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# The intracranial correlate of the 14&6/sec positive spikes normal scalp EEG variant

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HIGHLIGHTS

- The 14&6/sec positive spikes normal EEG variant is correlated exclusively with hippocampal activity.
- The variant is time-locked to high-amplitude spike bursts overlaid on low-amplitude slow waves.
- The scalp variant occurs ipsilateral to the hippocampus generating its intracranial correlate.

ABSTRACT

*Objective:* To investigate the intracranial correlate of the 14&6/sec positive spikes normal variant of scalp EEG.
*Methods:* Out of 35 adult refractory focal epilepsy patients who underwent intracranial electrode implantation with simultaneous scalp EEG electrodes, the 14&6/sec positive spikes variant was found in 4. We used three methods to identify and quantify intracranial correlates to the variant: visual inspection, time-referenced waveform averaging and 3D brain volume spectrum-based statistical parametric mapping (SPM).
*Results:* We discovered a novel and robust relationship between the scalp variant and an atypical hippocampal discharge. This intracranial correlate is an ipsilateral hippocampal burst of highly synchronized high-amplitude paroxysmal-like spikes of negative polarity, with a ramping up amplitude profile, which often ramps down and is accompanied by an underlying sequence of low-amplitude negative slow waves. The 14/sec positive spikes of the variant are time-locked to the negative peak of the hippocampal spikes, while the 6/sec positive spikes are time-locked to the negative spikes overlying the low-amplitude slow waves.
*Conclusions:* The 14&6/sec positive spikes variant correlates with bursts of negative polarity spikes in the ipsilateral hippocampus.
*Significance:* The identification of the hippocampal correlate of the 14&6/sec positive spikes variant fills a gap in our knowledge of normal intracranial variants. In clinical practice, this knowledge should reduce the chance that this electrophysiological signature is misinterpreted as epileptiform activity, which could inappropriately influence the interpretation of the intracranial study and subsequent surgical recommendation.


## 1. Introduction

The "14&6/sec positive spike" electroencephalographic (EEG) grapho-element is a well-known benign variant (Gibbs and Gibbs, 1952, 1963), also known as ctenoid due to its comb-like morphology (Lombroso et al., 1966; Schwartz and Lombroso,

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1968). It appears most often during sleep (Tsuzuki, 1967; Okuma et al., 1968) as a burst of arch-shaped waves of 13-17/sec and/or 5-7/sec frequency encompassing sharp components of positive polarity (IFSECN, 1974). The variant is distributed most often in a bilateral but asymmetrical and/or asynchronous fashion (Klass and Westmoreland, 1985), over the posterior cephalic regions (Niedermeyer, 2005), and is best highlighted by the use of an extended 10–20 inferior temporal electrode chain (Velizarova et al., 2011). The 14/sec and 6/sec components of the variant can appear independently, with the former being prominent in older children and adolescents and the latter prevailing in early childhood and adulthood (Gibbs and Gibbs, 1964; Eeg-Olofsson, 1971). The overall incidence of this normal EEG variant is age-dependent, ranging from 9% between 1 and 15 years (Gibbs et al., 1964; Eeg-Olofsson, 1971), peaking at 13% between 12 and 15 years (Metcalf, 1963; Weiner et al., 1966), and progressively decreasing in adulthood (Gibbs, 1990; Velizarova et al., 2011). Due to its spiky morphology, the 14&6/sec positive spike pattern has been investigated as a potential marker of brain pathology (DeLong et al., 1987). Excluding an increased appearance of the variant in hepatic coma patients (Poser and Ziegler, 1958; Silverman, 1964; Yamada et al., 1976, 1977) and encephalopathic children (Drury, 1989), the variant has been reliably shown to appear equally often in normal populations (Lombroso et al., 1966; Schwartz and Lombroso, 1968) and has been considered by some to be a sign of health rather than disease (Long and Johnson, 1968). The presence of the 14&6/sec positive spikes variant in coma and encephalopathies has been interpreted to be the result of its selective preservation and increased resistant character against structural and/or metabolic processes (Drury, 1989).

The intracranial source of the 14&6/sec positive spikes variant has not been investigated systematically so far, although the initial assumption was that of a subcortical origin (Gibbs and Gibbs, 1951). Niedermeyer first reported patterns similar to the scalp variant in depth electrode recordings from one patient, distributed across the thalamus and the basal ganglia, but also in the amygdala (Niedermeyer et al., 1967). Cortical recordings with subdural electrodes and simultaneous scalp EEG have identified similar patterns to the variant in the mesio-temporal structures (McLachlan and Luba, 2002). More recently, the use of stereotactic EEG (sEEG) with scalp EEG electrodes in a temporal lobe epilepsy (TLE) patient related the variant to the occurrence of hippocampal spike bursts (Jain et al., 2018). However, all previous reports were purely observational, lacking a robust correlation between the scalp variant and the intracranial electrocorticogram (ECoG), and most were based on intracranial recordings from epileptic tissue. Here, we link the 14 and 6/sec positive spikes normal EEG variant to its intracranial generator, based on recordings from non-epileptic tissue.

## 2. Methods

### 2.1. Patients

Patients for this study were derived from a pool of 173 consecutive adult patients diagnosed with refractory focal epilepsy who underwent stereotactic EEG (sEEG) or subdural electrode implantation at the University of Pittsburgh Medical Center (UMPC) from 2011 to 2018. Subdural or sEEG electrodes were implanted according to the epilepsy localization hypothesis determined during a multidisciplinary epilepsy patient management conference. Thirty-five (35) patients underwent simultaneous scalp-EEG recording during their intracranial investigation (see details in Abramovici et al., 2018; Antony et al., 2019). The entire scalp EEG recording over the full length of each patient's intracranial monitoring admission was reviewed retrospectively. The study was approved by the University of Pittsburgh Institutional Review Board (IRB).

### 2.2. Recording and analysis

Simultaneous scalp (EEG) and intracranial (ECoG) signals were recorded at 1 kHz, by a 128-channel Xltek digital system (Natus Medical Incorporated, Pleasanton, CA). Two board-certified epileptologists (A.U. and N.Z.) and an epilepsy surgery neurophysiologist (V.K.) reviewed the scalp EEG of the selected patients. An event marker was placed at the first spiky positive peak of each scalp EEG variant, in all seizure-free recordings of each patient before the first clinical seizure manifestation of their intracranial monitoring. Extracted scalp EEG segments were separated into 14/sec only and 14&6/sec positive spikes groups, where applicable. Each group was in turn waveform averaged at an interval of 4 sec (±2 sec from the reference event marker) after computational correction of the marker position to the closest most positive point of the peak (top of the peak) in MatLab (The Mathworks, Natic, MA, USA). Fast Fourier Transform (FFT)-based event-related time–frequency analysis was performed for each selected event within a time-window centered (time = 0) at the event marker, 0.05–25 Hz for the scalp EEG and 0.05–80 Hz for the intracranial ECoG, at a step of 0.05 Hz.

Spectrum-based statistical parametric mapping (SPM) analysis of the intracranial ECoG data in the 3D MRI space was performed in Brainstorm (Tadel et al., 2011) for the 14/sec positive spikes samples of Patient 3, implanted bilaterally and symmetrically in the frontal and temporal lobes (Supplementary Fig. 1c). The pre-operative MRI was co-registered with the post-implantation MRI using SPM12 (Wellcome Trust Centre for Imaging Neuroscience; http://www.fil.ion.ucl.ac.uk/spm), and in turn electrodes were localized in 3D MRI space according to the surgical plan and notes in Brainstorm. All raw ECoG channels were linked to the respective sEEG electrode contacts through the respective 3D MRI space coordinates. The statistical significance of the appearance of the alpha-beta (10–30 Hz) and separately gamma (30–80 Hz) ranges was evaluated in the 0.5 sec interval after the event marker for the 14/sec positive spikes group of samples (methodology as described by David et al., 2011). ECoG segments of 6 sec (−4 sec, +2 sec from the event markers) were used, and one-second pre-marker intervals of background ECoG activity from each were used as baselines for statistical comparison. Z-scores to quantify the probability of power increase within 1 s interval after the markers versus baseline power for each spectral band were derived by means of FDR (False Discovery Rate) control for multiple comparisons.

## 3. Results

The review of patient scalp EEG records revealed 14&6/sec positive spikes variant in 4 of 35 patients (11.4%). All four patients had undergone sEEG implantation, cumulatively covering the frontal, parietal, temporal and insular lobes. Patient 1 had parietal lobe epilepsy, while the rest had frontal lobe epilepsy; none of the 4 patients identified with the scalp variant suffered from TLE (see Table 1 for detailed clinical and surgical data). Mesial temporal lobe coverage included a single sEEG electrode targeting the head/anterior body region of the hippocampus (Supplementary Fig. 1).

Visual inspection of all the simultaneous EEG/ECoG records showed that the 14&6/sec positive spikes normal scalp EEG variant coincided with the appearance of an atypical hippocampal waveform in 3 patients whose sEEG implantation coverage included the mesial temporal structures (Figs. 1, 2). No intracranial activity coinciding with the 14&6/sec positive spikes variant was observed in the recordings of Patient 4 who had bilateral frontal sEEG coverage without concurrent coverage below the Sylvian fissure. The 14/sec counterpart of the variant was observed to appear at the

**Table 1**
Clinical and surgical data of sEEG-implanted patients that manifested the 14&6/sec positive spikes variant on their scalp EEG.

| | Age | Gender | Duration of epilepsy (years) | AEDs at the time of surgery | Scalp EEG electrodes[a] | 14&6/sec positive spikes groups and samples (n) | Intracranial electrode implantation plan (Entry/ Target) | ECoG-based seizure onset | Surgical outcome (Engel Classification – Postop interval) | Localization of 14&6/ sec positive spikes correlate |
|---|---|---|---|---|---|---|---|---|---|---|
| Patient 1 | 26 | F | 10 | CBZ, LCM, LTG, LVT | Obligate International 10–20 System electrodes + F10, T10, P9, P10 | 14/sec only (n = 149) 14&6/sec (n = 127) | Unilateral Left sEEG Coverage 1. Middle Fontal Gyrus/ Anterior Cingulate 2. Middle Frontal Gyrus/ Middle Cingulate 3. Middle Frontal Gyrus/ Superior Frontal Gyrus Mesial 4. Middle Frontal Gyrus/ Supplementary Motor Area 5.Precentral Gyrus Lateral/Precentral Gyrus Mesial 6. Middle Temporal Gyrus/Hippocampus 7.Inferior Parietal Lobule/ Precuneus Superior 8. Inferior Parietal Lobule/Precuneus Inferior | Precuneus | I – 4.8 years | Left Hippocampus |
| Patient 2 | 52 | M | 51 | CBZ, CLZ, LVT | Obligate International 10–20 System electrodes | 14/sec only (n = 118) | Unilateral Right sEEG Coverage 1. Inferior Frontal Gyrus/ Orbitofrontal Mesial 2. Middle Temporal Gyrus/Amygdala 3. Middle Temporal Gyrus/Hippocampus 4. Middle Temporal Gyrus/Basal Temporal Mesial 5. Superior Temporal Gyrus/Posterior Insula | Inferior Frontal | III – 3.8 years | Right Hippocampus |
| Patient 3 | 36 | M | 22 | OXC, VA, ZNS | Obligate International 10–20 System electrodes + F9, F10, T9, T10, P9, P10 | 14/sec only (Right n = 186, Left n = 61) | Bilateral Symmetric sEEG Coverage 1.Inferior Frontal Gyrus/ Orbitofrontal Mesial 2.Superior Frontal Gyrus/ Mesial Frontal 3. Middle Frontal Gyrus/ Anterior Cingulate 4. Middle Frontal Gyrus/ Middle Cingulate 5.Inferior Frontal Gyrus/ Anterior Insula 6. Precentral Gyrus/ Middle Insula 7. Postcentral Gyrus/ Posterior Insula 8. Middle Temporal Gyrus/Amygdala 9. Middle Temporal Gyrus/Hippocampus | Left Superior Frontal | I –13 months | Left & Right Hippocampus |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Patient 4 | 25 | M | 14 | CLZ, CZM, LCM, LVT, ZNS | Obligate International 10–20 System electrodes | 14/sec only (n = 102) | Bilateral sEEG Coverage<br>Left:<br>1.PrefrontalLateral/Basal Frontal Mesial<br>2. Inferior Frontal Gyrus/ Anterior Cingulate<br>3. Middle Frontal Gyrus/ Anterior Cingulate<br>4. Inferior Frontal Gyrus/ Anterior Insula<br>5.Superior Frontal Gyrus/ Supplementary Motor Area<br>6. Middle Frontal Gyrus/ Middle Cingulate<br>7. Middle Frontal Gyrus/ Supplementary Motor Area<br>8. Precentral Gyrus/ Paracentral Lobule<br>Right:<br>1. Inferior Frontal Gyrus/ Anterior Cingulate<br>2. Middle Frontal Gyrus/ Middle Cingulate<br>3. Middle Frontal Gyrus/ Supplementary Motor Area<br>4. Precentral Gyrus/ Paracentral Lobule | Left Superior & Middle Frontal | III –16 months | n/a |

[a] Most scalp electrode positions were approximations of the International10-20 System, as a result of concurrent sEEG.

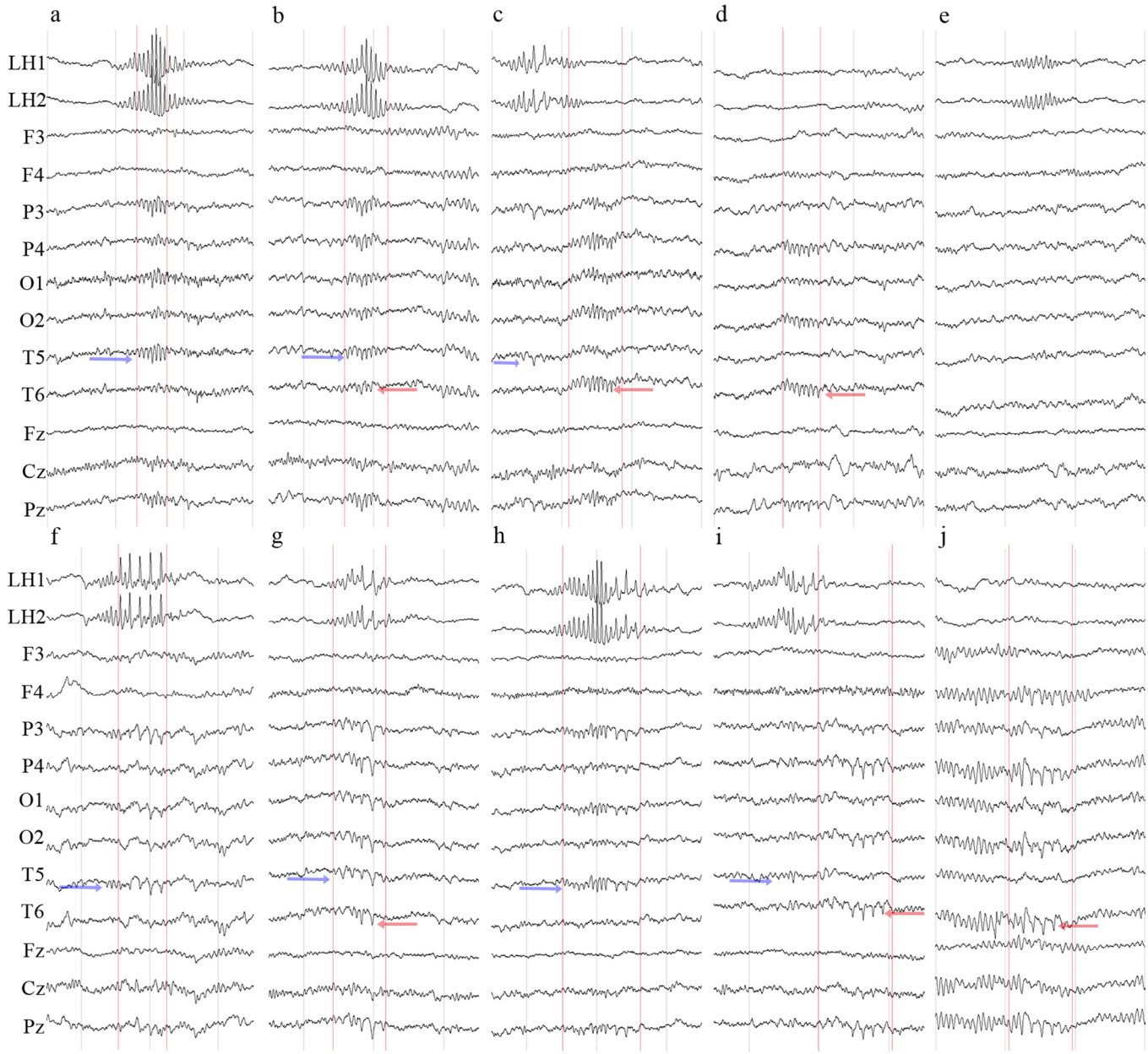


**Fig. 1.** Raw concurrent scalp EEG and intracranial ECoG samples from unilaterally implanted Patient 1, highlighting the manifestation of the 14&6/sec positive spikes variant and its intracranial correlate. (a) The 14/sec only positive spikes version of the variant appears on the left posterior (P3/O1/T5 max) scalp electrodes, at the same time with highly synchronized paroxysmal-like bursts of predominantly negative polarity spikes, following a profile of progressively increasing and then decreasing amplitude, originating in the ipsilateral hippocampus. Bilateral manifestations of the 14&6/sec positive spikes variant were also linked to the appearance of the hippocampal correlate waveform (b). This correlate waveform does not coincide with 14&6/sec positive spikes appearing in contralateral scalp electrodes (P4/O2/T6 max) (c, d), and also is not a hippocampal spindle judging by its waveform morphology and by the fact that hippocampal spindles are not coinciding with the scalp variant (e). (g) The 6/sec part of the 14&6/sec positive spikes variant coincides with the appearance of a low amplitude negative phase slow wave sequence among the negative spike burst. The 6/sec part of the variant can appear either mixed with the 14/sec component – often with bilateral distribution – (h) or in series (i). Similar to the 14/sec only version of the variant, the hippocampal correlate does not coincide with contralateral manifestations of the 14&6/sec positive spikes variant (j–l). Vertical red lines denote the duration of the scalp variant; right- and left-sided manifestations of the variant are denoted by red and blue arrows respectively. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

same time as a distinct hippocampal waveform comprised of highly synchronized paroxysmal-like bursts of predominantly negative polarity spikes, with a ramping up – often ramping down – amplitude profile (Figs. 1a, 2a). For lack of a widely accepted term to describe this discharge pattern, we will refer to the observed hippocampal activity as the "intracranial correlate" or "hippocampal correlate" of the 14&6/sec positive spikes scalp EEG variant. The intracranial correlate of the 14&6/sec positive spikes variant is not a hippocampal spindle, as the morphology of this correlate waveform is markedly different than that of hippocampal spindles, and hippocampal spindles did not coincide with the scalp variant (Figs. 1b, 2b). The intracranial hippocampal correlate coincides with ipsilateral manifestations of the variant (mean coincidence percentage: 100%, Figs. 1a, 2a) but does not coincide with contralateral manifestations (mean coincidence percentage: 37.1%, Figs. 1c, d, f, 2c, d, f). Bilateral manifestations of the 14&6/sec

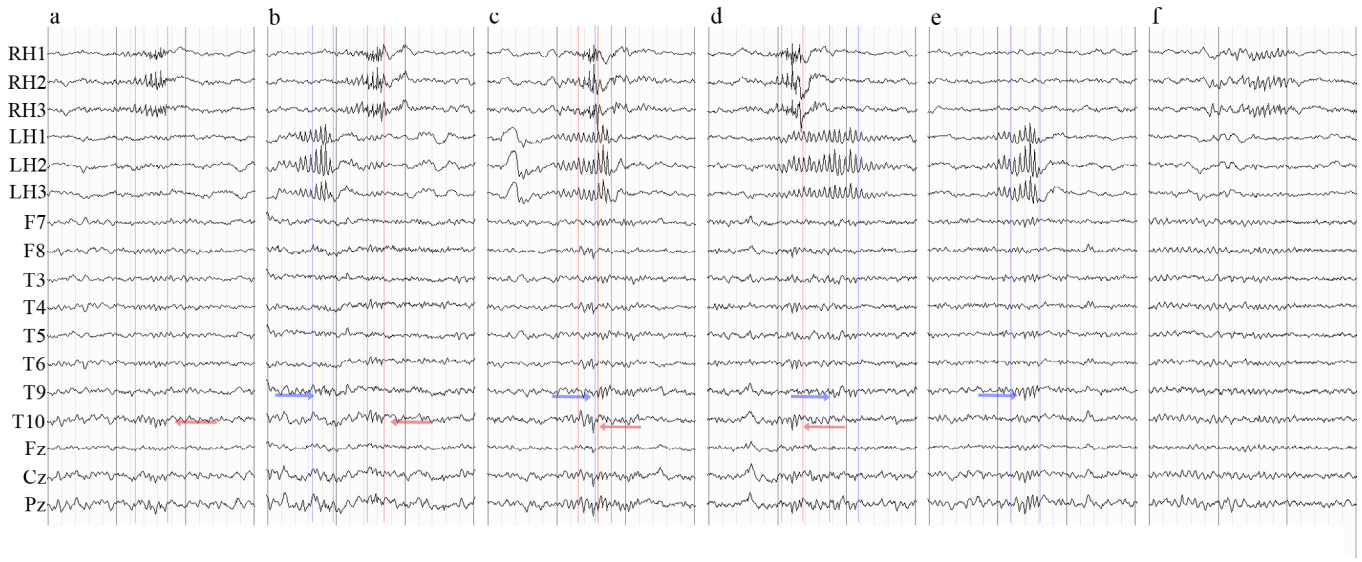


**Fig. 2.** Raw concurrent scalp EEG and intracranial ECoG samples from bilaterally implanted Patient 3, highlighting the manifestation of the 14&6/sec positive spikes variant and its intracranial correlate. (a) The 14/sec only positive spikes version of the variant appears on the right posterior (T4/T6/T10 max) scalp electrodes, at the same time with ipsilateral hippocampal activity similar to that of Patient 1 (Fig. 1) accompanied by high frequency content. Unilateral and bilateral manifestations of the 14&6/sec positive spikes variant were linked to the appearance of the hippocampal correlate waveform in the ipsilateral hippocampus (b–d). (e) Manifestations of the correlate waveform on the left hippocampus does not coincide with the appearance of right-sided 14&6/sec positive spikes, but with low-amplitude brief left-sided 14&6/sec positive spikes (T3/T5/T9 max, also see b, c, d); the weak amplitude of the variant on the left hemisphere is a result of necessary scalp electrode placement deviations from the International 10–20 System due to concurrent sEEG. As in Patient 1, this correlate waveform is not a hippocampal spindle (f). Vertical red lines denote the duration of the scalp variant; right- and left-sided manifestations of the variant are denoted by red and blue arrows respectively. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

positive spikes variant occur when the intracranial correlate is present simultaneously in both hippocampi (Fig. 2e).

The 6/sec counterpart of the 14&6/sec positive spikes variant was observed only in Patient 1 and coincided with a variation of the hippocampal discharge correlate, characterized by the additional appearance of a sequence of low amplitude negative slow waves underlying the negative spike burst (Fig. 1g). Variations in the manifestation of the 6/sec counterpart of the variant, appearing either mixed with the 14/sec component (Fig. 1h) or in series (Fig. 1i), were accompanied by respective variations in the slow waves' distribution among the spike bursts. Similar to the 14/sec counterpart of the variant, the hippocampal correlate complex does not coincide with contralateral manifestations of the 6/sec counterpart (Fig. 1j–l).

Temporal correlation of averaged waveforms by means of event-marker referencing from the first positive spike of the EEG variant resulted in concurrent non-baseline (noise) waveforms only in the deep hippocampal contacts of the three patients, in accordance with the visual evaluation (Figs. 3, 4, and Supplementary Fig. 2). Respective averaged waveforms from the rest of the implanted foci (see Table 1) resulted in baselines, without any discernable time-locked morphologic feature. The waveforms temporally correlated to the scalp 14&6/sec variant were the ones visually observed to coincide. More specifically, waveform temporal correlation verified the visually appreciated hippocampal correlate of the 14&6/sec positive spikes variant and further showed that: (a) the 14&6/sec positive spikes on the scalp are strongly time-locked (high synchrony) to the negatives spikes of the ipsilateral hippocampus (mean time interval from closest intracranial peak is −4.27 ms SD = 3.68 ms; −2.93 ms SD = 3.68 ms for Patient 1, −4.88 ms SD = 3.30 ms for Patient 2, −5.02 ms SD = 4.06 ms for Patient 3/right and −4.10 ms SD = 3.43 ms for Patient 3/left; Figs. 3c, 4c, 5c and Supplementary Fig. 2c), (b) the 14&6/sec scalp positive spikes are not time-locked (low synchrony) to the contralateral hippocampal activity (mean time interval from closest intracranial peak is 6.59 ms SD = 30.37 ms: 7.81 ms SD = 29.56 ms for Patient 1, 8.78 ms SD = 31.23 ms for Patient 2, 3.17 ms SD = 30.32 ms for Patient 3/right, and 4.29 ms SD = 31.24 ms for Patient 3/left; Figs. 3e, g, m, 4e, and Supplementary Fig. 2g), (c) the 14/sec constituent of the variant is time-locked to the high-amplitude (200–1000 μV) negative spikes of the hippocampal correlate (Figs. 3c, 4c, and Supplementary Fig. 2c), and (d) the 6/sec counterpart is time-locked to the high-amplitude negative spikes riding the underlying low-amplitude slow waves (mean time interval from closest intracranial peak is −3.90 ms SD = 3.06 ms ipsilateral, and 1.95 ms SD = 43.32 ms contralateral in Patient 1; Fig. 3k, o). Time-frequency analysis showed that the respective hippocampal correlates of the 14/sec and 6/sec counterparts have spectral profiles that are rich in frequencies higher that 14 Hz, owing to the sharpness of the negative spike bursts that comprise them (Fig. 3d, l, 4d, and Supplementary Fig. 2d); a spectral profile that appears reduced in power and higher frequency content in the contralateral hippocampus (Fig. 3h, p, 4f, and Supplementary Fig. 2h).

Temporal correlation between the right and left hippocampal correlates from the bilaterally implanted Patient 3 showed only a small but significant percentage of them appearing concurrently bilaterally (29 occurrences, 15.59% of right hippocampal correlates of the variant, 47.54% of left hippocampal correlates of the variant, Fig. 6a). Most right-sided manifestations did not coincide with left-sided manifestations of the hippocampal correlate, although a subgroup coincided with brief left hippocampal spindles (84.41%, Fig. 6b). Almost half of the left hippocampal correlates did not coincide with respective right-sided manifestations (52.46%, Fig. 6c).

3D brain volume SPM was applied only in the recordings of Patient 3, who was bilaterally and symmetrically sEEG-implanted in frontal and temporal regions (Supplementary Fig. 1c). The appearance of the 14&6/sec positive spikes variant was correlated exclusively with hippocampal activity (Figs. 4g, h, 5g, h); no other implanted parts of the brain resulted in statistically significant

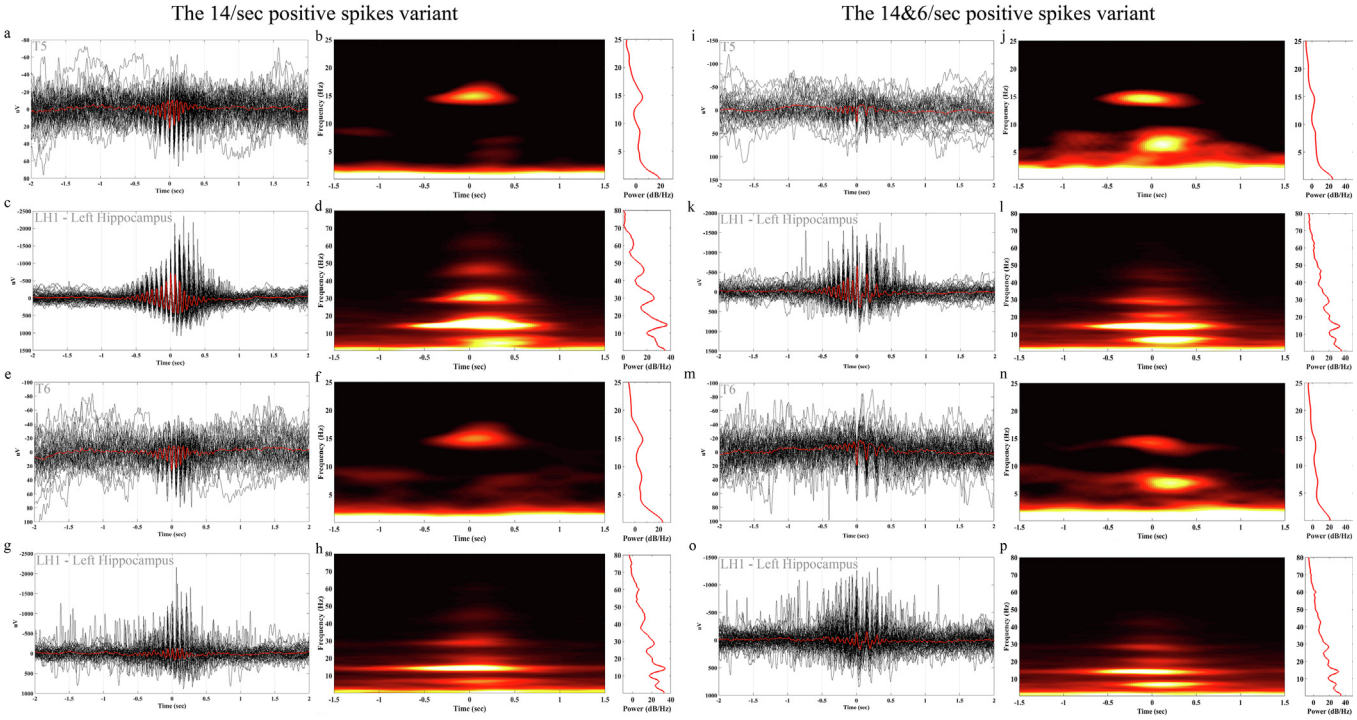


**Fig. 3.** Intracranial correlation study results for the 14/sec only and 14&6/sec positive spikes variant of Patient 1. (a) Waveform average of the 14/sec only positive spikes variant (red trace) over the T5 scalp electrode, time-referenced (t = 0) by event-markers at the first positive spike peak, with the individual samples superimposed (black traces, n = 82), and (b) the respective 3D time-frequency and 2D power-frequency plots. (c) Waveform average of the hippocampal correlate, time-referenced to the same scalp 14&6/sec positive spikes event-marker on the T5 electrode, with all individual samples superimposed. Note the highly systematic time-lock in both the average and single sample waveforms, reflecting a high degree of temporal synchronization between the hippocampal waveform and the scalp EEG variant. (d) 3D time-frequency and 2D power-frequency plots for the hippocampal correlate of the scalp 14&6/sec positive spikes variant. Note the rich in higher than 14 Hz frequency spectral profile of the hippocampal correlate of the variant, owing to the consistent sharpness of its spiky constituents. (e–h) Respective data derived from the contralateral electrode (T6, n = 67), showing poor temporal relation with the left hippocampal waveforms and suggesting incidental coincidence (depicted in Fig. 1e). (i) Waveform average of the 14&6/sec positive spikes variant recorded at T5, with the individual samples superimposed (n = 55), and the respective 3D time-frequency and 2D power-frequency plots (j). (k) Waveform average of the hippocampal correlate with all individual samples superimposed. Note both the highly systematic time-lock signifying high temporal synchronization between the hippocampal waveform and the scalp positive spikes, as well as the time-lock of the 6/sec constituent with the emergence of the low-amplitude slow wave underlying the negative spike burst. (l) 3D time-frequency and 2D power-frequency plots for the hippocampal correlate of the scalp 14&6/sec positive spikes variant. (m–p) Respective data derived from the contralateral electrode (T6, n = 72), showing poor temporal relation with the left hippocampal waveforms and suggesting incidental coincidence (Fig. 1h). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

correlated activity. In the alpha/beta 10–30 Hz range, the right hemispheric manifestation of the variant was correlated to a higher degree with ipsilateral hippocampal activity (right hippocampus $Z_R = 24.413$) compared to the contralateral one (left hippocampus $Z_L = 11.165$) (Fig. 4g); the left manifestation of the variant was correlated to a higher degree with activity in the left hippocampus ($Z_L = 25.159$) compared to the right ($Z_R = 12.588$) (Fig. 5g). In the gamma 30–80 Hz range, the right-hemispheric variant was almost exclusively correlated with ipsilateral hippocampal activity ($Z_R = 7.909$; $Z_L = 3.270$) (Fig. 4h); the left hemispheric manifestation of the 14&6/sec positive spikes variant was also exclusively correlated with ipsilateral hippocampal gamma activity ($Z_L = 7.551$; $Z_R = 0.00$) (Fig. 5h).

## 4. Discussion

This intracranial correlation study resulted in the identification of the source of the 14&6/sec positive spikes normal scalp EEG variant. Using three methods (visual evaluation, waveform averaging, and SPM) we documented a strong relationship between the scalp variant and an atypical hippocampal discharge. The intracranial correlate of the 14&6/sec positive spikes variant is an ipsilateral hippocampal burst of highly synchronized paroxysmal-like negative polarity spikes, with a ramping up – often ramping down – amplitude profile, often accompanied by an underlying sequence of low amplitude negative slow waves. The 14/sec positive spikes of the variant are time-locked to the negative peak of the hippocampal spikes, while the 6/sec positive spikes are time-locked to the negative spikes riding the low-amplitude slow waves. The 14&6/sec variant, although predominantly correlated with the manifestation of the ipsilateral atypical hippocampal discharge, is sometimes accompanied by a concurrent contralateral correlate due to intra-hippocampal synchronization. The rate of inter-hippocampal synchronization producing simultaneous bilateral hippocampal correlate discharges is low but appreciable. The quantitatively highlighted hippocampal correlate of the 14&6/sec positive spikes variant is expected to become an important asset in future evaluations of intracranial activity in the human hippocampus.

An important advantage of this study derives from the confidence that the hippocampi recorded were not exhibiting epileptic behavior. Although the mesial temporal structures were initially targeted in our surgical planning because phase I data showed extension of the extratemporal interictal discharges to the anterior temporal electrodes (T1/T3 and T2/T4, more than T5/T6), phase II did not confirm their involvement in seizure generation. More specifically: (a) no interictal activity was recorded from the hippocampal contacts, in the presence of abundant extratemporal interictal activity, and (b) the hippocampus was not involved in the electrographic onset of the recorded extratemporal clinical seizures. More importantly, and for the purposes of this study, no interictal activity was observed or found by analysis to be

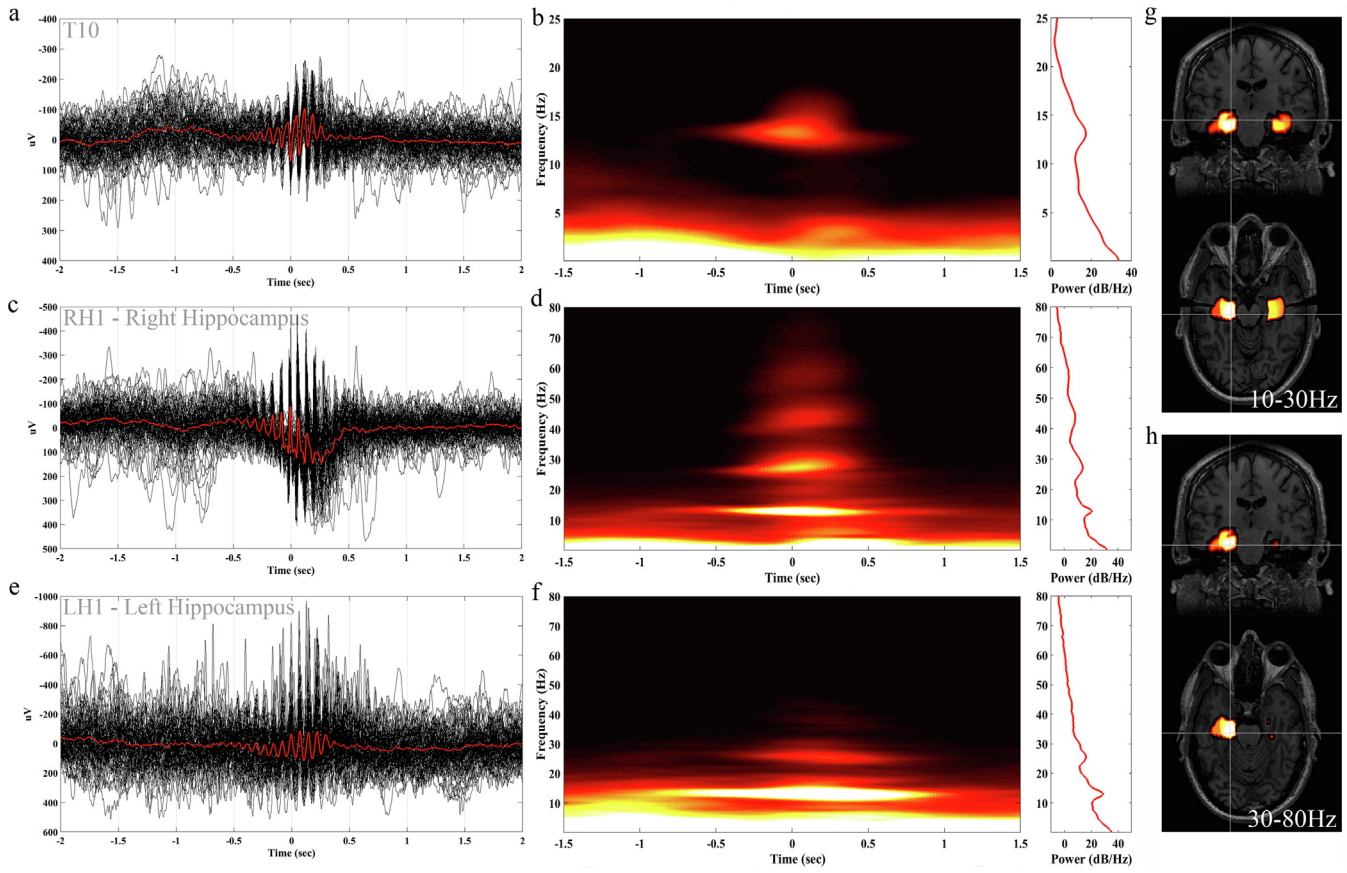


**Fig. 4.** Intracranial correlation study results for the right hemispheric 14/sec only positive spikes variant of Patient 3. (a) Waveform average of the 14/sec only positive spikes variant recorded at the T10 scalp electrode (red trace), with the individual samples superimposed (black traces, n = 186), and (b) the respective 3D time-frequency and 2D power-frequency plots. (c) Waveform average of the hippocampal correlate (red), time-referenced to the same scalp 14/sec only positive spikes event-marker on T10, with all individual samples superimposed (black). As shown for Patient 1 in Fig. 1, a highly systematic time-lock exists in both the average and single sample waveforms, reflecting a high degree of temporal synchronization between the hippocampal waveform and the scalp EEG variant. (d) 3D time-frequency and 2D power-frequency plots for the hippocampal correlate of the scalp 14/sec only positive spikes variant. (e, f) Similar to the c, d pair, showing the respective waveforms and spectral content from the left hippocampus. The spectral profile of the hippocampal correlate is rich in higher than 14 Hz frequencies, as a result of the systematic sharpness of its spikes. (g, h) 3D volume statistical parametric mapping for the intracranial correlate of the right lateralized 14/sec only positive spikes variant, appearing highly correlated to the emergence of alpha/beta (10–30 Hz) and gamma (30–80 Hz) frequencies in the right hippocampus ($Z_R$ = 24.413 and 7.909, respectively), and less correlated to left hippocampal activity ($Z_L$ = 11.165 and 3.270, respectively). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

correlated with the manifestation of the scalp 14&6/sec variant. Two of our patients (Patients 1 and 3) achieved excellent seizure control after surgical treatment of extratemporal regions. And although Patients 2 and 4 achieved a lower level of post-operative seizure control level, they fall within the documented outcome expectations of non-lesional frontal lobe epilepsy.

Attempts to identify the intracranial correlate of the 14&6/sec positive spikes variant have been few, seeking to identify intracranial discharges that share common spectral features with the scalp variant, without the possibility of temporal correlation due to the lack of concurrent scalp EEG (Niedermeyer et al., 1967; McLachlan and Luba, 2002). Moreover, intracranial recording coverage was limited to the cortical surfaces, as recording were made by subdural electrodes. Most recently, a brief observational report from an sEEG-implanted TLE patient described hippocampal discharges, that visually resemble the ones we identified, coinciding with manifestations of the 14&6/sec positive spikes variant (Jain et al., 2018); however, sEEG recordings were derived from epileptic tissue, which makes the distinction between epileptic and non-epileptic spike discharges difficult. Our study surpassed the limitations of previous investigations, as we used recordings from non-TLE patients, extensively implanted with sEEG electrodes across the brain, and used three methods to establish a robust relationship between the scalp EEG 14&6/sec positive spikes variant and its hippocampal correlate.

The hippocampal correlate of the 14&6/sec positive spikes variant is a discharge distinct from the well-documented hippocampal spindles (Brazier, 1970; Montplaisir et al., 1981; Nakabayashi et al., 2001; Andrillon et al., 2011; Sarasso et al., 2014; Frauscher et al., 2015). There are three features that support this distinction: (a) The markedly asymmetric polarity of the 14&6/sec positive spikes correlate vs the traditionally symmetric polarity of the spindles, forming their characteristic waxing and waning shape (Andrillon et al., 2011); the amplitude of the negative portion of the hippocampal correlate is 2–4 times higher than that of the positive part (Figs. 1, 2). (b) The spiky morphology of the hippocampal correlate's negative portion vs the rather smooth biphasic peaks of the hippocampal spindles (Brazier, 1970; Andrillon et al., 2011; Frauscher et al., 2015). (c) The strong association (time-locking) of the hippocampal correlate with the 14&6/sec positive spikes variant vs the rather independent nature of the hippocampal spindles that may or may not coincide with scalp sleep spindles (Nakabayashi et al., 2001).

Although the spiky parts of 14&6/sec scalp variant are predominantly positive in polarity, the sharp elements of its intracranial correlate are negative. This contrast may be explained by the

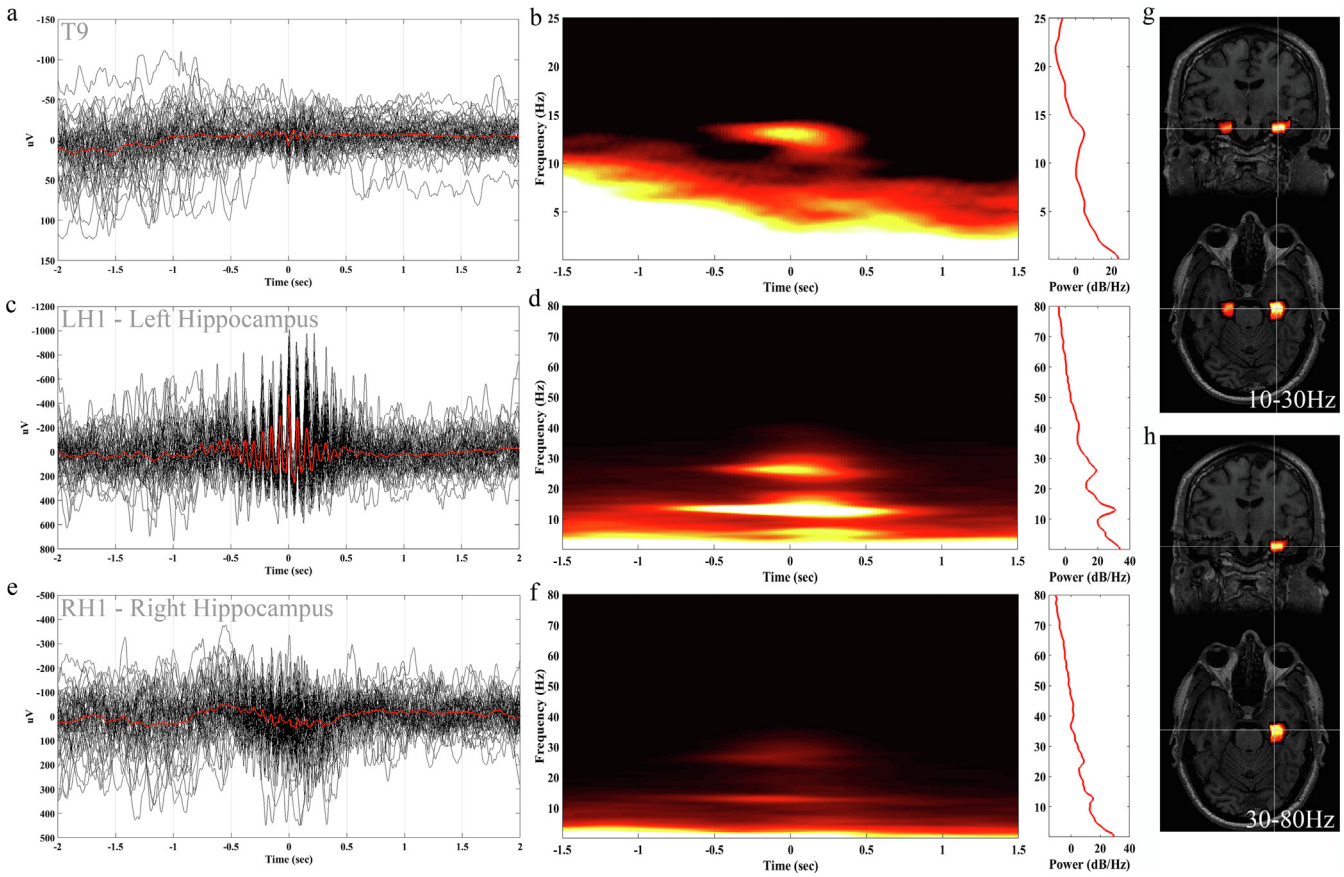


**Fig. 5.** Intracranial correlation study results for the left hemispheric 14/sec only positive spikes variant of Patient 3. (a) Waveform average of the 14/sec only positive spikes variant recorded at the T9 scalp electrode (red trace), with the individual samples superimposed (black traces, n = 60), and (b) the respective 3D time-frequency and 2D power-frequency plots. (c) Waveform average of the hippocampal correlate (red), time-referenced to the same scalp 14/sec only positive spikes event-marker on T9, with all individual samples superimposed (black). (d) 3D time-frequency and 2D power-frequency plots for the hippocampal correlate of the left-sided scalp 14/sec only positive spikes variant. (e, f) Similar to the c, d pair, showing the respective waveforms and spectral content from the right hippocampus. (g, h) The left hemispheric variant appears highly correlated to the emergence of alpha/beta (10–30 Hz) and gamma (30–80 Hz) frequencies in the ipsilateral hippocampus ($Z_L$ = 25.159 and 7.551, respectively), and less correlated to contralateral hippocampal activity ($Z_R$ = 12.588 and 0.00, respectively). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

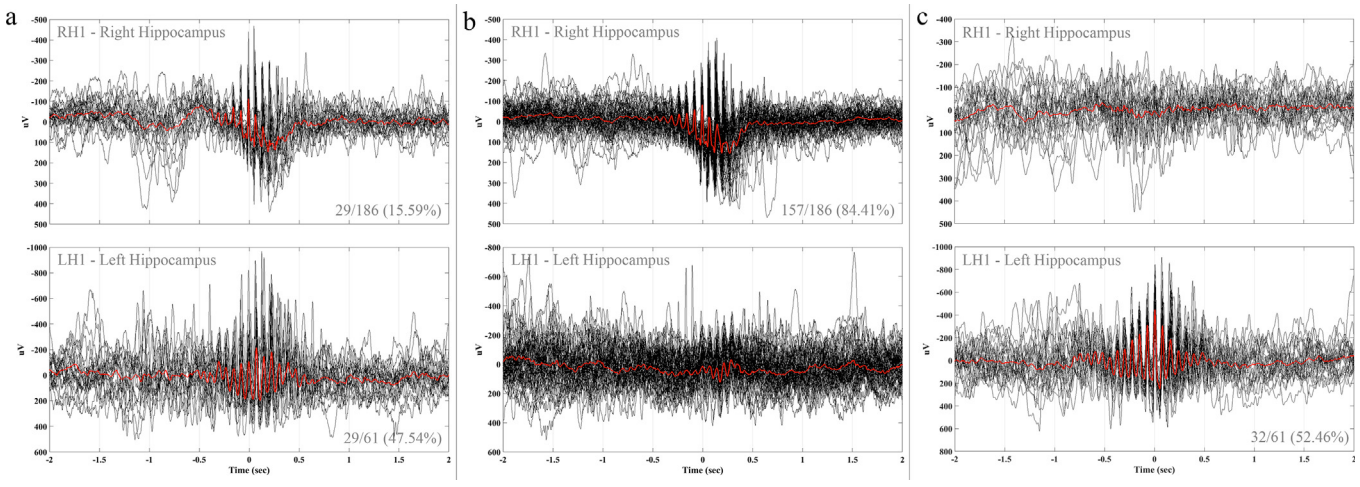


**Fig. 6.** Intracranial temporal correlation results for the hippocampal correlate, derived from the data of Patient 3 bilaterally implanted in the hippocampus. (a) A small but significant percentage of the hippocampal correlate appeared concurrently bilaterally. (b) The majority of right-sided manifestations were not accompanied by left-sided manifestations. In a subgroup of them, brief hippocampal spindles were developed on the left hippocampus after the appearance of the right-sided hippocampal correlate, giving rise to a minor oscillation in the average waveform. (c) Almost half of the left-sided hippocampal correlate manifestations were not accompanied by right-sided correlates of the variant.

anatomical orientation of the hippocampus, and its longitudinal connections. The hippocampus is mostly known for its trisynaptic circuit in the transverse direction (Andersen, 1975), however connections between the anterior and posterior hippocampus have also been described through the CA3 network (Lorente de Nò, 1934; Miles et al., 1988; Li et al., 1994). It is possible that the hippocampal correlate of the 14&6/sec positive spikes variant is a discharge specific to the longitudinal networks of the hippocampus, spreading along the anterior-posterior hippocampal axis and appearing with inversed polarity over the posterior scalp EEG electrodes. This hypothesis can also explain the fact that the use of an additional lower temporal electrode line increases the identification yield for the 14&6/sec positive spikes variant (Velizarova et al., 2011).

Another observation regards the extracranial distribution of the 14&6/sec positive spikes variant. Our study shows that the hippocampal correlate of the variant most often lateralizes to either hippocampus (Figs. 2a, 2, 6b and c) or appears bilaterally in an asynchronous manner (Fig. 2c and d), and seldom appears in bilateral synchrony (Figs. 2c and 6a). These findings suggest that the intracranial behavior of the variant's hippocampal correlate resembles the extracranial behavior of the variant, as it has been well known that the variant has an asymmetrical and/or asynchronous posterior EEG distribution (Klass and Westmoreland, 1985; Niedermeyer, 2005).

Our study was limited in terms of subject number, mainly by the inherent low incidence of the 14&6/sec positive spikes variant. In addition, extended sEEG coverage often did not allow additional scalp EEG electrodes to be placed using our system, given its amplifier channel limitations. Moreover, although the sEEG implantation of our patients cumulatively covered most of the brain, thereby reliably localizing the appearance of the correlate discharge in the hippocampus, none of these patients had sEEG electrodes in the posterior body and tail of the hippocampus, as well as the occipital lobe. Given that the temporal lobe receives projections from occipital areas (Catani et al, 2003), this study cannot currently rule out the involvement of the occipital lobe in the generation of the 14&6/sec positive spikes variant. However, the previous study of McLachlan and Luba (2002) that included occipital coverage did not report findings from that area. The resolution of a potential more posterior contribution was further limited by the fact that all our patients happen to have had only one electrode in the head of the hippocampus, thereby depriving us from monitoring the posterior body and the tail. Our typical scheme of 2–3 orthogonal electrodes covering the mesial temporal lobe along its longitudinal axis would have allowed us to study the intracranial correlate of the 14&6/sec positive spikes variant in relation to anterior-posterior location within the hippocampus, and potentially to estimate the involvement of posterior brain areas. However, our patients were not investigated for TLE but for suspected involvement of the mesial temporal structures during the progression of their seizures, which resulted in advantageous recordings from non-epileptic hippocampal tissue.

## 5. Conclusions

Using visual inspection, time-referenced waveform averaging, and SPM, we quantitatively identified the intracranial correlate of the 14&6/sec positive spikes scalp EEG normal variant. The occurrence of the 14&6/sec positive spikes variant coincides with hippocampal activity exclusively. We found that the scalp variant is time-locked to a specific hippocampal waveform, characterized by bursts of high-amplitude negative spikes, with a ramping up – often ramping down – amplitude profile, often overlaid on low-amplitude slow waves. More specifically, the 14/sec counterpart of the variant is time-locked to the negative peak of the hippocampal spikes, while the 6/sec counterpart is time-locked to the negative spikes riding the low-amplitude slow waves. The 14&6/sec positive spikes variant manifests ipsilateral to the hippocampus that generates its intracranial correlate.

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## Declaration of Competing Interest

The authors have no conflict of interest to declare.

## Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.clinph.2019.05.024.

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