Available online at www.sciencedirect.com
SCIENCE DIRECT®
Neuroscience Letters 390 (2005) 15–20
Neuroscience Letters
www.elsevier.com/locate/neulet

Voltage-gated calcium channels are not involved in generation and propagation of spreading depression (SD) in the brainstem of immature rats

Frank Richter[a,*], Alfred Lehmenkühler[b], Hans-Georg Schaible[a]

[a] *Institute of Physiology I – Neurophysiology, Friedrich Schiller University Jena, Teichgraben 8, D-07740 Jena, Germany*
[b] *St. Vincent-Hospital Düsseldorf, Center for Pain Therapy, Schloßstraße 85, D-40477 Düsseldorf, Germany*

Received 16 June 2005; received in revised form 18 July 2005; accepted 26 July 2005

Abstract

Spreading depression (SD) can be elicited in the brainstem of rats younger than 13 days when excitability is enhanced by acetate superfusion [F. Richter, S. Rupprecht, A. Lehmenkühler, H.-G. Schaible, Spreading depression can be elicited in brain stem in immature but not adult rats, J. Neurophysiol. 90 (2003) 2163–2170]. To investigate whether voltage-gated calcium channels (VGCCs) modify initiation and propagation of SD in this type of tissue, we applied specific blockers to L-, T-, P/Q-, and N-type VGCCs locally or systemically. SD-related d.c. potentials and concomitant increases in extracellular potassium concentration ($[K^+]_e$) were unaffected by the L- and T-type VGCC blocker flunarizine that was applied either systemically (up to 2 mg/kg body weight) or by superfusion onto the brainstem (40 μM). In addition, local application of the P/Q-type VGCC blocker ω-agatoxin (1 μM) or of the N-type VGCC blocker ω-conotoxin (1 μM) to the brainstem surface did not influence SD. The results indicate that VGCCs do not modify the generation or propagation of SDs in the brainstem of the immature rat. Blockade of N-type VGCCs disturbed the normal breathing rhythm. Application of alpha-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid (AMPA) (250–1000 μM) that elicited SD in the immature cortex, failed to elicit SD in the immature brainstem. In summary, it is likely that K^+ initiates and propagates brainstem SDs.
© 2005 Elsevier Ireland Ltd. All rights reserved.

Keywords: SD-related d.c. shift; Agatoxin; Conotoxin; Flunarizine

Spreading depression (SD) in the cerebral cortex [12,13] is a depolarization wave characterized by a transient negative d.c. potential, propagating slowly from a focus across the hemisphere. It is accompanied by a transient release of potassium into the extracellular space and an influx of water, sodium, calcium and chloride into cells [2,21]. For a long time, SD in the cerebral cortex is thought to be the neuronal mechanism involved in the migraine aura [10,16,18] and a mechanism that causes expansion of the ischemic penumbra zone after stroke (cf. [27]).

Voltage-gated calcium channels (VGCCs) play a modulating role in cortical and hippocampal SD. Leaner and tottering mice with mutations of P/Q-calcium channels showed increased resistance to cortical SD and a lack of self-regenerated repetitive cortical SD waves [1]. The blockade of N-, P/Q-, or L-type calcium channels by specific blockers inhibited repetitive cortical SD in adult rats, but did not prevent prick-induced non-repetitive SD [17]. Conversely, the blockade of T-type calcium channels by amiloride induced spontaneously occurring repetitive SD waves in human cortical slices [4]. In hippocampal organotypic cultures, uncoupling of glial gap junctions by heptanol blocked both calcium waves and SD waves [8]. Blockade of P/Q-type calcium channels but of neither L- nor N-type calcium channels prevented SDs in hippocampal organotypic cultures [9].

Recently, we have shown that SDs can be elicited in the immature rat brainstem [19] when 75% of the extracellular chloride content was substituted by an equimolar amount of acetate as it has been demonstrated for the turtle cerebellum

* Corresponding author. Tel.: +49 3641 938811; fax: +49 3641 938812.
E-mail address: frank.richter@mti.uni-jena.de (F. Richter).

0304-3940/$ – see front matter © 2005 Elsevier Ireland Ltd. All rights reserved.
doi:10.1016/j.neulet.2005.07.046

[11,14]. The mechanisms underlying generation and propagation of brainstem SDs have not been described. Since VGCCs are involved in transmitter release and control of excitability, blockade of VGCCs should interfere with brainstem SDs if the same mechanisms are involved as in cortical and hippocampal SDs (see also [22]). The elevation of extracellular $[K^+]$ due to neuronal release that was proposed originally by Grafstein [6] could either by itself or together with glutamate [23] initiate or propagate SD.

To test whether VGCCs are involved in brainstem SDs, we recorded SD-related d.c. deflections in the brainstem of immature rats before and after the blockade of VGCCs in the brainstem. For comparison, we also recorded SD in the cerebral cortex. The experiments were approved by the local animal protection committee and the regional government of Thüringen (reg. 02-10/01). The preparation was previously described in detail [19]. Briefly, in spontaneously breathing, urethane-anesthetized (1.5 g/kg, i.p., Sigma–Aldrich GmbH, Seelze, Germany) Wistar rat pups aged 11–12 days a trephination (diameter 2–3 mm) was made over the occipito-temporal cortex, and the underlying dura was slitted. The brainstem was exposed from the occipital bone to the first cervical vertebra by incising the neck muscles, the atlanto-occipital ligament and by removing the underlying dura mater. A feedback-controlled heating pad maintained body temperature at 37 °C, and a mechanograph monitored thorax movements.

To record d.c. potentials at different vertically and horizontally separated sites in the brainstem, an array was used that consisted of three microelectrodes (horizontal tip separation of 400 μm, vertical tip separation of 400 μm) filled with 150 mM NaCl. It was lowered into the brainstem and reached a maximum depth of 1200 μm in a region close to the caudal trigeminal nucleus. For simultaneous registration of d.c. shifts and of changes in extracellular potassium concentration ($[K^+]_e$), a double-barreled potassium-selective microelectrode filled with Corning K^+ exchanger 477317 (W.P.I., Berlin, Germany) was used. To observe SD propagation, one d.c. microelectrode filled with 150 mM NaCl was glued with 400 μm horizontal tip separation to this electrode. SD-related d.c. shifts in the cerebral cortex were recorded at a depth of 1200 μm using a single barreled microelectrode filled with 150 mM NaCl. The reference electrode for DC-recording was placed onto the nasal bone. For further techniques see [19].

Exposed brain areas were kept moist with regular artificial cerebrospinal fluid (ACSF (in mM): NaCl 138.4, KCl 3.0, $CaCl_2$ 1.3, $MgCl_2$ 0.5, NaH_2PO_4 0.5, urea 2.2, glucose 3.4, warmed to 37 °C and equilibrated with 5% CO_2 in O_2). After ACSF superfusion for 1 h, small KCl crystals weighing 0.1–0.5 mg were placed onto the cortex and onto the brainstem surface at a distance of about 1 mm to the most superficial electrode. Prior to KCl application, superfusion was stopped and the surface of the cortex and the brainstem was carefully dried with a thread of cotton wool. After KCl application for about 1 min, superfusion was continued for 60 min with ACSF in which 100 mM of the chloride (75%) was exchanged by 100 mM acetate. After stopping the superfusion, another KCl crystal was applied. Superfusion was continued after the first SD-related d.c. shift or when a sustained negative d.c. shift occurred that was associated with an impairment of breathing. At the end of the experiment, the animal was sacrificed by an overdose of urethane.

In seven rat pups aged 11–12 days, the non-selective L- and T-type calcium channel blocker flunarizine (flunarizine hydrochloride, Sigma, Germany) was administered intraperitoneally at concentrations of 1–2 mg/kg body weight (according to Shimazawa [20]), when KCl application had elicited SD in the brainstem. In four of these rats in addition, flunarizine at 40 μM diluted in ACSF was applied topically to the surface of the cortex and the brainstem.

To check whether acetate-ACSF interferes with SD repetition or with the effects of the VGCC blockers, we superfused the cerebral cortex of three adult rats with acetate-ACSF and tested whether KCl still elicited repetitive SD after 1–2 h of superfusion. Subsequently, we applied the VGCC blockers (1 μM ω-agatoxin, 1 μM ω-conotoxin, or 40 μM flunarizine) topically to the exposed cortical surface, and we monitored the propagation of prick-induced SD and the propagation and repetition rates of KCl-induced SD in the untreated and treated cortical areas, respectively. For further methodological details see [17].

In a group of five 11-day-old rats, we tested whether the topical application of a solution containing ω-agatoxin IVA at 1 μM (P/Q-type calcium channel blocker, dissolved in 165 mM NaCl, pH 7.4; Bachem, Heidelberg, Germany) or of ω-conotoxin GVIA at 1 μM (N-type calcium channel blocker, dissolved in 165 mM NaCl, pH 7.4; Bachem, Heidelberg, Germany) blocks the propagation of SD in the brainstem. The same doses prevented repetitive SDs in adult cerebral cortex [17] and influenced single neuron activity in the adult rat brainstem [3]. About 50 μl of the ω-agatoxin IVA solution was applied to the dried surface of the brainstem with a micropipette and stayed there for 20 min. Then, it was quickly removed by a cotton thread, and within 30 s a KCl crystal was placed to the brainstem. The KCl crystal was washed away with acetate-ACSF when an SD wave had moved across the electrode area. Then the brainstem was dried with a cotton thread and the same blocker was applied for another 20 min. The procedure was repeated a third time, so that each blocker was applied for 60 min, only interrupted by two intervals of about 5 min each. After thoroughly washing the brainstem with acetate-ACSF the same protocol was repeated with topical application of ω-conotoxin GVIA.

To further test whether glutamatergic mechanisms are involved in the generation of SDs in the immature brainstem, we applied the specific AMPA receptor agonist alpha-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid (AMPA: 250, 500 and 1000 μM; Sigma, Seelze, Germany) topically to the acetate-ACSF conditioned brainstem in 6 rats aged 11–12 days. In addition, we tested whether the same concentrations of AMPA evoke cortical SDs.

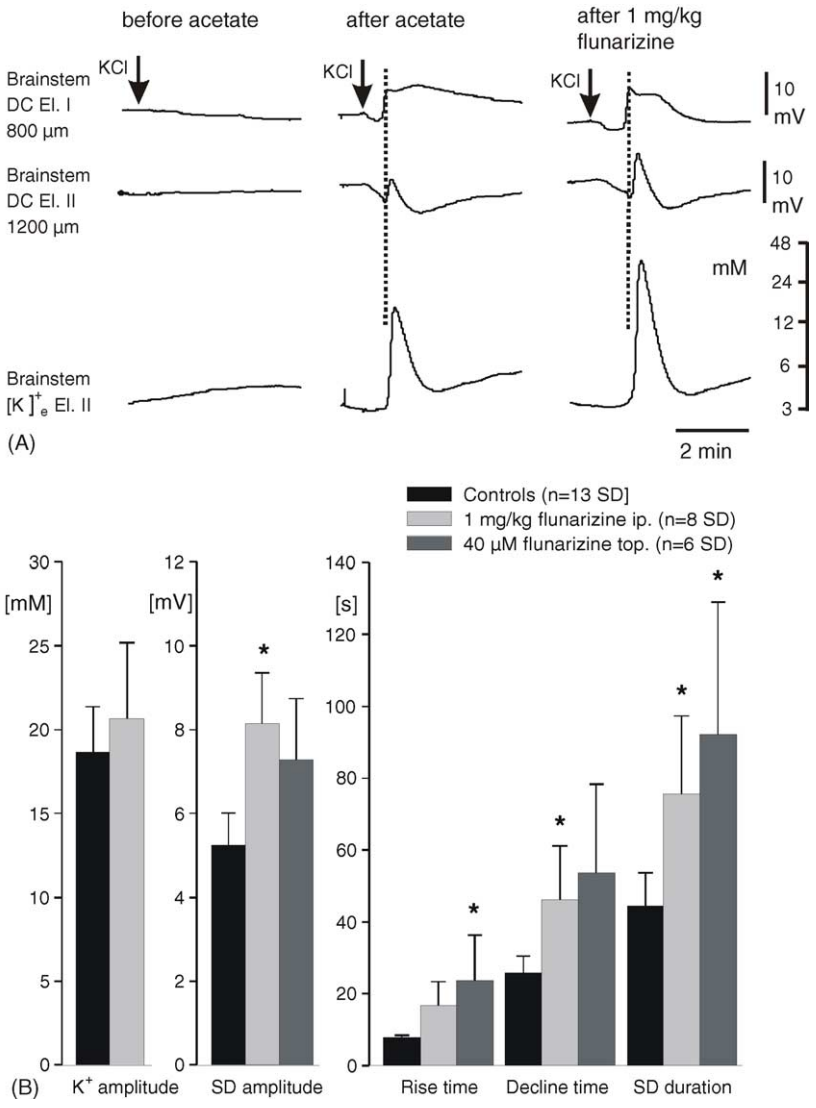

Fig. 1. (A) Failure of systemic flunarizine to influence SD generation in the immature brainstem. Simultaneous recordings of SD-related d.c. deflections and concomitant changes in extracellular potassium concentration at depths of 800 and 1200 μm in the brainstem of an 11-day-old rat. The lateral spacing of the electrode tips was 400 μm. Panels from left to right: prior to acetate-ACSF, KCl failed to elicit SD in the brainstem. After acetate, KCl elicited single SD-related d.c. shifts in the brainstem that were accompanied by an elevation in $[K^+]_e$. A similar brainstem SD was observed after systemic flunarizine. The thin dotted lines accent the delayed peak times at the second brainstem d.c. electrode (middle trace), indicating the propagation of the SD wave. (B) Comparison of SD parameters amplitude (measured as amplitude of the SD-related d.c. shift), rise time to peak, decline time and duration after acetate, after 1 mg/kg flunarizine i.p. and after application of 40 μM flunarizine to the surface of the brainstem in the course of the experiments. The bars show mean values ± S.E.M. An asterisk marks statistically significant differences to controls ($p < 0.05$).

Data were recorded on PC by using custom signal acquisition programs. The SDs were analyzed regarding occurrence, amplitude, and shape. Direct current (d.c.) shifts were classified as SD if $[K^+]_e$ elevations amounted at least 12 mM with concomitant d.c. shifts of about 3 mV, and if peaks showed a time difference at the horizontally separated electrodes in the brainstem indicating propagation. Statistical comparisons between values before and after treatment were made by one-way ANOVA with Tukey's post-test. Significance was accepted at $p < 0.05$.

No SD-related d.c. shifts were elicited by a KCl crystal in the cortex or in the brainstem of rat pups prior to acetate superfusion confirming our previous study [19]. A superfusion with acetate-ACSF for 90–120 min was required to condition the immature brain tissues for SD. We never observed a migration of the SD from the brainstem to the cerebral cortex.

After administration of flunarizine systemically, we were still able to elicit, with a KCl crystal, the typical non-repetitive SD-related d.c. deflections in the brainstem (Fig. 1A). Flunarizine did not stop SD generation and propagation in the brainstem (14 attempts elicited 14 SD), but significantly prolonged the SD-related d.c. shift at this site (Fig. 1B). No effect of flunarizine was obtained in the cortex of these rats. Neither

the intraperitoneal nor the topical application of flunarizine interfered with the normal breathing rhythm of the rat pups. In adult rat neocortex, the same doses of flunarizine inhibited repetitive SDs after KCl application (data not shown).

Both the P/Q-type blocker ω-agatoxin IVA and the N-type VGCC blocker ω-conotoxin GVIA inhibited the repetition rate of KCl-induced SD in the cerebral cortex of adult rats [17]. In the present study, we confirmed these effects and showed that acetate-ACSF did not influence the inhibiting effects of both VGCC blockers on repetitive SDs in the cerebral cortex of adult rats (data not shown). Topical application of ω-agatoxin IVA to the surface of the brainstem of rat pups did not prevent SD-related d.c. shifts (15 attempts elicited 13 SD). Neither a single application of ω-agatoxin IVA for 20 min nor a triple application for 20 min each inhibited SD elicitation and propagation (Fig. 2A). After ω-agatoxin IVA application brainstem SD-related d.c. shifts were slightly smaller in amplitude, and their duration lasted significantly longer than during acetate-ACSF controls (Fig. 2B). Breathing frequency was unchanged by ω-agatoxin IVA. Subsequent application of ω-conotoxin GVIA (Fig. 2A) did not influence the occurrence of SD-related d.c. shifts either (11 attempts elicited 10 SD), but slightly decreased their amplitudes and increased their rise times (Fig. 2B). In three out of five rat pups, the second application of ω-conotoxin GVIA caused a progressive reduction in breathing frequency after about 10 min, and a final respiratory arrest with asphyxia occurred after 15–20 min.

The application of AMPA was barely effective to elicit SD in the immature brainstem. In all rat pups, a KCl crystal elicited SD in the brainstem after conditioning with acetate-ACSF, but AMPA elicited only at a concentration of 1000 μM in one of the six animals a d.c. deflection that fulfilled our criteria of SD (Fig. 3). In the cerebral cortex of the same animals, a concentration of 250 μM AMPA was sufficient to elicit a single SD wave (four attempts elicited three SD), whereas 500 or 1000 μM AMPA evoked long-lasting and slowly recurring depolarizations in the cerebral cortex that did not conform to typical SD (500 μM: four SDs in five applications; 1000 μM: one SD in four applications).

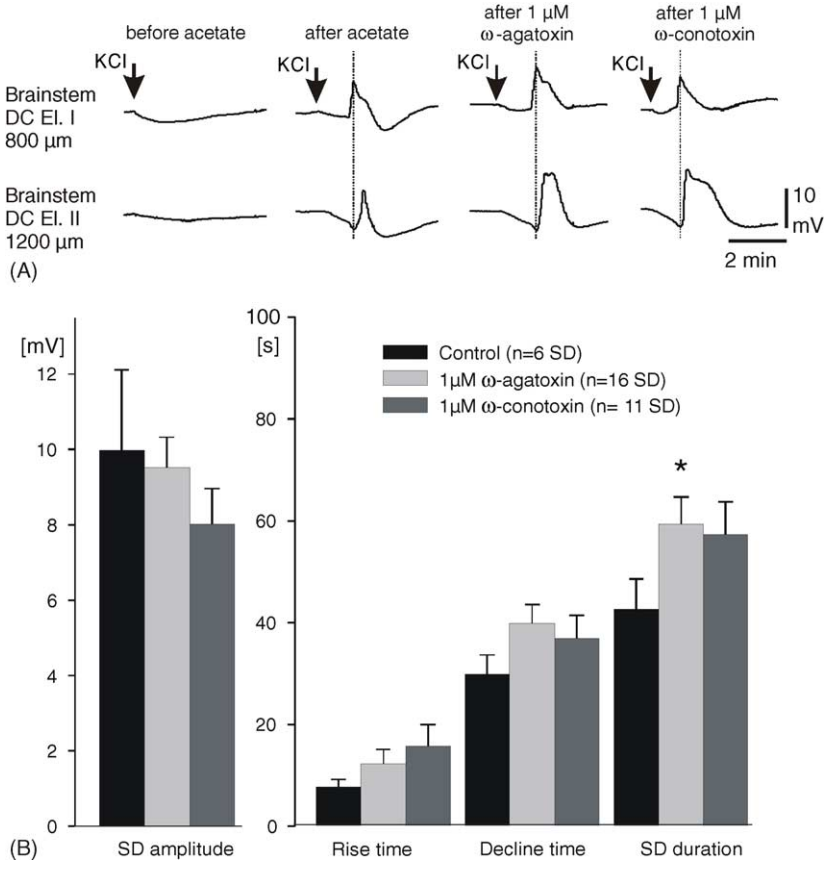

Fig. 2. (A) SD propagation in the brainstem at two different depths (800 and 1200 μm) with a separation of the electrode tips by 800 μm after topical application of specific P/Q-type and N-type VGCC blockers to the brainstem surface. The recordings were made subsequently in an 11-day-old rat. Panels from left to right: prior to acetate-ACSF KCl failed to elicit SD in the brainstem. KCl-evoked SD after acetate-ACSF in the brainstem. The application of ω-agatoxin for three times per 20 min did not change elicitability of the brainstem SD. First application of ω-conotoxin for 20 min did not interfere with the KCl-induced SD in the brainstem. Scale bars refer to all traces. (B) Comparison of SD parameters amplitude (measured as amplitude of the SD-related d.c. shift), rise time to peak, decline time and duration in controls after acetate, after topical application of ω-agatoxin and after topical application of ω-conotoxin to the surface of the brainstem in the course of the experiments. The bars show mean values ± S.E.M. An asterisk marks statistically significant differences to controls ($p < 0.05$).

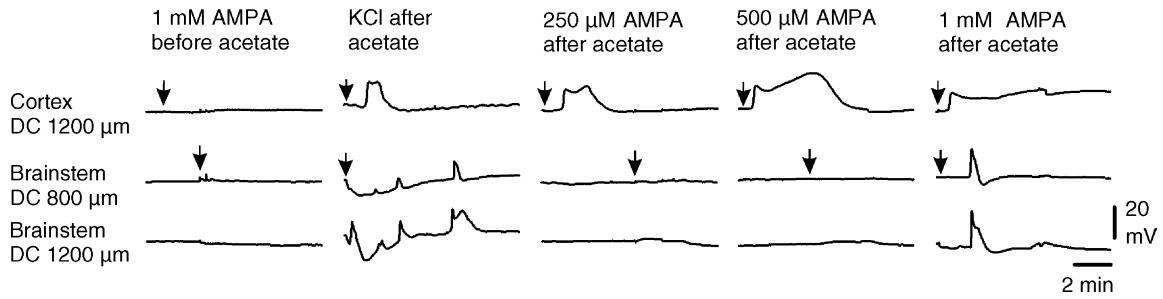

Fig. 3. The application of AMPA at different concentrations elicited SD in the cerebral cortex of 11-day-old rats, but not in the brainstem. Panels from left to right: 1000 μM AMPA (arrows) prior to acetate conditioning was ineffective to elicit any d.c. deflections either in the cortex or in the brainstem. After 60 min of acetate conditioning KCl (arrows) elicited SDs both in the cortex and in the brainstem. AMPA (250 μM) (arrows) after acetate elicited SD in the cerebral cortex but not in the brainstem. After application of 500 μM AMPA (arrows) following acetate conditioning the SD wave in the cortex was prolonged and returned only slowly to baseline, no AMPA-related d.c. deflections in the brainstem were seen. Application of 1000 μM AMPA (arrows) after acetate conditioning caused a long-lasting negative d.c. deflection in the cortex, similar to a terminal negativation. In the brainstem of this particular animal, however, this concentration of AMPA elicited one SD.

The present data show that blockade of L-, T-, P/Q-, and N-type VGCCs does not interfere with the generation and propagation of SD in the immature brainstem. Thus SDs in the brainstem significantly differ from that in neocortex and hippocampus. Application of the L- and T-type VGCC blocker flunarizine to the immature brainstem or cortex did not prevent SDs. This is different from adult animals where flunarizine raises the SD-threshold in the cerebral cortex [24,25] and decreases the repetition rate of KCl-induced SD [20]. The prolonged duration of SD in the immature brainstem after application of flunarizine could be attributed to a non-specific blockade of channels that are involved in the recovery from SD. The lack of effect of blockade of T-type VGCCs on brainstem SD also contrasts with the results in hippocampal slices, where low doses of the T-type channel blockers Ni^{2+} or amiloride induced SD [4,5].

The extracellular diffusion of excitatory amino acids, e.g. glutamate, that mediate propagation and self-ignition of repetitive SD in the cerebral cortex is closely related to their synaptic release [21]. The latter depends on activation of presynaptic VGCCs. In the adult neocortex, topical application of the same doses of P/Q-type and N-type VGCC blockers for 60 min (ω-agatoxin IVA and ω-conotoxin GVIA, respectively) inhibited repetitive SD and prolonged the duration of single CSD. A KCl crystal, however, was still able to elicit a single CSD that traveled across the cortical area in which VGCCs had been blocked, indicating that elevation of $[K^+]_e$ alone is able to generate and propagate single SD [17]. In the immature brainstem, KCl elicited only a single, non-repetitive SD. Therefore, we suggest that VGCCs are not involved in the propagation of KCl-induced SD. Indeed, in the immature brainstem, application of ω-agatoxin IVA and ω-conotoxin GVIA did not prevent generation and propagation of single KCl-elicited SD. In the adult cortex, acetate-ACSF did not reduce the repetition rate of KCl-induced SD and the inhibitory effect of the VGCC blockers. It is unlikely, therefore, that the lack of effect of VGCC blockers on brainstem SD is due to the acetate-ACSF conditioning. The particular mechanism, how the chloride/acetate exchange conditions the brain tissue for SD, is not clear. Besides lowering the chloride content, it might additionally change the pH- and calcium-activity gradient across cellular elements, resulting in an increased neuronal excitability.

The precise localization of different VGCCs in the brainstem (presynaptically to the neuron, at interneurons, etc.) is assumed to be similar to that in other brain tissues [3,26]. P/Q-type and N-type channels are located presynaptically, thereby controlling the release of transmitters such as glutamate. The failure of VGCC blockers to inhibit SD in the immature brain tissue might have several reasons. (i) KCl-induced SD at this age propagate independently from the release of excitatory amino acids, and therefore, VGCCs are not required for SD. (ii) The VGCCs are not functional at this developmental age. The latter is unlikely, because ω-conotoxin GVIA reduced breathing, suggesting that at least N-type channels are present. AMPA, which is known to elicit SD in the adult cerebral cortex [15] and in the chicken retina [7], barely elicited SD in the immature brainstem. At the same age when AMPA in the immature cerebral cortex was able to elicit a typical SD, it failed in the brainstem, though KCl was effective. Therefore, we conclude that glutamatergic mechanisms do not contribute to the generation and propagation of KCl-induced SD in the brainstem. This is in line with our previous findings that blockade of NMDA receptors by MK-801 or ketamine did not abolish KCl-induced SD waves in the immature brainstem [19].

In summary, as shown in our previous report and in the present study, the immature rat brainstem is able to generate SD under suitable conditions. This type of SD resembles in its phenomenology (e.g. amplitudes of transient d.c. shifts and accompanying elevations in $[K^+]_e$, propagation velocity) the SDs in the cerebral cortex. Since VGCC blockade does not modulate SDs in the immature brainstem, in this type of tissue both generation as well as propagation of SD probably obey the classical potassium theory of Grafstein [6], whereas in the cerebral cortex glutamate is additionally necessary [21,22].

Acknowledgements

This work was supported by IZKF Jena (IZKF B378-10102). We thank Mrs. Helga Müller for excellent technical assistance.

References

[1] C. Ayata, M. Shimizu-Sasamata, E.H. Lo, J.L. Noebels, M.A. Moskowitz, Impaired neurotransmitter release and elevated threshold for cortical spreading depression in mice with mutations in the α1A subunit of P/Q type calcium channels, Neuroscience 95 (2000) 639–645.
[2] J. Bureš, O. Burešova, J. Křivanek, The Mechanism and Applications of Leão's Spreading Depression of Electroencephalographic Activity, Academia, Prague, 1974.
[3] A. Ebersberger, S. Portz, W. Meissner, H.-G. Schaible, F. Richter, Effects of N-,P/Q- and L-type calcium channel blockers on nociceptive neurons of the trigeminal nucleus with input from the dura, Cephalalgia 24 (2004) 250–261.
[4] A. Gorji, D. Scheller, H. Straub, F. Tegtmeier, R. Kohling, J.M. Hohling, I. Tuxhorn, A. Ebner, P. Wolf, H. Werner Panneck, F. Oppel, E.J. Speckmann, Spreading depression in human neocortical slices, Brain Res. 906 (2001) 74–83.
[5] A. Gorji, D. Scheller, F. Tegtmeier, R. Kohling, H. Straub, E.J. Speckmann, $NiCl_2$ and amiloride induce spreading depression in guinea pig hippocampal slices, Cephalalgia 20 (2000) 740–747.
[6] B. Grafstein, Mechanism of spreading cortical depression, J. Neurophysiol. 19 (1956) 308–316.
[7] S. Kertesz, G. Kapus, G. Levay, Interactions of allosteric modulators of AMPA/kainate receptors on spreading depression in the chicken retina, Brain Res. 1025 (2004) 123–129.
[8] P.E. Kunkler, R.P. Kraig, Calcium waves precede electrophysiological changes of spreading depression in hippocampal organ cultures, J. Neurosci. 18 (1998) 3416–3425.
[9] P.E. Kunkler, R.P. Kraig, P/Q Ca^{2+} channel blockade stops spreading depression and related pyramidal neuronal Ca^{2+} rise in hippocampal organ culture, Hippocampus 14 (2004) 356–367.
[10] M. Lauritzen, Pathophysiology of the migraine aura. The spreading depression theory, Brain 117 (1994) 199–210.
[11] M. Lauritzen, M.E. Rice, Y.C. Okada, C. Nicholson, Quisqualate, kainate and NMDA can initiate spreading depression in the turtle cerebellum, Brain Res. 475 (1988) 317–327.
[12] A.A.P. Leão, Spreading depression of activity in the cerebral cortex, J. Neurophysiol. 7 (1944) 359–390.
[13] A.A.P. Leão, Further observations on the spreading depression of activity in the cerebral cortex, J. Neurophysiol. 10 (1947) 409–419.
[14] C. Nicholson, J.M. Phillips, C. Tobias, R.P. Kraig, Extracellular potassium, calcium and volume profiles during spreading depression, in: E. Syková, P. Hńik, L. Vyklicky (Eds.), Ion-Selective Microelectrodes and Their Use in Excitable Tissues, Plenum Press, New York, London, 1981, pp. 211–223.
[15] T.P. Obrenovitch, J. Urenjak, E. Zilkha, Intracerebral microdialysis combined with recording of extracellular field potential: a novel method for investigation of depolarizing drugs in vivo, Br. J. Pharmacol. 113 (1994) 1295–1302.
[16] D. Pietrobon, J. Striessnig, Neurobiology of migraine, Nat. Rev. Neurosci. 4 (2003) 386–398.
[17] F. Richter, A. Ebersberger, H.-G. Schaible, Blockade of voltage-gated calcium channels in rat inhibits repetitive cortical spreading depression, Neurosci. Lett. 334 (2002) 123–126.
[18] F. Richter, A. Lehmenkühler, Spreading depression can be restricted to distinct depths of the rat cerebral cortex, Neurosci. Lett. 152 (1993) 65–68.
[19] F. Richter, S. Rupprecht, A. Lehmenkühler, H.-G. Schaible, Spreading depression can be elicited in brain stem in immature but not adult rats, J. Neurophysiol. 90 (2003) 2163–2170.
[20] M. Shimazawa, H. Hara, T. Watano, T. Sukamoto, Effects of Ca^{2+} channel blockers on cortical hypoperfusion and expression of c-Fos-like immunoreactivity after cortical spreading depression in rats, Br. J. Pharmacol. 115 (1995) 1359–1368.
[21] G.G. Somjen, Mechanisms of spreading depression and hypoxic spreading depression-like depolarization, Physiol. Rev. 81 (2001) 1065–1096.
[22] A. van Harreveld, Compounds in brain extracts causing spreading depression of cerebral cortical activity and contraction of crustacean muscle, J. Neurochem. 3 (1959) 300–315.
[23] A. van Harreveld, Two mechanisms for spreading depression in the chicken retina, J. Neurobiol. 9 (1978) 419–431.
[24] A. Wauquier, Is there a common pharmacological link between migraine and epilepsy? Funct. Neurol. 1 (1986) 515–520.
[25] A. Wauquier, D. Ashton, R. Marrannes, The effects of flunarizine in experimental models related to the pathogenesis of migraine, Cephalalgia 5 (Suppl. 2) (1985) 119–123.
[26] R.E. Westenbroek, L. Hoskins, W.A. Catterall, Localization of Ca^{2+} channel subtypes on rat spinal motor neurons, interneurons, and nerve terminals, J. Neurosci. 18 (1998) 6319–6330.
[27] O.W. Witte, H.J. Bidmon, K. Schiene, C. Redecker, G. Hagemann, Functional differentiation of multiple perilesional zones after focal cerebral ischemia, J. Cereb. Blood Flow Metab. 20 (2000) 1149–1165.