Original article

Epileptic Disord 2010; 12 (4): 292-302

Accentuated cortico-cortical evoked potentials in neocortical epilepsy in areas of ictal onset

Masaki Iwasaki[1,4], Rei Enatsu[1], Riki Matsumoto[1,3], Eric Novak[2], Baburaj Thankappen[1], Zhe Piao[1], Tim O'Connor R[1], Karl Horning[1], William Bingaman[1], Dileep Nair[1,2]

[1] Epilepsy Center, Neurological Institute, Cleveland Clinic
[2] Biostatistics, Department of Qualitative Health Sciences, Cleveland Clinic, USA
[3] Department of Neurology, Kyoto University Graduate School of Medicine
[4] Department of Neurosurgery, Tohoku University Graduate School of Medicine, Japan

Received November 11, 2009; Accepted August 20, 2010

ABSTRACT – *Objective.* To determine whether patients with neocortical epilepsy show evidence for increased excitability measured by cortico-cortical evoked potentials (CCEPs) in ictal-onset regions. *Methods.* In patients undergoing intracranial recordings with subdural electrodes for epilepsy surgery, we measured amplitudes, latencies, and stimulus thresholds of CCEPs near ictal onset zones (iCCEPs), and compared with adjacent neocortex not associated with ictal EEG (nCCEP). CCEP amplitude and latency measurements were made with each stimulation site, using graded stimulation intensities. *Results.* Ten patients were included in this study. CCEPs were recorded in eight of 10 patients. The first negative (N1) iCCEP amplitude was higher than that of nCCEP in seven of the eight patients. In the group analysis, this difference was statistically significant. In three of these patients, the difference was individually significant. In one patient, the amplitude was higher in nCCEP than iCCEP and the area selected as nCCEP was within primary eloquent cortex. There was no significant difference seen in latency changes or stimulus threshold. *Conclusions.* Accentuated CCEP amplitudes near ictal onset zones could reflect an increased excitability of the cortex associated with the epileptogenic zone in some patients with neocortical epilepsy. The response of the neocortex to low-frequency stimulation may vary depending on the presence or absence of intrinsic epileptogenicity.

Key words: cortical stimulation, epilepsy, evoked potentials, ictal-onset zone, subdural electrode, low frequency stimulation

The purpose of a presurgical evaluation in patients with medically intractable focal epilepsy is to estimate the location and the extent of the epileptogenic zone. The location of the epileptogenic zone is not directly measurable; rather it is assumed by the concordance of data which resolve the location of the irritative zone, ictal onset zone, functional deficit zone, and epileptic lesion (Carreno and Lüders, 2001). When there is a lack of concordance of data, or if functional and epileptic regions are in close proximity, further evaluation using intracranial EEG recordings may be warranted. The placement of subdural and depth electrodes is based on the hypothesis generated by the noninvasive phase of the testing. Invasive electrodes only identify the reflection of the

Correspondence:
D. Nair
Epilepsy Center, Desk S51,
Cleveland Clinic, 9500 Euclid Avenue,
Cleveland, OH 44195, USA
<nairdccf.org>

doi: 10.1684/epd.2010.0334

spread of ictal activity if the true ictal onset is either deep in the brain or at the margins of the subdural or depth electrode array (Nair *et al.*, 2008). For purposes of this study, regions identified by invasive recordings as ictal onset have been described as the "ictal onset zone," although the precise area of the true ictal onset cannot always be delineated by this methodology.

Cortical excitability is closely related to the pathophysiology of epilepsy. Increased excitability of cortex in epileptic patients can be associated with increased amplitudes of evoked potentials. Giant somatosensory evoked potentials have been reported in patients with progressive myoclonic epilepsy (Shibasaki *et al.*, 1985; Dawson, 1947). Regions of epileptogenicity within the cortex have occasionally been reported to elicit seizures with high frequency direct cortical stimulation (Penfield and Jasper, 1954; Walker, 1949). The lack of clear association might be a result of the propagation of the high-frequency stimulation to distant cortical regions, such as the symptomatic or epileptogenic zones (Schulz *et al.*, 1997; Ishitobi *et al.*, 2000). It remains controversial whether the epileptogenic zone has a decreased threshold to induce a seizure as a result of an external electrical stimulus.

Local cortical responses to low-frequency stimulation have been reported and termed "direct cortical responses" (DCRs) (Adrian, 1936). DCRs from single or paired pulse stimulation have been studied in patients with mesial temporal lobe epilepsy (Wilson *et al.*, 1990; Wilson *et al.*, 1998; Valentin *et al.*, 2005 For Valentin 2005 indicate when 2005a and when 2005b]) and in patients with neocortical epilepsy (Valentin *et al.*, 2002; Valentin *et al.* 2005a; Valentin *et al.*, 2005b; Matsumoto *et al.*, 2005). They suggested that a cortical imbalance between excitation and inhibition is likely to be the pathophysiological basis for human partial epilepsy (Wilson *et al.*, 1990; Wilson *et al.*, 1998; Valentin *et al.*, 2005a). In the current study, we measured similar local cortical responses resulting from the direct electrical stimulation of various sites in temporal, frontal, and parietal neocortex. We refer to these responses as cortico-cortical evoked potentials (CCEPs) which we reported in our prior studies of functional cortical connectivity (Matsumoto *et al.*, 2004; Matsumoto *et al.*, 2007). We hypothesized that the ictal onset zone may produce CCEPs with larger amplitudes, shorter latencies, as well as lower stimulation thresholds, compared to regions outside the ictal onset zone.

Patients and methods

Study subjects

Ten patients (six male patients) with medically intractable neocortical epilepsy were prospectively recruited for this study. This study had the approval of the institutional review board committee of the Cleveland Clinic (IRB # 4513) and informed consent was obtained from all patients prior to the study. Consecutive patients were selected who underwent invasive video-EEG monitoring and were found to have electrocorticographic patterns suggestive of a focal ictal onset.

The study was performed extraoperatively in the epilepsy monitoring unit after the standard presurgical evaluation and after restarting antiepileptic medication. The patients were awake and relaxed during testing. The subdural electrode arrays consisted of platinum disc electrodes (diameter: 3.97 mm, inter-electrode distance: 10 mm) embedded in a silicon membrane (custom-made, Cleveland Clinic Foundation, Ohio). The relationship between electrode position and major cerebral sulci was identified on a three-dimensional reconstructed post-operative T1-weighted MRI (1.5 T, 2 mm slice thickness without interslice gap) or by high resolution volumetric CT (2 mm slice thickness) using the signal voids created by the electrodes (Hadar and Bingaman, 2002; Matsumoto *et al.*, 2004).

Cortico-cortical evoked potentials

The method of CCEPs has been described in more detail elsewhere (Matsumoto *et al.*, 2004). In brief, bipolar electrical stimulation was applied to adjacent electrodes using a Grass S88 stimulator (Astro-Med, Inc., RI). A constant current, monophasic square wave pulse with 0.3 ms duration, was delivered at a frequency of 1 Hz. An alternating stimulus polarity was used to counterbalance the stimulus artefacts and maintain charge balance.

Evoked potentials were recorded from the cortical surface by the electrodes around the stimulation site using the Epoch 2000 evoked potential measuring system (Axon Systems, Inc., NY) for the first seven patients and the Nihon Khoden EEG system (EEG 1000 Nihon Khoden, Japan) for the last three patients. We began using a second system so that raw data could be recorded for off-line analysis. An extracranial scalp electrode (contralateral mastoid) was used as the reference. The signals were band pass filtered between 1 and 800 Hz and sampled at 2,000 Hz for the first seven patients. In the last three patients, filtering was set from 1 to 300 Hz with a sampling rate of 1,000 Hz. Each average consisted of 10 to 54 stimuli *(table 1)* with a recording time of 200 ms duration and a 20 ms prestimulus baseline.

In each patient, two sites were selected for recording CCEPs; the ictal onset zone CCEP (iCCEP) and a site close to the ictal onset but not associated with ictal patterns (nCCEP). The electrodes selected for iCCEP recordings were based on onset of the initial ictal patterns of either a low voltage fast activity or repetitive spiking activity. The nCCEP electrodes were within the same lobe of brain as the iCCEP electrodes but not associated with ictal EEG activity in the first 30 seconds.

CCEPs were recorded with graded stimulus intensities in order to investigate the relationship between intensity and amplitude, as well as intensity and latency of the response. The bipolar stimulation of each subdural electrode pair was titrated from a low stimulus intensity of 1.0 mA, followed by increments of 1.0 or 2.0 mA, up to a maximum of 15 mA. The intensity was always delivered either below afterdischarge threshold or without eliciting a clinical response (such as muscle twitch or a sensation). The typical morphology of CCEP responses was characterized by a prominent negative deflection (N1). The N1 amplitude was measured from the pre-stimulus baseline to the peak of the N1 potential for each electrode. The group analysis was performed by taking the amplitude of the N1 response at the electrode showing the maximum response at the maximum stimulation intensity level for each patient. The maximum intensity level was defined as the highest stimulation applied to both iCCEP and nCCEP data without artefact. A higher stimulation was ignored if applied in one zone but not the other. Stimulus intensity versus N1 amplitude curve was also evaluated individually for each patient by comparing the iCCEP amplitude curve to the nCCEP amplitude curve. The electrode showing the highest N1 amplitude at greatest current intensity was used for the analysis of latency and amplitude at the various stimulation intensity grades.

Statistics

For each patient, a t-test for paired data was conducted to identify any difference between amplitude readings in the ictal onset zone and the non-ictal zone. However, for extremely small samples, or when data deviated from normality, a binomial sign test was used. Similar analysis was conducted for latency values. A binomial sign test was also used to determine if the amplitude measurement threshold occurred earlier in the ictal zone than in the non-ictal zone. Statistical significance was determined at the 5% level (*i.e.* $\alpha = 0.05$). When results were not available for both iCCEP and nCCEP, a pairing could not be made and the intensity level was not included.

Results

Demographics

The patient characteristics and details of invasive evaluation are described in *table 2*. Five patients had neocortical temporal lobe epilepsy (patients 1, 4, 6, 9 and 10), four had frontal lobe epilepsy (patients 2, 3, 7and 8) and one had perirolandic epilepsy (patient 5). Three patients had prior failed epilepsy surgery (patients 3, 4 and 7). Location of ictal onset zone measurements for iCCEP varied among patients: patient 1 had two ictal onset zones (right entorhinal cortex and posterior aspect of the right superior temporal gyrus); patient 2 (posterior aspect of right medial frontal gyrus); patient 3 (right orbitofrontal cortex); patient 4 (left middle temporal gyrus anterior to the glioma); patient 5 (left lower postcentral gyrus); patient 6 (parahippocampal gyrus); patient 7 (left orbitofrontal gyrus); patient 8 (right middle frontal gyrus); patient 9 (left superior temporal gyrus); and patient 10 (right middle temporal gyrus). The type and dosage of anticonvulsant medication during the recordings varied for each patient.

CCEPs were successfully recorded in eight of 10 patients. There were no symptoms or seizures induced during the procedure. Stimulus artefacts precluded analysis in two patients (patients 6 and 7). Response threshold intensity ranged between 3 and 8 mA for iCCEPs, and between 4 and 15 mA for nCCEPs. In Patient 1, two sets of iCCEP and nCCEP were studied for two separate ictal onset zones. CCEPs were not always recorded from all regions surrounding the stimulation, such as when the stimulation was performed at the edge of electrode array.

General characteristics of cortico-cortical evoked potentials

Cortico-cortical evoked potentials (CCEPs) consisted of two components: an initial stimulus artefact and a series of cortical potentials. The stimulus artefact occurred within 3-5 ms and consisted of a baseline drift with quick decay following the stimulus. CCEPs were then characterized by a prominent negative peak, labelled as N1, with a latency ranging from around 5 to 82 ms. The electrode which showed the largest N1 amplitude was typically adjacent to the stimulating pair.

Amplitude difference

The iCCEP and nCCEP amplitude measurements were compared and differences calculated for each maximum stimulus intensity level. The amplitude was higher in the iCCEP measurements than in nCCEP in seven of 10 patients. In one of 10 patients, the amplitude was higher in nCCEP than iCCEP measurements. CCEPs could not be recorded in two of 10 patients .The mean amplitude difference between iCCEP and nCCEP across all patients was 209.0 which was significantly greater in the ictal onset zone ($p = 0.002$).

For each patient, the iCCEP and nCCEP amplitude measurements were compared and differences calculated for each stimulus intensity level *(figures 1-3)*. The mean amplitude differences between iCCEPs and nCCEPs, and the number of iCCEP amplitude measurements that were greater than nCCEP amplitude measurements, can be seen in *table 1*. Patients 2, 3 and 4 had individual iCCEP amplitudes that were significantly greater than nCCEP amplitudes (with p-values equal to 0.0091, 0.0125 and

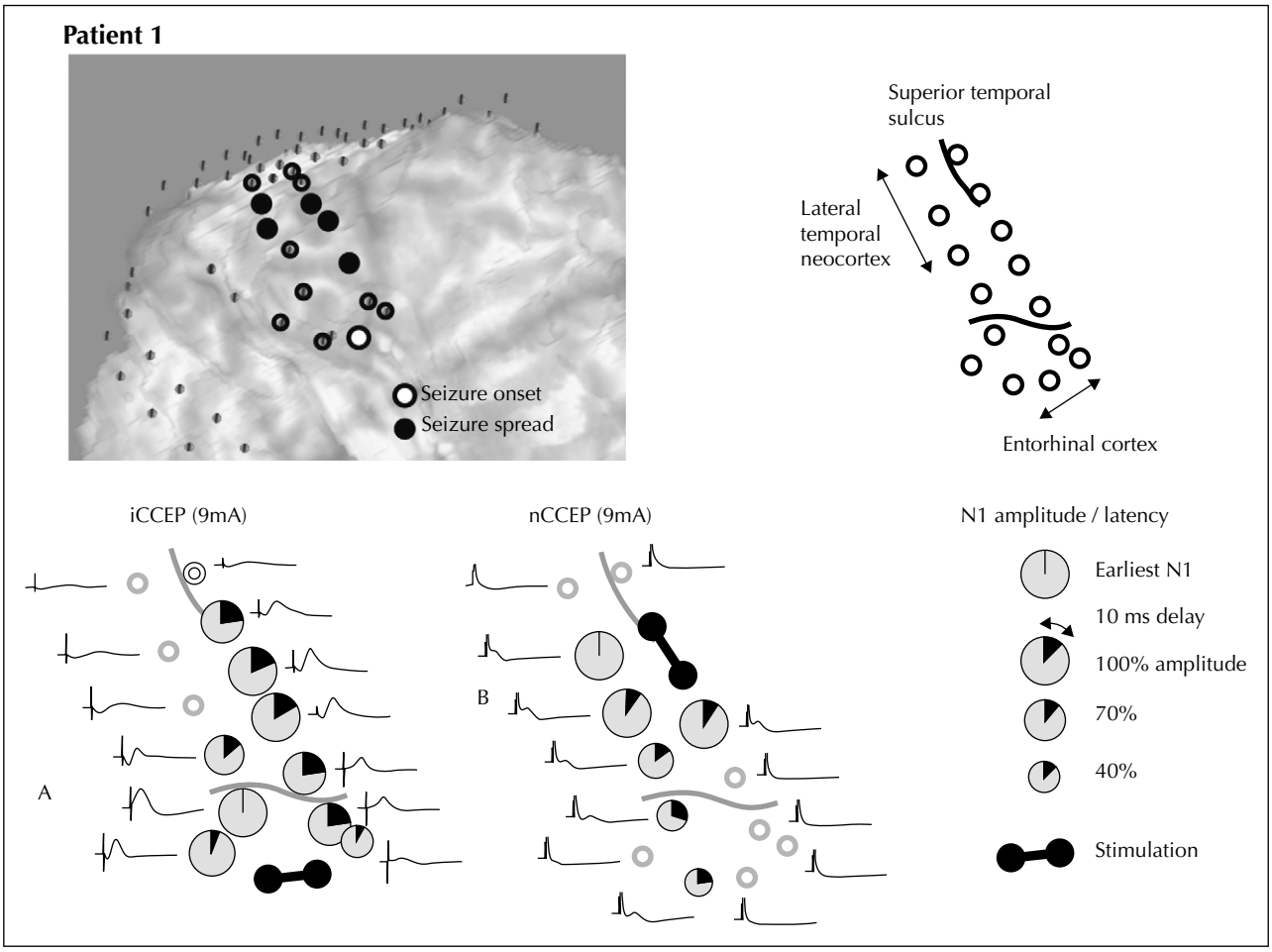

Figure 1. Illustration of the CCEPs in patient 1 (top row). The volume-rendered brain MRI of the right fronto-temporal region is displayed from the anterior inferior oblique view. One six-contact subdural strip electrode is placed over the right anterior basal temporal cortex towards the entorhinal cortex. One 2x6 subdural grid electrode is placed on the anterior lateral temporal cortex towards the temporo-polar region. CCEPs were recorded from circled electrodes. Anatomical relationship is depicted next to the MRI. One form of ictal EEG initiated at the electrode over the entorhinal cortex and spread to the anterior lateral temporal region (bottom row). CCEPs after stimulation of the ictal onset zone and after stimulation outside the ictal onset zone are compared. Stimulus intensity was 9 mA for both. Size of the full circles is proportional to the amplitude of N1 peak for the given stimulation. In this way, a distribution of the response is shown for the given stimulation and they cannot be used for comparison between stimulations (such as between iCCEP or nCCEPs data sets). 100% amplitude is assigned to the electrode with the largest N1 response. The angle of pie (painted in black) shows the relative latency of N1, *i.e.* latency delay from the earliest N1.

0.0437, respectively). An example of the amplitude and stimulus intensity curves, demonstrating how iCCEP stimulation produced a left shift in the curve compared to the nCCEP curve, is seen in *figure 2*. Two ictal onset zones were stimulated in this patient as shown in *figure 2*, however the first ictal onset zone had a maximum stimulation intensity of 4 mA with higher intensities producing a motor response. In the second ictal onset zone, higher stimulation intensities could be delivered without afterdischarges or eliciting a clinical response, therefore this area was used in the statistical analysis. However both regions, as shown in the graph, had similar slope and left shift as compared to the non-ictal site.

There was no statistical difference of the individual amplitude measurements between iCCEP and nCCEP in patients 1, 5, 8, 9 and 10. Two ictal and non-ictal zones were recorded for patient 1. Although the amplitudes in the first ictal onset zone were slightly higher than in the non-ictal zones, this was not true for the second ictal and non-ictal pair in this patient. In patients 8, 9 and 10, the iCCEPs were of larger amplitude than the nCCEPs but did not reach statistical significance individually. The percentage of times in which the iCCEP amplitudes were greater than the nCCEP at all the incremental stimulations combined were also more frequent in patients 2, 3, 4 and 10, but this did not reach statistical significance.

Table 1. Amplitude of CCEPs responses to graded stimulation intensity. Two positions of ictal and non-ictal stimulation were tested for patient 1.

Patient Number	Stimulation Intensity (mA)	Number of Averages	iCCEP Amplitude (μV)	nCCEP Amplitude (μV)	Mean Difference (iCCEP-nCCEP) (μV)	iCCEP > nCCEP	Percentage of Stimulations with Greater iCCEP amplitudes
1	9	20	191	106		Yes	
(1st position)	7	20	126	143		No	
	5	20	25	0		Yes	
	3	20	0	0	- 26.7	No	25%
1	9	20	337	521	p = 0.7460	No	(2/8)
(2nd position)	7	20	138	270		No	
	5	20	0	119		No	
	3	20	0	0		No	
2	15	10	No data	1032.6	616.3	-	83.3%
	12	10	No data	904.2	p = 0.0091*	-	(5/6)
	10	10	1560	751.1		Yes	p = 0.1094
	9	10	1486.6	No data		-	
	8	10	1376	257		Yes	
	7	10	1131.8	No data		-	
	6	10	1025.2	32		Yes	
	5	10	824.6	No data		-	
	4	10	532.4	0		Yes	
	3	10	244.6	0		Yes	
	2	10	0	0		No	
3	14	10	No data	0	801.8	-	100.0%
	12	10	No data	0	p = 0.0125*	-	(4/4)
	10	10	1111	0		Yes	p = 0.0625
	9	10	1108	No data		-	
	8	10	1093	0		Yes	
	6	10	703	0		Yes	
	4	10	300	0		Yes	
	3	10	0	No data		-	
	2	10	No data	0		-	
4	11	20	575	144	190.3	Yes	60.0%
	9	20	372.8	76.5	p = 0.0437*	Yes	(3/5)
	7	20	224.2	0		Yes	p = 0.5000
	5	20	0	0		No	
	3	20	0	0		No	
5	6	20	123	326.4	-100.5	No	33.3%
	5	20	66.8	236.8	p = 0.8375	No	(1/3)
	4	20	51.8	0		Yes	p = 0.8750
8	8	54	268	181	34.77	Yes	50.0%
	4	54	111	59.1	p = 0.1008	Yes	(2/4)
	2	54	0	0		No	p =0.6875
	1	54	0	0		No	
9	15	54	186.9	61.1	48.15	Yes	50.0%
	8	54	66.8	0	p = 0.1051	Yes	(2/4)
	4	54	0	0		No	p = 0.6875
	2	54	0	0		No	
10	8	52	746	564	69.87	Yes	66.7%
	4	52	65.8	38.2	p = 0.343	Yes	(2/3)
	2	52	0	0		No	p = 0.5000

iCCEP: CCEPs of ictal onset zone; nCCEP: CCEPs of neocortex uninvolved with the ictal EEG pattern; "Yes" means the amplitude of iCCEP was larger than nCCEP; "No" means the amplitude of iCCEP was smaller or the same as nCCEP; * Indicates significance; No data: stimulation not applied.

Table 2. Summary of patient characteristics and details of their invasive evaluation.

Patient Number	Age (Sex)	Etiology/ Pathology	MRI location of lesion	Ictal onset zone	Anticonvulsant (Dosage)	Surgery	Seizure outcome (Follow-up)
1	27 (F)	Encephalomalacia	Right F-P	First: Entorhinal Second: Posterior STG	Levetiracetam (1,000 mg bid) Zonisamide (600 mg qd) Phenytoin (1,000 mg load)	Right T Lobectomy	Engel Class II 75% Reduction (1.5years)
2	15 (M)	Cortical Dysplasia	Normal	Posterior MFG	Phenytoin (900 mg load)	Right Mesial F Resection	Engel Class Ia Single Postsurgical Seizure (1.5 years)
3	47 (F)	Unknown (Gliosis)	S/P Right T Lobectomy	OF	Phenytoin (750 mg load)	Right OF and T Resection	Engel Class Ia Two Postsurgical Seizures (1.5 years)
4	24 (M)	Low Grade Glioma	S/P Left F Tumour	MTG: Anterior to lesion	Levetiracetam (1,500 mg bid) Phenytoin (200 mg bid)	Left T Resection	Engel Class Ia Seizure Free (1.5 years)
5	45 (M)	Encephalomalacia	Remote hemorrhagic contusion Left O	Lower Postcentral Gyrus	Levetiracetam (1,000 mg bid) Carbatrol (500 mg AM, 200 mg noon, 600 mg PM-load) Klonopin (1 mg AM, 0.5 mg noon and PM)	Left Peri-rolandic Resection	Engel Class Ib Only Auras (1.5 years)
6	36 (M)	Cortical Dysplasia	Normal	Parahippo-campal Gyrus	Topiramate (175 mg AM, 200 mg PM) Carbamazepine (1,000 mg bid)	Right T Lobectomy	Engel Class III 50% Reduction (2 years)
7	9 (M)	Cortical Dysplasia	S/P Left OF Resection	OF	Oxcarbazepine (600 mg AM, 900 mg PM)	Left OF Resection	Engel Class Ia Seizure Free (2 years)
8	35 (F)	Cortical Dysplasia	Normal	Middle frontal gyrus	Carbamazepine (400 mg bid) Pregabalin (50 mg AM, 100 mg PM)	Right superior and middle frontal gyrus	Engel Class Ia Seizure Free (1 year)
9	12 (M)	Cortical Dysplasia	Left STG and Parietal operculum	STG	Oxcarbazepine (600 mg bid) valproate (250 mg bid)	Left temporo-parietal resection	Engel Class Ia Seizure Free (1 year)
10	52 (F)	Unknown (Gliosis)	Normal	MTG	Lamotrigine (100 mg bid) Carbamazepine (200 mg bid)	Right middle temporal gyrus	Engel Class II 75% Reduction (1 year)

S/P: status post; F: frontal; T: temporal; F-P: frontoparietal; OF: orbitofrontal; O: opercular; STG: superior temporal gyrus; MFG: medial frontal gyrus; MTG: middle temporal gyrus; bid: twice daily: qd: daily.

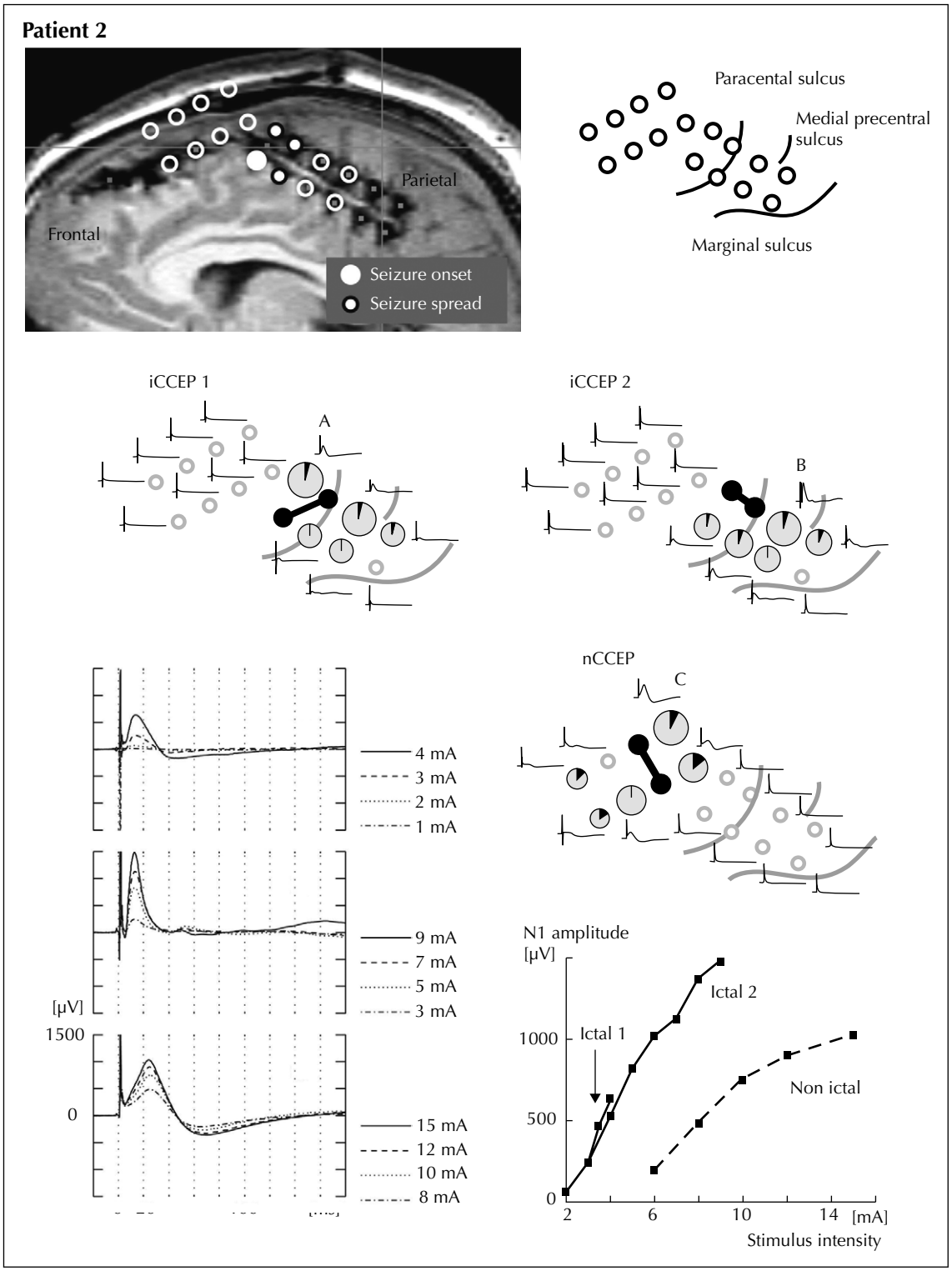

Figure 2. Illustration of the CCEPs in patient 2. Sagittal section of the patient's MRI shows one 2 x 6 grid placed in the right mesial fronto-parietal cortex and another 2 x 6 grid placed anteriorly over the mesial frontal and superior frontal gyrus. CCEPs were recorded from 14 of these electrodes, as shown in the image. The stimulation at the ictal onset zone produced larger N1 responses. The stimulus intensity *versus* N1 amplitude curve from the iCCEP showed greater increment than that from the nCCEP.

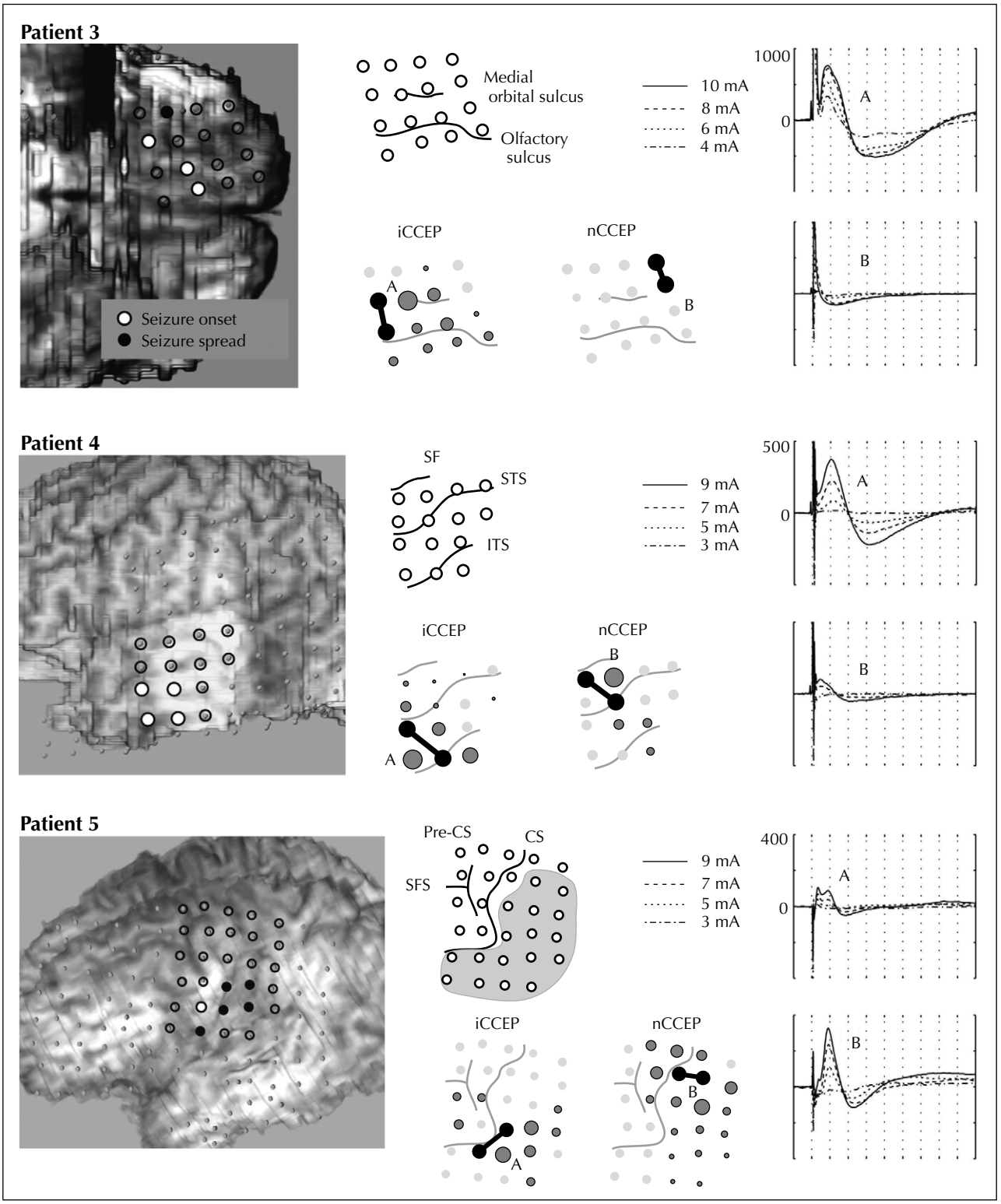

Figure 3. Results for patients 3, 4 and 5. CCEPs were recorded from the right orbito-frontal cortex (patient 3), left lateral temporal cortex p 4), and left perirolandic cortex (patient 5). The latencies and CCEP waveforms are shown on the right with the iCCEP (A) and the nCCEP (B). The stimulation at the ictal onset zone evoked larger N1 in patients 3 and 4. In patient 5, the stimulation outside the ictal onset zone produced larger responses. The grey-painted area in patient 5 shows the area of cortical atrophy.

Latency difference

Similar to the amplitude analysis, iCCEP and nCCEP maximum intensity latency measurements were compared both as a group as well as individually. Patients 2 and 5 had shorter iCCEP latency values (12.8 ms and 5.4 ms) than nCCEP latency values (25.7 ms and 18.3 ms) but this did not reach statistical significance. Latencies were not found to be statistically shorter in duration in the other patients, with iCCEP responses ranging from 5.4 to 82 ms compared to the nCCEP responses ranging from 12.7 to 67 ms.

Amplitude threshold

In order to determine whether amplitude thresholds were seen at lower stimulus intensities for iCCEP measurements compared to nCCEP, a determination was made for each of the eight patients indicating whether iCCEP was observed prior to nCCEP. Six of the eight patients (75%) had amplitude thresholds that were lower for iCCEP than nCCEP *(table 1)*. For patient 3, iCCEPs were recorded first at 4 mA whereas nCCEPs were never recorded up to 14 mA. However, a lower amplitude threshold in iCCEP regions did not reach statistical significance for all cases.

Seizure free outcome after surgery

All eight patients underwent epilepsy surgery with removal of tissue, including the tissue associated with the ictal onset zone. Of the eight patients in whom CCEPs could be recorded, six (patients 2, 3, 4, 5, 8 and 9) had good seizure outcome *(table 2)*. Patients 4, 8 and 9 were completely seizure free for one year or longer, whereas patients 2 and 3 had one or two postoperative seizures but eventually became seizure free. Although patient 5 had a good clinical surgical outcome, he continued to have frequent auras. This suggests that the epileptogenic zone was incompletely resected. Patient 1 had a reduction of her seizures following surgery but continued to have seizures after surgery. The three patients in whom iCCEPs amplitudes showed significant individual differences all had good seizure free outcome.

Discussion

The principal finding of this study is that, based on group analysis in patients with focal neocortical epilepsy, low-frequency stimulation at ictal onset zones produces a more marked increase in CCEP amplitude compared to the stimulation outside the ictal onset zone. While individual differences in amplitudes elicited in three patients were significantly higher in the ictal onset zone, this was not true for the remaining five patients. The measurements of amplitude change in this study were performed using graded stimulation intensities. By doing so, the stimulation and amplitude curves shifted to the left of the curve for CCEP in the ictal onset zones. The presence of enhanced amplitudes in ictal onset zones may be a reflection of the increased excitability of the cortex associated with epileptogenicity in these regions in some patients with neocortical epilepsy. It should be noted that regions selected as controls (nCCEP) were not systematically analyzed for any underlying pathology. Therefore, nCCEP may not necessarily represent responses from normal cortex, however the comparisons with iCCEPs were made in regions of similar anatomical localization. Patient 5 was the only patient in whom the underlying cortex showed an imaging difference between iCCEP and nCCEP. In this patient, the ictal onset electrode had an underlying encephalomalacia whereas the non-ictal electrode did not. This selected non-ictal region also showed eloquent function by standard cortical stimulation. This may explain why this patient also showed the opposite relationship in which the nCCEPs were larger than iCCEPs. It may be important in future studies to compare not only similar anatomical regions but differentiate primary versus association cortices, as well as lesional versus non-lesional areas.

The latencies of these responses occurred within a short period after the stimulation onset (5 and 82 ms). It is known that electrical pulse stimulation of the neocortical surface produces stereotypical synaptic responses adjacent to the stimulation called DCRs (Adrian, 1936; Barth and Sutherling, 1988). DCRs are characterized by a prominent negative peak around 10-20 ms, followed by slow positive and negative potentials lasting up to 200 to 300 ms (Barth and Sutherling, 1988; Goldring *et al.*, 1961; Purpura *et al.*, 1957). Our study noted larger variations and more prolonged latencies than described in DCR studies. Some explanations include the variations noted in abnormal cortex or the effect of anticonvulsant medication. However, the more likely explanation may involve the significant differences in the electrode sizes and interelectrode distances, both of which were much smaller in the DCR studies. The larger surface area of the recording electrodes used in our study would enable recording over a larger population of pyramidal neurons which may have some variable jitter in their individual responses (Matsumoto *et al.*, 2004). If so, then this jitter may have blunted the N1 peak, producing a longer latency. Latencies recorded in our study were not significantly decreased in the ictal onset zone compared to the non-ictal onset regions as might be expected when comparing regions of potentially different synaptic excitability. Other reports that have studied late responses, ranging from 100 ms to one second after an inter-ictally applied single-pulse stimulation, effectively predicted the topography of seizure onset in temporal lobe epilepsy (Valentin *et al.*, 2002) as well as in frontal lobe epilepsy (Valentin *et al.*, 2005a). When resected, these areas tended to pre-

dict good surgical outcome in frontal and temporal lobe epilepsy (Valentin *et al.*, 2005b). These late responses were not evaluated in our study. Early responses, similar to CCEPs, have been recorded in other studies (Valentin *et al.*, 2002, 2005a, 2005b) but were not associated with regions of seizure onset. The main difference between the methodologies of these studies and ours is that our study compared amplitudes of the evoked response to the stimulation intensity given in a graded fashion. This allowed for a comparison of amplitude response to the intensity of stimulation between various neocortical sites.

The electrical stimulation used in our study was similar to that in the previous DCR studies. The CCEPs most likely share common generator mechanisms with DCRs in the initiation of the response and involve the cortico-cortical projection neurons for the relatively distant transmission of the responses. We speculate that CCEPs vary depending on the efficiency of excitatory synaptic connections between the stimulated cortex and the remote cortex. Previous investigators revealed that positive potential (P1) and N1 DCRs were associated with the occurrence of frequent action potentials and excitatory postsynaptic potentials (EPSPs) in the apical dendrites of pyramidal cells (Barth and Sutherling, 1988; Creutzfeldt *et al.*, 1966). The cortical stimulation produces multisynaptic excitatory responses in the local cortical circuits (Douglas *et al.*, 1995), mainly mediated by the ascending recurrent axon collaterals of the pyramidal neurons and partly by excitatory interneurons (Barth *et al.*, 1989). This excitatory process is followed by inhibitory postsynaptic potentials (IPSPs) and longer periods of hyperpolarization (DCR-P2) (Barth and Sutherling, 1988). The amplitude and time course of the N1 probably reflects the summation of EPSPs occurring after multisynaptic excitations and secondary inhibition by IPSPs. The region of maximum amplitude change was noted in the electrode adjacent to the electrodes being stimulated. This may reflect increased local excitability of the epileptic cortex.

Initial attempts to study cortical stimulation as a surrogate marker for the epileptogenic zone involved looking at afterdischarge thresholds and induction of typical auras or seizures (Lesser *et al.*, 1984; Luders *et al.*, 1988; Penfield and Jasper, 1954; Walker, 1949). We assessed whether stimulus threshold to evoke CCEP was lower in regions near ictal onset compared to non-ictal onset zones but found no statistical significant difference.

This preliminary study suggests that CCEPs close to ictal onset regions may be affected by the excitability of the cerebral cortex in some patients with focal epilepsy. We did not show this to be the case in each patient individually, but this was demonstrated when comparison was performed within the entire group of patients studied. It is interesting that the three patients who did show significant individual amplitude differences all had good seizure outcome following epilepsy surgery. Whether the finding of a significant difference of amplitude between iCCEP and nCCEP regions in an individual patient has good correlation with seizure-free outcome after surgery is unclear based on the small number of patients in which this was observed in our study.

A previous study suggested that cellular responses to external electrical stimulation are enhanced in the epileptogenic cortex (Matsumoto *et al.*, 2005). *In vivo* cellular recordings from lateral temporal cortex in patients with intractable epilepsy showed that "epileptic" neurons with spontaneous high-frequency bursts are more likely to generate evoked single unit activities after direct cortical stimulation than do "normal" firing neurons (Wyler and Ward Jr., 1981). Techniques similar to CCEPs have been applied before in human subjects to evaluate epileptogenicity. Wilson *et al.* (1990) used evoked potentials elicited by stimulation of the limbic structures to investigate the "preferred pathway" of epileptic activity. They did not find increased "response probability" when stimulating the epileptogenic region. Rutecki *et al.* (1989) found that the entorhinal-evoked hippocampal potentials showed different waveform configuration among patients with and without hippocampal sclerosis. Stereotactic depth electrode studies (Buser and Bancaud, 1983) reported that evoked responses in the amygdala after hippocampal stimulation were exclusively observed in patients with temporal lobe epilepsy but less in other types of epilepsy. It was speculated that epileptogenesis could generate new synaptic pathways which were normally absent.

Some limitations of this study include the evaluation of only predefined regions of the cortex in each case and the variation of ictal EEG patterns, which makes the identification of the electrodes involved in the initial changes challenging. This study did not attempt to evaluate the ictal patterns and their association to CCEP amplitudes; however, this would be an interesting consideration for future studies. Another issue that may have affected these findings is that the patients were receiving the typical doses of anticonvulsants at the time of this study. What effect anticonvulsants would have had on the CCEP recordings is not clear. For two of our patients, CCEP recording could not be accomplished due to stimulus artefacts. This might limit the usefulness of this technique in all patients with intracranial recordings. Techniques that resolve stimulus artefacts could enhance the usefulness of this technique and future efforts should focus on several regions of the cortex without knowledge of the ictal EEG information.

In conclusion, the CCEPs recorded, following stimulation of ictal onset zones, were shown to increase in comparison to similar regions of cortex not involved in the ictal activity in our group of patients with focal neocortical epilepsy. This difference was only individually significant in three of eight patients. The seizure-free outcome following surgery was good for the three in whom the difference was individually significant. The number of patients studied in this study was small. Thus, we consider this study to

be preliminary and a larger study necessary to validate this finding. The ability to accurately record regions of ictal onset in invasive EEG can be challenging (Ebner and Lüders, 2001). Therefore, exploring other surrogate markers for epileptogenicity could aid in the detection of these regions. □

Acknowledgments.
We wish to thank Dr Hans Lüders for his comments on the protocol for this study as well as Ms Elaine Gottlieb for her editorial comments of the paper.

Disclosure.
None of the authors has any conflict of interest or financial support to disclose.

References

Adrian ED. The spread of activity in the cerebral cortex. *JPhysiol* 1936; 88: 127-61.

Barth DS, Sutherling W. Current source-density and neuromagnetic analysis of the direct cortical response in rat cortex. *Brain Res* 1988; 450: 280-94.

Barth DS, Di S, Baumgartner C. Laminar cortical interactions during epileptic spikes studied with principal component analysis and physiological modeling. *Brain Res* 1989; 484: 13-35.

Buser P, Bancaud J. Unilateral connections between amygdala and hippocampus in man. A study of epileptic patients with depth electrodes. *Electroencephalogr Clin Neurophysiol* 1983; 55: 1-12.

Carreno M, Luders H. General Principles of Presurgical Evaluation. In: Lüders H, Comair Y, eds. *Epilepsy Surgery*. Philadelphia: Lippincott Williams & Wilkins, 2001: 185-99.

Creutzfeldt OD, Watanabe S, Lux HD. Relations between EEG phenomena and potentials of single cortical cells. I. Evoked responses after thalamic and erpicortical stimulation. *Electroencephalogr Clin Neurophysiol* 1966; 20: 1-18.

Dawson GD. Investigations on a patient subject to myoclonic seizures after sensory stimulation. *J Neurol Neursurg Psychiatry* 1947; 10: 141-62.

Douglas RJ, Koch C, Mahowald M, Martin KA, Suarez HH. Recurrent excitation in neocortical circuits. *Science* 1995; 269: 981-5.

Ebner A, Luders H. Subdural Electrodes. In: Lüders H, Comair Y, eds. *Epilepsy Surgery*. Philadelphia: Lippincott Williams & Wilkins, 2001: 593-6.

Goldring S, Jerva MJ, Holmes TG, O'Leary JL, Shields JR. Direct response of human cerebral cortex. *Arch Neurol* 1961; 4: 590-8.

Hadar EJ, Bingaman WE. Surgery for hemispheric malformations of cortical development. *Neurosurg Clin N Am* 2002; 13: 103-11 (ix.).

Ishitobi M, Nakasato N, Suzuki K, Nagamatsu K, Shamoto H, Yoshimoto T. Remote discharges in the posterior language area during basal temporal stimulation. *Neuroreport* 2000; 11: 2997-3000.

Lesser RP, Luders H, Klem G, Dinner DS, Morris HH, Hahn J. Cortical afterdischarge and functional response thresholds: results of extraoperative testing. *Epilepsia* 1984; 25: 615-21.

Lüders H, Lesser RP, Dinner DS, Morris HH, Wyllie E, Godoy J. Localization of cortical function: new information from extraoperative monitoring of patients with epilepsy. *Epilepsia* 1988; 29 (Suppl. 2): S56-65.

Matsumoto R, Nair DR, LaPresto E, Bingaman W, Shibasaki H, Luders HO. Functional connectivity in human cortical motor system: a cortico-cortical evoked potential study. *Brain* 2007; 130: 181-97.

Matsumoto R, Kinoshita M, Taki J, *et al*. In vivo epileptogenicity of focal cortical dysplasia: a direct cortical paired stimulation study. *Epilepsia* 2005; 46: 1744-9.

Matsumoto R, Nair DR, LaPresto E, *et al*. Functional connectivity in the human language system: a cortico-cortical evoked potential study. *Brain* 2004; 127: 2316-30.

Nair DR, Burgess R, McIntyre CC, Lüders H. Chronic subdural electrodes in the management of epilepsy. *Clin Neurophysiol* 2008; 119: 11-28.

Penfield W, Jasper H. *Epilepsy and the Functional Anatomy of the Human Brain*. Boston, MA: Little Brown, 1954.

Purpura DP, Pool JL, Ransohoff J, Frumin MJ, Housepian EM. Observations on evoked dendritic potentials of human cortex. *Electroencephalogr Clin Neurophysiol Suppl* 1957; 9: 453-9.

Rutecki PA, Grossman RG, Armstrong D, Irish-Loewen S. Electrophysiological connections between the hippocampus and entorhinal cortex in patients with complex partial seizures. *J Neurosurg* 1989; 70: 667-75.

Schulz R, Luders HO, Tuxhorn I, *et al*. Localization of epileptic auras induced on stimulation by subdural electrodes. *Epilepsia* 1997; 38: 1321-9.

Shibasaki H, Yamashita Y, Neshige R, Tobimatsu S, Fukui R. Pathogenesis of giant somatosensory evoked potentials in progressive myoclonic epilepsy. *Brain* 1985; 108 (Pt 1): 225-40.

Valentin A, Alarcon G, Garcia-Seoane JJ, *et al*. Single-pulse electrical stimulation identifies epileptogenic frontal cortex in the human brain. *Neurology* 2005a; 65: 426-35.

Valentin A, Alarcon G, Honavar M, *et al*. Single pulse electrical stimulation for identification of structural abnormalities and prediction of seizure outcome after epilepsy surgery: a prospective study. *Lancet Neurol* 2005b; 4: 718-26.

Valentin A, Anderson M, Alarcon G, *et al*. Responses to single pulse electrical stimulation identify epileptogenesis in the human brain in vivo. *Brain* 2002; 125: 1709-18.

Walker AE. Electrocorticography in Epilepsy. *Electroencephalogr Clin Neurophysiol Suppl* 1949; 2: 30-9.

Wilson CL, Khan SU, Engel Jr J, Isokawa M, Babb TL, Behnke EJ. Paired pulse suppression and facilitation in human epileptogenic hippocampal formation. *Epilepsy Res* 1998; 31: 211-30.

Wilson CL, Isokawa M, Babb TL, Crandall PH. Functional connections in the human temporal lobe. I. Analysis of limbic system pathways using neuronal responses evoked by electrical stimulation. *Exp Brain Res* 1990; 82: 279-92.

Wyler AR, Ward Jr AA. Neurons in human epileptic cortex. Response to direct cortical stimulation. *J Neurosurg* 1981; 55: 904-8.