ELSEVIER

Epilepsy Research 26 (1997) 373–380

EPILEPSY RESEARCH

Epileptiform activity but not synaptic plasticity is blocked by oxidation of NMDA receptors in a chronic model of temporal lobe epilepsy

O. Quesada, J.C. Hirsch, H. Gozlan, Y. Ben-Ari, C. Bernard*

INSERM U29, Hôpital de Port Royal, 123 Bld de Port Royal, 75014 Paris, France

Received 1 July 1996; revised 1 September 1996; accepted 12 September 1996

Abstract

Simultaneous extracellular recordings were performed in stratum radiatum and stratum pyramidale of hippocampal slices 7 days following unilateral intracerebroventricular injections of kainic acid. In this ex vivo experimental model of human temporal lobe epilepsy, stimulation of the surviving commissural fibres in stratum radiatum produced graded epileptiform activity in the CA1 area. The oxidizing reagent 5,5′-dithiobis (2-nitrobenzoic acid) (DTNB) acting at NMDA receptors redox sites decreases NMDA receptor-mediated responses by half and suppresses evoked epileptiform discharges. We have examined the effect of DTNB on NMDA-dependent bidirectional synaptic plasticity and EPSP/spike coupling. DTNB treatment did not prevent either long-term potentiation induced by tetanic stimulation or long-term depression induced by low frequency stimulation of field EPSPs. Application of DTNB alone did not induce EPSP/spike dissociation. However, both high and low frequency stimulations induced EPSP/spike potentiation indicating that neurons had a high probability to discharge in synchrony. These results suggest that oxidizing reagents may provide novel antiepileptic treatments since they decrease NMDA-dependent evoked epileptiform activity but do not interfere with either NMDA-dependent synaptic plasticity or the probability of synchronous discharge.

Keywords: Temporal lobe epilepsy; Anticonvulsant drug; NMDA receptors; Long-term potentiation; Long-term depression

1. Introduction

A common feature to human epileptic tissue as well as to most chronic models of temporal lobe epilepsy (TLE) is that *N*-methyl-D-aspartate

* Corresponding author. Tel.: +33 153737931; fax: +33 146341656; e-mail: bernard@cochin.inserm.fr

0920-1211/97/$17.00
PII S0920-1211(96)01004-2

(NMDA) receptors participate in synaptic transmission and epileptiform activity [3,4,23,30] in contrast to non-lesioned tissue [1]. The pharmacological blockade of NMDA receptors would indeed prevent evoked epileptiform discharges [3,4] but would also impair NMDA-dependent alpha-amino-3-hydroxy-5-methylisoxazolepropionic acid (AMPA) receptor-mediated bidirectional synaptic plasticity [5]. Alternatively, one may try to manipulate the multiple regulatory sites on NMDA receptors, which fine tune their activity in order to decrease but not abolish, NMDA receptor-mediated neurotransmission. Potentially, this could prevent the over activation of these receptors without affecting NMDA receptor-triggered synaptic plasticity. In the present study, attention was focused on redox potential modulations.

The model of TLE which we have used is the kainic acid (KA) lesioned rat hippocampus, a chronic model of TLE relevant to the human condition [6,24,31]. Following unilateral intracerebroventricular injection of KA, most CA3 pyramidal cells degenerate. As a consequence, their Schaffer collaterals disappear, leaving the commissural fibres as the main excitatory input to CA1 pyramidal cells [25]. The deafferentation triggers a reactive synaptogenesis in the proximal dendrites of the CA1 pyramidal cells [26,28] which seems to originate from their axon collaterals [27]. Seven days following the lesion, CA1 pyramidal cells demonstrate NMDA-dependent epileptiform activity [3] associated with a large participation of NMDA receptors in synaptic transmission [30]. In this preparation as in control tissue, NMDA-dependent bidirectional synaptic plasticity can be induced [8,9,12] and also EPSP/spike potentiation, i.e. an increase of the probability of synchronous discharge, which occurs whatever the electrical conditioning stimulus used [10–12]. Finally, we have recently reported that in this model the oxidizing reagent 5,5′-dithiobis (2-nitrobenzoic acid) (DTNB) irreversibly decreases NMDA receptor-dependent evoked epileptiform activity [19,29]. This phenomenon occurs via the oxidation of NMDA receptors by DTNB, which decreases by 50% NMDA receptor-mediated responses [19]. It was then suggested that DTNB was potentially an important candidate for pharmacological treatment of TLE. In order to demonstrate that manipulation of the redox site was less deleterious than the full blockade of NMDA receptors it was necessary to assess the effect of DTNB on synaptic plasticity. We have thus investigated in the kainate rat the consequences of applying DTNB on bidirectional NMDA-dependent AMPA receptor-mediated synaptic plasticity and on EPSP/spike coupling (i.e. the probability of neurons to discharge in synchrony). These are key issues if DTNB, or other oxidizing drugs, are to be used for pharmacological treatment of TLE.

2. Materials and methods

2.1. Experimental preparation

Kainic acid lesions were performed on male Wistar albino rats (180 g) following established procedures [21]. Rats were anaesthetized with sodium pentobarbitone, 100 mg/kg intraperitoneal and placed in a stereotaxic frame. A hole was drilled in the cranium at 3.5 mm posterior to bregma and 3.9 mm lateral to the midline. The needle of an injection syringe was inserted into the lateral ventricle at a depth of 4.0 mm below the surface of the cortex. KA (0.5 μg) in a volume of 0.5 μl of pH 7.4 phosphate buffer was then infused into the lateral ventricle over a 30 min period. The syringe was left in position for another 10 min to allow diffusion away from the tip before the needle was removed and the scalp incision sutured.

At post-operative day 7, animals were anaesthetized with 0.4 ml of chloralose 7% and intracardially perfused with 100 ml of oxygenated ice cold modified artificial cerebrospinal fluid (ACSF, pH 7.4). Hippocampi ipsilateral to the lesion were removed and sliced (400 μm) using a McIlwain tissue chopper. Slices were incubated in oxygenated ACSF for at least 1 h at room temperature before recording and then placed in a submerged in vitro slice chamber (temperature: 30–32°C). The medium was aerated with 95% 0_2 and 5% CO_2. The ACSF was composed of (in mM) NaCl, 124.0; KCl, 3.0; KH_2PO_4, 1.25;

$NaHCO_3$, 26; $MgSO_4$-$7H_2O$, 1.3; $CaCl_2$, 2.0 and D-glucose, 10. The initial stages (intracardiac perfusion and slicing) were performed with a modified ACSF in which NaCl was replaced with an equimolar concentration of sucrose; Ca^{2+} was lowered to 0.5 mM and Mg^{2+} was raised to 6 mM.

Extracellular population spikes (PSs) and field EPSPs were recorded using 5–10 MΩ resistance glass microelectrodes filled with ACSF. Bipolar twisted NiCr wire stimulation electrodes (trimel insulated apart from 50 μm at the tip) were placed in the stratum radiatum of CA1 area proximally to stratum pyramidale and 0.5 mm from the recording electrodes. Stimuli (40 μs pulses, 0.5–15 μA) were delivered at 30 s (0.033 Hz) intervals. Responses in hippocampi ipsilateral to the lesion were selected if they were characterized by an epileptiform response composed of at least three PSs at saturation. Tetanic stimulations were applied through the same bipolar electrode and consisted of four trains of ten shocks at 100 Hz every 1 s. LFS consisted of a train of 900 shocks at 1 Hz during 15 min. Population spike amplitudes (PSAs) were measured as the distance between the peak negativity and the line joining the two positivities of the PS. Population EPSP slopes were measured as the initial slope taken during the first 0.5 ms after the afferent volley. Increase or decrease of PSAs and population EPSP slopes were considered as long-term if they were sustained for at least 30 min. The amplitude of the afferent volley was monitored throughout the experiments and was seen to be stationary. After obtaining a stable baseline (10 min), DTNB (200 μM) was applied for 15 min and then washed from the solution over 30 min, before applying the conditioning train. EPSP/spikes curves were constructed from full input/output curves obtained at the beginning of the experiment, 30 min after the end of application of DTNB and 30 min after the conditioning train. The quantification of EPSP/spike dissociation, I_{ES}, has been described in detail elsewhere [11,12]. Briefly, for each experiment, the population EPSP whose slope was 75% of the maximum evoked prior to any manipulation was taken as control. All PSAs whose corresponding population EPSP slope matched the control were measured using the input/output curves. The index of dissociation, I_{ES}, was taken as $I_{ES} = (PSA_{after}-PSA_{control})/PSA_{control}$. At the end of the experiment D-APV (50 μM) was applied in order to fully block NMDA receptors in order to assess the level of decrease of epileptiform activity achieved by DTNB treatment.

2.2. Data analysis

Extracellular signals were fed to a WPI amplifier. All data were digitized (10 kHz) with a Labmaster interface card to a personal computer and analyzed with Acquis1 program (G. Sadoc, DIPSI, Asnieres, France). Parameters were compared with a Student's *t*-test for paired samples, for evaluation of differences between means. All values are expressed as mean ± S.D.

2.3. Drugs

Kainic acid and DTNB were purchased from Sigma and Calbiochem respectively. D-APV was obtained from Dr Boddeke (Sandoz). D-APV and DTNB were diluted to the desired concentrations in oxygenated ACSF and bath applied.

3. Results

3.1. Extracellular characteristics of field recordings

In slices ipsilateral to the lesion, paired pulse inhibition was impaired as previously reported [14] and the test stimuli evoked epileptiform discharges composed from four to six PSs (Fig. 1 and Fig. 2). Data was collected from 13 slices from 13 lesioned animals which fulfilled the selection criteria. The mean population EPSP slope was 1.79 ± 0.79 mV/ms and the mean amplitude of the first four PSs were 2.56 ± 0.49, 1.13 ± 0.21, 0.54 ± 0.12, 0.32 ± 0.09 mV respectively ($n = 13$). These values are in keeping with those previously reported [12].

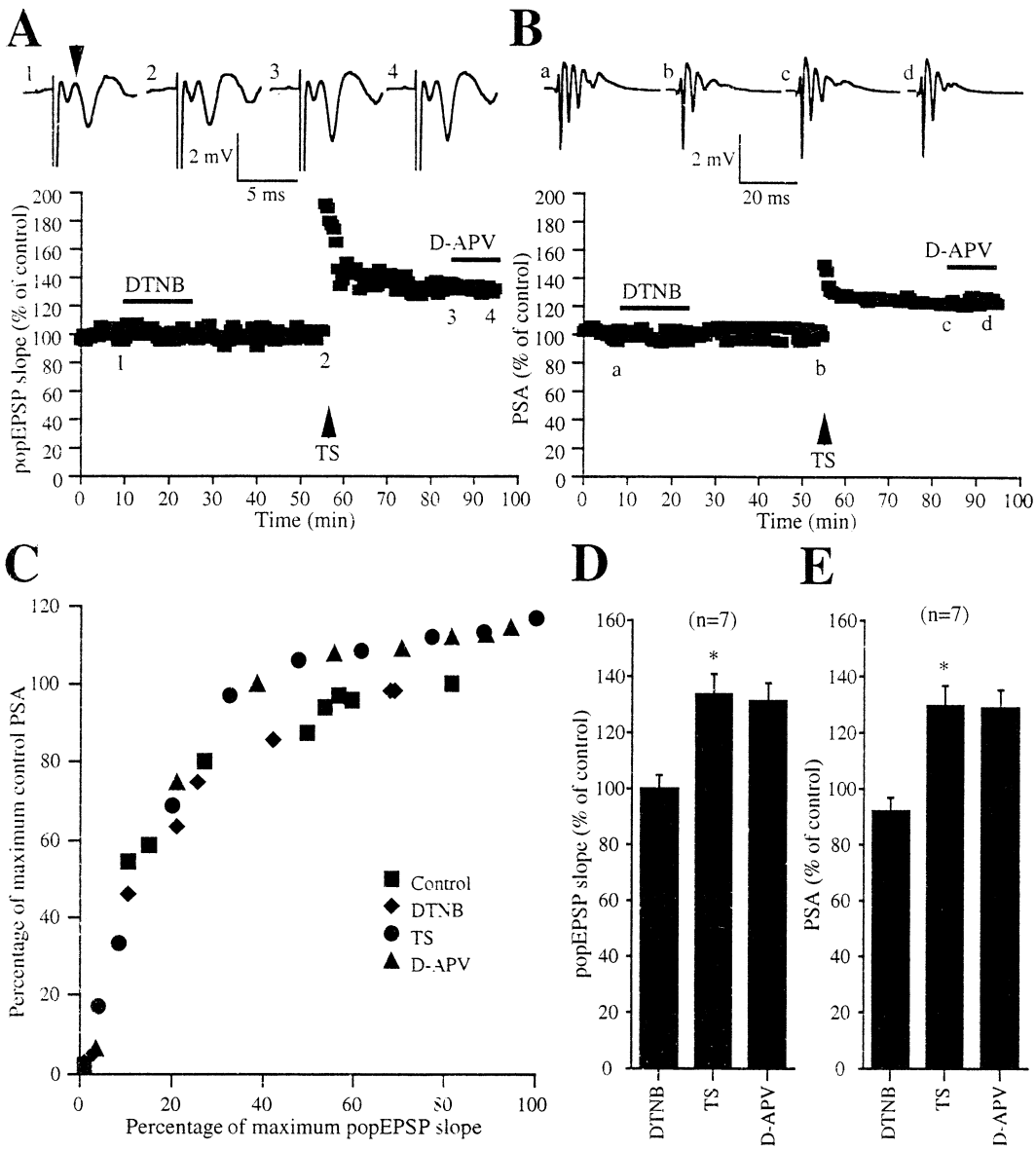

Fig. 1. Example of the effect of DTNB treatment on TS-induced synaptic and network plasticity. (A) Time course of the field EPSP slopes measured every 30 s; 15 min application of DTNB did not modify the synaptic responses. Subsequent TS (arrowhead) potentiated the synaptic responses whereas 10 min D-APV treatment made no further changes. (B) Same as A, for the population spike amplitude. Drugs and conditioning stimulus had similar effects as in A. Above the graphs are shown averages of five consecutive popEPSPs and PSs measured before (1 and a, respectively) and after DTNB (2b), TS (3c) and D-APV (4d). The times at which these traces were recorded are indicated on the graphs. The start of the EPSP is indicated by an arrowhead in A1. Note that DTNB and D-APV had identical effects on the PSAs (compare b and d traces). (C) E/S curves obtained by plotting the PSAs as a function of the popEPSPSs before (■) and after DTNB (◆), TS (●) and D-APV (▲). Note the left shift of the curve after TS indicating E/S potentiation. DTNB and D-APV had no effect on E/S dissociation when compared to control and TS curves respectively. (D–E) Summary of experiments in which synaptic responses and population spikes amplitudes were 100 ± 4 and $92 \pm 16\%$ of control after DTNB, 134 ± 9 and $130 \pm 7\%$ of control after TS and 131 ± 11 and $129 \pm 12\%$ of control after D-APV, respectively. As in the Fig. 2, * $P < 0.0001$ relates to the value prior to the last chemical or electrical conditioning stimulus.

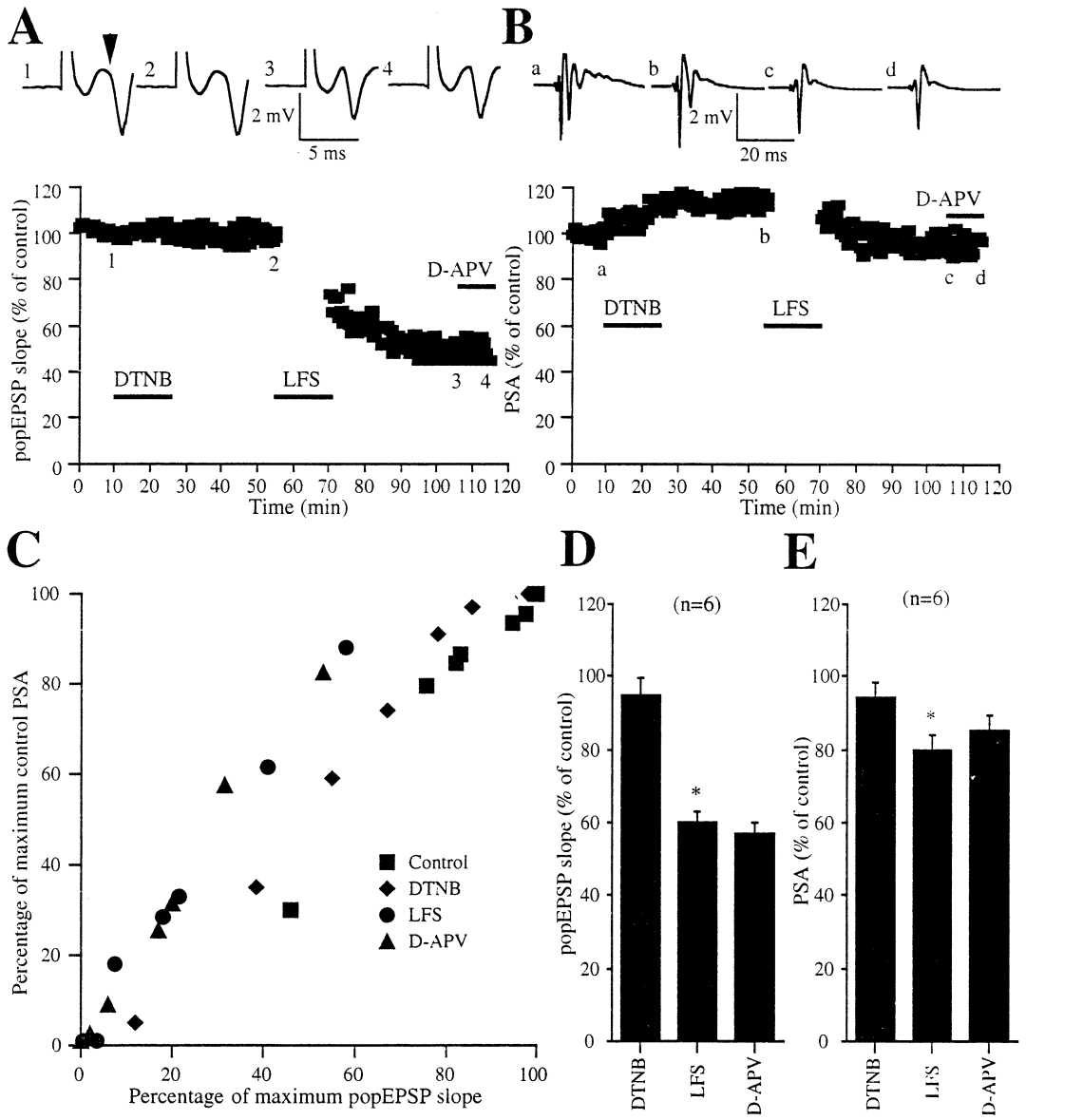

Fig. 2. Example of the effect of DTNB treatment on LFS-induced synaptic and network plasticity. (A) DTNB did not modify the synaptic responses, they were depressed by LFS and D-APV treatment made no further changes. (B) Same as A for the PSA. Note that D-APV induced a slight potentiation of the PSA. (C) E/S curves obtained before (■) and after DTNB (◆), LFS (●) and D-APV (▲). Note the left shift of the curve after DTNB. LFS increased this E/S potentiation. (D–E) Summary of experiments in which synaptic responses and population spikes amplitudes were 95 ± 7 and $94 \pm 12\%$ of control after DTNB, 60 ± 5 and $80 \pm 8\%$ of control after LFS and 57 ± 9 and $85 \pm 10\%$ of control after D-APV, respectively.

3.2. DTNB reduces evoked epileptiform activity and does not affect EPSP/spike coupling

In lesioned slices, DTNB treatment did not affect either the population EPSP slope or the first PSA, 97 ± 5 and $91 \pm 15\%$ of control, respectively ($P > 0.05$, $n = 13$). However, the second, third and fourth PSs were considerably decreased to 69 ± 6 ($P < 0.0001$), 29 ± 7 ($P < 0.0001$) and 0% ($P < 0.0001$) of control, respectively ($n = 13$). In agreement with previous reports [29], DTNB nearly abolished the late part of the evoked epileptiform discharge. DTNB did not produce E/S dissociation; $I_{ES} = 0 \pm 9\%$, indicating that no shift of the E/S curves occurred ($P > 0.05$, $n = 13$).

3.3. DTNB treatment does not prevent the induction of long-term potentiation

In a first series of experiments ($n = 7$), we tested whether LTP could be induced following DTNB treatment. An example of stable TS-induced LTP after application of DTNB is shown in Fig. 1. Population EPSP slopes and PSAs were 134 ± 9 and $130 \pm 7\%$ of control, respectively ($P < 0.0001$). In all experiments, TS resulted in E/S potentiation, $I_{ES} = 25 \pm 9\%$ ($P < 0.001$). These values were not statistically different from those previously reported in the absence of DTNB treatment using the same preparation and the same experimental protocol [8–10,12]. Application of D-APV at the end of the experiment had no further effect on either population EPSP slopes or PSAs, 131 ± 11 and $129 \pm 12\%$ of control, respectively ($P > 0.05$). No further E/S dissociation was seen.

3.4. DTNB treatment does not prevent the induction of long-term depression

In the second series of experiments ($n = 6$), we tested whether LTD could be induced following DTNB treatment. An example of stable LFS-induced LTD after application of DTNB is shown in Fig. 2. Population EPSP slopes and PSAs were 60 ± 5 and $80 \pm 8\%$ of control, respectively ($P < 0.0001$). In all experiments, LFS resulted in E/S potentiation, $I_{ES} = 19 \pm 5\%$ ($P < 0.001$). These values were not statistically different from those measured in the absence of DTNB treatment [12]. Application of D-APV at the end of the experiment had no further effect on either population EPSP slopes or PSAs, 57 ± 9 and $85 \pm 10\%$ of control, respectively ($P > 0.05$). No further E/S dissociation was seen.

4. Discussion

Under control conditions, excitatory synaptic transmission in the hippocampus is mediated primarily by AMPA receptors. LTP and LTD of synaptic responses, which are believed to underlie memory and learning processes, are mainly expressed by AMPA receptors but depend upon activation of NMDA receptors [5]. In pathological conditions such as epilepsy [4,23,30] or ischaemia [15], NMDA receptors participate in synaptic transmission. Furthermore, in human tissue and various experimental chronic models of TLE, epileptiform discharges are usually triggered by the activation of NMDA receptors [3]. However, LTP and LTD of AMPA receptor-mediated responses occur in epileptic tissue and are also dependent on the activation of NMDA receptors [8,12]. As such, drugs that decrease NMDA receptor-mediated synaptic responses could be used to attenuate epileptiform discharges thus providing an alternative to present treatments which focus on boosting inhibition. However, a blockade of NMDA receptors is not desirable since it would prevent synaptic plasticity, i.e. memory and learning processes. Alternatively, one may try to manipulate the multiple regulatory sites on NMDA receptors which fine tune their activity. These regulatory sites can be affected by various compounds such as kinases and phosphatases (intracellularly); glycine, polyamines, histamine, Mg and Zn ions (allosteric sites); as well as pH and redox potential variations [2,7,22]. In this study, attention was focused on redox potential modulations as potential new candidates for the treatment of epilepsies.

Four results support this working hypothesis. Firstly, oxidizing drugs have been reported to be neuroprotective in a model of hypoxic/ischaemic

brain injury [20]. Secondly, it has been suggested that ischaemic insults are associated with a redox modification [17]. Thirdly, DTNB decreases by 50% NMDA receptor-mediated responses [17,18] without affecting AMPA, presynaptic metabotropic glutamate, $GABA_A$ and $GABA_B$ receptor-mediated responses, in CA1 pyramidal cells in non lesioned tissue [13]. Finally, DTNB does not affect synaptic plasticity of AMPA receptor-mediated responses but impairs synaptic plasticity of pharmacologically isolated NMDA receptor-mediated responses [13].

DTNB was as efficient as D-APV in blocking evoked epileptiform activity recorded in the pyramidal cell layer of the CA1 area of the KA lesioned rat hippocampus since D-APV applied at the end of the experiments did not further reduce epileptiform discharges in agreement with recent studies [19,29]. This blockade of epileptiform activity occurred via a 50% decrease of pharmacologically isolated NMDA receptor-mediated responses [19]. In the present study, we report that in the kainic rat as in control tissue, DTNB treatment does not prevent the induction of LTP and LTD of AMPA receptor-mediated synaptic responses. Noteworthy is the absence of effect of DTNB on E/S coupling in contrast to D-APV: in lesioned as well as in control tissues, D-APV treatment produces E/S potentiation, i.e. an increase of the probability of the neurons to discharge in synchrony [11,12]. As such D-APV, despite the blockade of the late part of the burst, modifies the behaviour of the network toward more excitability. DTNB treatment had no such effect. However, subsequent TS or LFS readily produced E/S potentiation as previously described in normal [11] and epileptic tissue [10,12]. The absence of effect of DTNB on E/S coupling may be due to the fact that the 50% decrease of NMDA responses is not sufficient to modify the threshold of E/S coupling. It must be noted that DTNB produced a slight E/S potentiation in three out of 13 cases as shown in Fig. 2.

In conclusion, we suggest that oxidizing agents offer a promising alternative to the classical boost of inhibition for treating TLE. Redox agents exist endogenously [16] and oxidizing drugs have been used successfully in animals for treating infarct [20]. It remains to be determined whether DTNB would have the same effect in vivo than in vitro, i.e. decrease of epileptiform discharges and no effect on synaptic plasticity. The possible deleterious effect of the drug also needs to be investigated.

Acknowledgements

The study was supported by INSERM and DRET. O. Quesada was funded by the CoNaCyT (Mexico). We would like to thank SANDOZ for the gift of D-APV.

References

[1] Andreasen, M., Lambert, J. and Jensen, M., Effects of the new non-*N*-methyl-D-aspartate antagonists on synaptic transmission in the in vitro rat hippocampus, *J. Physiol. (Lond.)*, 414 (1989) 317–336. (Abstract)

[2] Aniksztejn, L., Otani, S. and Ben-Ari, Y., Quisqualate metabotropic receptors modulate NMDA currents and facilitate induction of long-term potentiation through protein kinase C, *Eur. J. Neurosci.*, 4 (1992) 500–505.

[3] Ashwood, T.J. and Wheal, H.V., The expression of *N*-methyl-D-aspartate-receptor mediated component during epileptiform synaptic activity in the hippocampus, *Br. J. Pharmacol.*, 91 (1987) 815–822.

[4] Avoli, M. and Olivier, A., Bursting in human epileptogenic neocortex is depressed by an *N*-methyl-D-aspartate antagonist, *Neurosci. Lett.*, 76 (1987) 249–254.

[5] Bear, M.F. and Malenka, R.C., Synaptic plasticity, LTP and LTD, *Curr. Opin. Neurobiol.*, 4 (1994) 389–399.

[6] Ben-Ari, Y., Limbic seizure and brain damage produced by kainic acid: mechanisms and relevance to human temporal lobe epilepsy, *Neuroscience*, 14 (1985) 375–403.

[7] Ben-Ari, Y., Aniksztejn, L. and Bregestovski, P., Protein kinase C modulation of NMDA currents: An important link for LTP induction, *Trends Neurosci.*, 15 (1992) 333–339.

[8] Bernard, C. and Wheal, H.V., Simultaneous expression of long-term depression of NMDA and long-term potentiation of AMPA receptor-mediated synaptic responses in the CA1 area of the kainic acid-lesioned hippocampus, *Eur. J. Neurosci.*, 7 (1995) 1651–1655.

[9] Bernard, C. and Wheal, H.V., Plasticity of AMPA and NMDA receptor-mediated epileptiform activity in a chronic model of temporal lobe epilepsy, *Epilepsy Res.*, 21 (1995) 95–107.

[10] Bernard, C. and Wheal, H.V., Simultaneous expression of EPSP/spike potentiation and EPSP/spike depression in the hippocampus, *Neuroscience*, 67 (1995) 73–82.

[11] Bernard, C. and Wheal, H.V., Expression of EPSP/spike potentiation following low frequency and tetanic stimulation in the CA1 area of the rat hippocampus, *J. Neurosci.*, 15 (1995) 6542–6551.

[12] Bernard, C. and Wheal, H.V., A role for synaptic and network plasticity in controlling epileptiform activity in CA1 in the kainic acid-lesioned rat hippocampus in vitro, *J. Physiol. (Lond.)*, 495 (1996) 127–142.

[13] Bernard, C., Hirsch, J.C., Khazipov, R., Ben-Ari, Y. and Gozlan, H, Redox modulation of synaptic responses and plasticity in rat CA1 hippocampal neurons, *Exp. Brain Res.*, in press (1996).

[14] Cornish, S. and Wheal, H.V., Long-term loss of paired pulse inhibition in the kainic acid-lesioned hippocampus of the rat, *Neuroscience*, 28 (1989) 563–571.

[15] Crepel, V., Hammond, C., Krnjevic, K., Chinestra, P. and Ben-Ari, Y., Anoxia-induced LTP of isolated NMDA receptor-mediated synaptic responses, *J. Neurophysiol.*, 69 (1993) 1774–1778.

[16] Gozlan, H. and Ben-Ari, Y., NMDA receptor redox sites: are they targets for selective neuronal protection? *Trends Pharmacol. Sci.*, 16 (1995) 368–374.

[17] Gozlan, H., Diabira, D., Chinestra, P. and Ben-Ari, Y., Anoxic LTP is mediated by the redox modulatory site of the NMDA receptor, *J. Neurophysiol.*, 72 (1994) 3017–3022.

[18] Gozlan, H., Khazipov, R., Diabira, D. and Ben-Ari, Y., In CA1 hippocampal neurons, the redox state of NMDA receptors determines LTP expressed by NMDA but not AMPA receptors, *J. Neurophysiol.*, 73 (1995) 2612–2617.

[19] Hirsch, J.C., Quesada, O., Esclapez, H., Gozlan, H., Ben-Ari, Y. and Bernard, C., Enhanced NMDAR-dependent epileptiform activity is controlled by oxidizing agents in a chronic model of temporal lobe epilepsy, *J. Neurophysiol*, in press (1996).

[20] Jensen, F., Gardner, G., Williams, P., Gallop, P., Aizenman, E. and Rosenberg, P., The putative essential nutrient pyrroloquinoline quinone is neuroprotective in a rodent model of hypoxic/ischemic brain injury, *Neuroscience*, 62 (1994) 399–406.

[21] Lancaster, B. and Wheal, H.V., A comparative histological and electrophysiological study of some neurotoxins in the rat hippocampus, *J. Comp. Neurol.*, 211 (1982) 105–114.

[22] McBain, C.J. and Mayer, M.L., *N*-methyl-D-aspartic acid receptor structure and function, *Physiol. Rev.*, 74 (1994) 723–760.

[23] Mody, I. and Heinemann, U., NMDA receptors of dentate gyrus granule cells participate in synaptic transmission following kindling, *Nature*, 326 (1987) 701–704.

[24] Nadler, J.V., Kainic acid as a tool for the study of temporal lobe epilepsy, *Life Sci.*, 29 (1981) 2031–2042.

[25] Nadler, J.V., Perry, B. and Cotman, C., Selective reinnervation of hippocampal area CA1 and fascia dentate after destruction of CA3-CA4 afferents with kainic acid, *Brain Res.*, 182 (1980) 1–9.

[26] Nadler, J.V., Perry, B., Gentry, C. and Cotman, C., Loss and reacquisition of hippocampal synapses after selective destruction of hippocampal synapses after selective destruction of CA3-CA4 afferents with kainic acid, *Brain Res.*, 191 (1980) 387–403.

[27] Perez, Y., Morin, F., Jutras, I., Beaulieu, C. and Lacaille, J.-C., CA1 pyramidal cells in hyperexcitable slices of kainate-treated rats show sprouting of local axon collaterals, *Eur. J. Neurosci.*, 8 (1996) 736–748.

[28] Phelps, S., Mitchell, J. and Wheal, H.V., Changes to synaptic ultrastructure in field CA1 of the rat hippocampus following intracerebroventricular injection of kainic acid, *Neuroscience*, 40 (1991) 687–699.

[29] Quesada, O., Hirsch, J., Ben-Ari, Y. and Bernard, C., Redox sites of NMDA receptors can modulate epileptiform activity in hippocampal slices from kainic acid treated rats, *Neurosci. Lett.*, 212 (1996) 171–174.

[30] Turner, D.A. and Wheal, H.V., Excitatory synaptic potentials in kainic acid-denervated rat CA1 pyramidal neurons, *J. Neurosci.*, 11 (1991) 2786–2794.

[31] Wheal, H.V., Function of synapses in the CA1 region of the hippocampus: their contribution to the generation or control of epileptiform activity, *Comp. Biochem. Physiol. [C]*, 93A (1989) 211–220.