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Review

Minireview: pH and synaptic transmission

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ABSTRACT

As a general rule a rise in pH increases neuronal activity, whereas it is dampened by a fall of pH. Neuronal activity per se also challenges pH homeostasis by the increase of metabolic acid equivalents. Moreover, the negative membrane potential of neurons promotes the intracellular accumulation of protons. Synaptic key players such as glutamate receptors or voltage-gated calcium channels show strong pH dependence and effects of pH gradients on synaptic processes are well known. However, the processes and mechanisms that allow controlling the pH in synaptic structures and how these mechanisms contribute to normal synaptic function are only beginning to be resolved.

1. Introduction

The strong acidification of synaptic vesicles by the vacuolar H^+-ATPase, which energizes the neurotransmitter loading of synaptic vesicles [1], is a main reason for the large fluctuations in synaptic pH. Synaptic vesicle exocytosis results in the release of protons into the synaptic cleft as well as in the incorporation of the vacuolar H^+-ATPase into the presynaptic membrane. Thus synaptic transmission causes a relatively short but strong acidification of the synaptic cleft [2–4]. The extracellular acidosis is subsequently followed by a long, yet transient increase in extrasynaptic pH [5]. In the hippocampus this alkaline transient can be detected within milliseconds [6,7] and reaches magnitudes as large as 0.1–0.2 pH units [8]. Mechanisms underlying this rise in pH are not fully understood but most likely presynaptic Ca^{2+}/H^+-ATPase [9,10], extracellular carbonic anhydrases [8], and $GABA_A$-receptor mediated bicarbonate efflux [11] are involved. Increased synaptic/neuronal activity can also cause a prolonged extracellular acidification because of the increased cell metabolism [5,12,13].

Although several studies have successfully monitored neuronal pH shifts in the brain [2,14,15], only very little is known about pH transients in neuronal microdomains because of technical limitations [16,17]. Direct experimental data on pH fluctuations and pH regulation in intracellular synaptic compartments so far have only been obtained for motor endplates because of their significantly larger dimensions compared to central synapses [4,18]. Zhang et al. used the pH-sensitive properties of the yellow fluorescent protein to analyse the presynaptic pH in mouse motor endplates. This study not only supports the importance of presynaptic pH regulators but further provided evidence that the release of vesicles in the peripheral nervous system is accompanied by a transient intracellular acidification. Here, the increase in pH was mainly caused by the activation of plasma membrane Ca^{2+}/H^+-ATPase and was followed by an unexpected, longer lasting alkalinisation is due to the transient incorporation of the vacuolar H^+-ATPase into the presynaptic membrane [4]. Focal injections of BCECF-AM in combination with slice imaging as used for measuring calcium transients in small synaptic compartments with the calcium-sensitive dye Fura [19], genetically encoded pH indicators [18], which also allow ratiometric imaging [20,21], may help to establish adequate and fast pH measurement in small compartments like central pre- and postsynaptic terminals in the future.

Despite these technical limitations the occurrence of rather large, spatially and timely limited, pH fluctuations in the different synaptic compartments is generally accepted and clearly implies that pH regulatory elements are essential to maintain proper synaptic function. Since many synaptic elements are strongly pH dependent, limitations and alterations in synaptic pH homeostasis could potentially feed-back on neuronal activity itself. Intriguingly,

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it has already been shown that direct release of protons during vesicle exocytosis can act as a negative feedback on closely associated calcium channels in the mammalian retina [3,22]. In this system, synaptic cleft acidification of retinal cells is thought to underlie surround inhibition and thereby helps to form the receptive field (for review see [23]). The discovery of acid-sensing ion channels (ASICs) is another example for a pH-induced feedback mechanisms [24]. At least four genes and their alternatively spliced transcripts code for subunits of such ion channels, which belong to the degenerin/epithelial Na^+ channel superfamily and are characterized by a strong H^+-sensitivity as well as their permeability for cations. ASICs are widely expressed in the mammalian nervous system and have been shown to localize mostly to somato-dendritic regions of neurons [25,26]. ASICs have been implicated in many neurological disorders like e.g., ischemic stroke, epileptic seizures and pain (for review see [27]). Interestingly, one study suggested that seizure termination critically depends on ASIC activation by the fall in extracellular pH in response to epileptic neuronal activity [28].

2. Effects of pH transients on presynaptic function

Loading of synaptic vesicles with different neurotransmitters depends on vesicular proton gradients [29]. Hence, variations in intracellular pH could directly interfere with neurotransmitter loading. It has been shown that the glutamate uptake by astrocytes is pH sensitive and provides a mechanism which can protect neurons from glutamatergic excitotoxicity due to reversed glutamate uptake under ischemic conditions [30].

The function of proteins, enzymatic activity as well as protein–protein interactions are sensitive to alterations in pH and thus changes in pH can impact on the release of synaptic vesicles, which depends on the concerted action of a complex machinery of different proteins (for review see [31]). In particular, the initial rise in the presynaptic calcium concentration mediated via voltage-gated calcium channels [32] is pH dependent, as the opening and the conductivity of presynaptic voltage-gated calcium channels strongly depend on both extracellular and intracellular pH [33]. Protons can directly bind to sensors within the pore of the channel and thereby reduce channel conductance [34,35], shield membrane-bound charges and thus shift the channel activation voltage to more positive values [36,37]. The rise in presynaptic calcium is augmented by release of calcium from intracellular stores which is mediated via inositol 1,4,5-trisphosphate and ryanodine receptors. Both receptors also show strong pH dependence [38,39]. Studies on spontaneous vesicle release by electrophysiological methods confirmed that lowering of intracellular pH in hippocampal neurons indeed results in a decreased rate of synaptic vesicle release and hence limited excitability [40,41]. Further studies are necessary to investigate if presynaptic pH modulates synapse function mainly by alterations in calcium transients or if multiple effects add up.

3. Effects of pH transients on postsynaptic function

NMDA receptors are strongly modulated by changes in extracellular pH [42,43]. An increase in extracellular pH facilitates the activation of NMDA receptors, whereas a decrease in extracellular pH inhibits ion channel function [42–44]. The transient increase in extracellular pH elicited by high-frequency stimulation of afferents in the hippocampus has been shown to be sufficient to augment NMDA-receptor responses in vitro [45]. This is most likely also relevant in vivo both in physiological and pathophysiological conditions. In contrast, kinetics and amplitudes of AMPA- and Kainate-receptors are only marginally modulated by alteration of extracellular pH [46].

Interestingly, $GABA_A$ receptor mediated currents are enlarged by low extracellular pH, whereas a high pH rather inhibits the GABA response [47–49]. $GABA_A$ receptors also conduct bicarbonate. As a consequence, GABAergic transmission can cause alterations of both intra- and extracellular pH [11]. In contrast to the direction of chloride fluxes, which can vary in dependence of the existing chloride gradients, which are set by the cation-chloride co-transporters NKCC1 and KCC2 [50–52], the existing gradients always drive HCO_3^- out of the neurons under physiological conditions. Both gradients contribute to the balance between neuronal excitation and inhibition. Only little is known about the role of pH for signaling via $GABA_B$ receptors or receptors of other neurotransmitters.

In conclusion, electrical stimulation or synchronized neuronal activity results first in an initial transient alkaline shift of the extracellular pH that is followed by a prolonged acidosis (for review see [5]). The short-lived initial increase in pH has been shown to be sufficient to augment glutamatergic excitation by activation of NMDA receptors in acute slice experiments [45] and most likely inhibits GABAergic transmission. In contrast, under conditions of sustained stimulation [53] or pathological neuronal activity [12], the following long-lasting acidosis is predicted to diminish glutamatergic neurotransmission and boost GABAergic inhibition, which was confirmed for cultured neurons [54].

This indicates that intrinsic pH transients serve as a feedback mechanism to keep the delicate balance between neuronal excitability and inhibition but also implies that neuronal and especially synaptic pH has to be tightly controlled.

4. Mechanisms to regulate synaptic pH

In general, cellular pH homeostasis is established by transport or buffering of acid equivalents. In neurons acid loading is largely established by Na^+ independent Cl^-/HCO_3^- exchangers [55], whereas Na^+/H^+ exchangers [56], Na^+-driven Cl^-/HCO_3^- exchangers and Na^+/HCO_3^- co-transporters [57] mediate acid extrusion. Another family of bicarbonate transporters, which can be distinguished from the family of SLC4 transporters [55,58], are classified as members of the SLC26 family [59], however, if at all, members of the SLC26 family of bicarbonate transporters are thought to play a minor role for neuronal pH homeostasis [60]. Neuronal pH is also affected by monocarboxylate transporters [61] but their role in the brain under physiological conditions is limited whereas they are more important in tissues with a high energy demand like in tumors [62]. Although so far no conclusive data exist that the plasma membrane calcium ATPase also plays a direct role for pH regulation, a brain-specific isoform with a predominant synaptic localization has been described [63], which may contribute to synaptic pH homeostasis [9,10].

Bicarbonate is a very important pH buffering system because it can be regulated by respiration. Carbonic anhydrases promote the interconversion of carbon dioxide and water to bicarbonate and protons, and thereby significantly contribute to the intra- and extracellular buffering capacity in the brain [64].

For a more general comprehensive review on cellular pH sensors and regulators, see [65], [5] and [66]. In the following we will mainly focus on the Na^+/H^+ exchanger NHE1, the Na^+ coupled anion-exchangers NCBE and NDCBE, and Na^+-HCO_3^- co-transporters, all mediating acid extrusion.

5. NHE1

The transmembrane Na^+-gradient is established by the Na^+/K^+ ATPase. The Na^+ gradient is then used to energize the electroneutral exchange of one extracellular sodium for one proton by Na^+/H^+ exchangers (NHE) [67]. So far 9 different isoforms of Na^+/H^+ exchangers have been identified and all of these are expressed in

the central nervous system (for review see [68]). NHE1/SLC9A1 is ubiquitously expressed and a multifunctional protein which does not only contribute to intracellular pH regulation but also volume regulation, cell migration, and also interacts with components of the cytoskeleton [69]. Because of the lack of suitable antibodies localization studies for Slc9a1 are limited. However, most studies suggest that Slc9a1 localizes to presynaptic nerve terminals of GABAergic neurons [54,70,71]. Disruption of Slc9a1 in mice resulted in a severe phenotype with locomotor deficits, epileptic seizures, neurodegeneration, and early mortality [72,73]. Slc9a1 deficient neurons had a lower steady-state pH and a delayed recovery from acid loads [74]. The epileptic phenotype in Slc9a1 knockout mice is therefore surprising, because an increase in pH is generally associated with an increase of neuronal excitability. However, disruption of Nhe1 results in a more complex phenotype with increased Na^+-current density in hippocampal neurons [75,76] as well as increased neuronal cell death [72]. There is also indirect evidence from electrophysiological recordings with pharmacological inhibitors of Na^+/H^+ exchange like amiloride, suggesting that the Na^+/H^+ exchanger, most likely NHE1, localizes to inhibitory and excitatory presynaptic nerve terminals [54,70,71]. In an elegant study by Dietrich and Morad the impact of extracellular pH buffering on the spontaneous release of GABAergic vesicles in cerebellar granule cells was investigated. The results from this study suggest that Nhe1 activity may not only affect presynaptic vesicle release by increasing intracellular pH but also boost GABAergic neurotransmission by increasing $GABA_A$ receptor responses at the postsynapse via the extracellular pH [54].

6. Na^+ coupled anion-exchangers

From early pH recordings in the squid giant axon and in snail neurons it became evident that Na^+-dependent Cl^-/HCO_3^- exchange plays an essential role in the control of intracellular pH of neurons [77,78]. This observation has been supported by the demonstration of Na^+-driven Cl^-/HCO_3^- exchange in different preparations of hippocampal neurons [79–82]. But the molecular correlate remained unclear, until a first cDNA was cloned from drosophila [83] and from a mouse insulinoma cell line [84]. In mammals Na^+-dependent Cl^-/HCO_3^- exchange is mediated by NDBCE (SLC4A8) and NCBE (SLC4A10). The initial transport characterization of NCBE/SLC4A10 as Na^+-dependent Cl^-/HCO_3^- exchanger was confirmed for rat [84–86], whereas the human cDNA was rather characterized as an electroneutral Na^+/HCO_3^- co-transporter (NBCn2) with Cl^- self-exchange activity [87]. Some of the controversy may be explained by the different expression systems used in the different studies like mammalian cells and *Xenopus* oocytes, temperature and composition of solutions, the transfection/injection efficiency or molecular tagging of the transport proteins.

In drosophila disruption of Na^+-dependent Cl^-/HCO_3^- exchange results in early lethality of the larvae [83]. Surprisingly, the phenotype of Slc4a8 knockout mice is very mild with some minor deficits in different behavioral paradigms [41,88], whereas Slc4a10 knockout mice experience a critical period within their first week of life with a decreased gain of body weight during postnatal development. They also display a drastic reduction of brain ventricle size [89] and visual impairment [90].

Detailed expression analysis in the brain revealed a significant overlap between both transporters. In the hippocampus Slc4a8 as well as Slc4a10 are expressed in pyramidal neurons [41]. The synaptic expression of Slc4a8 was further analyzed by ultrastructural analysis [41,91]. Transmission electron microscopy of freeze-fractured synaptosomes of wild-type mice revealed that Slc4a8 co-localizes with different presynaptic markers like e.g., syntaxin [41] or SNAP-25 (Fig. 1). The presynaptic expression of Slc4a8

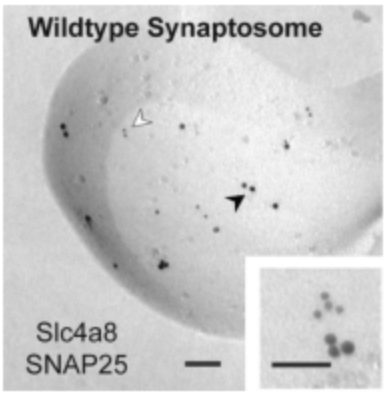

Fig. 1. Presynaptic expression of Slc4a8/NDCBE. Transmission electron microscopy of a freeze-fractured synaptosomes isolated from wild-type mouse brains immunogold-labeled for Slc4a8 (large grains 10 nm) and the presynaptic marker SNAP25 (small grains 5 nm). Images show the proteoplasmic side of synaptosome membranes. Scale bars correspond to 100 and 50 nm.

was nicely supported by the electrophysiological characterization of Slc4a8 knockout mice [41] and confirmed in an independent study [91]. In agreement with the classification of NDCBE as an acid extruder, Slc4a8 deficient neurons displayed a lower steady state pH and a defective pH regulation. Electrophysiological analysis and FM-imaging further showed a decrease in spontaneous and stimulated release of glutamatergic synaptic vesicles in knockout neurons. In accordance with a predominant presynaptic localization, there was no effect on the post-synaptic kinetics of AMPA receptor currents detected. Moreover, the release of GABAergic vesicles as evidenced from recordings of mIPSCs in acute hippocampal brain slices did not differ between genotypes [41]. In contrast to NDCBE, NCBE has also been detected in hippocampal interneurons and co-localizes with pre- and postsynaptic markers of GABAergic synapses in the hippocampus [89]. The recovery of hippocampal principal cells from acid loads was delayed in acute brain slices of Slc4a10 knockout mice, although there was no difference in the steady state pH. Here, this affected network excitability was studied in the 4-aminopyridine model of interictal discharges in acute brain slices. Although the frequency of the interictal-like events at baseline levels did not differ between genotypes, the decreased frequency upon a propionate pulse was prolonged in the knockout [89]. Interestingly, disruption of either Slc4a8 or the Slc4a10 in mice increased the seizure threshold in different seizure inducing paradigms [41,89]. In contrast, Slc9a1 and Slc4a3 knockout mice are more susceptible to seizures [72,92].

7. Na^+/HCO_3^- co-transporters

Na^+/HCO_3^- co-transporters also mediate net acid extrusion. The electrogenic Na^+/HCO_3^- co-transporter Slc4a4 (also called NBCe1) and the electroneutral Na^+/HCO_3^- co-transporter Slc4a7 (also called NBCn1) are broadly expressed in the central nervous system [93,94]. Slc4a7 was shown to co-localize with PSD-95, a postsynaptic protein of glutamatergic synapses [93]. Slc4a7 expression was increased upon metabolic acidosis and this up-regulation was associated with glutamate excitotoxicity [95]. Slc4a7 knockout mice have been reported to display severe sensory deficits [96], however, a detailed analysis of its role for synaptic transmission is missing to date.

Recent data suggest that Slc4a4 helps to prevent the large, prolonged, Ca^{2+}-dependent alkaline shift upon depolarization of neurons [97]. As prolonged positive shifts in membrane potential, which might cause a sustained net alkaline shift, are a recurrent condition during normal brain function, depolarization activated

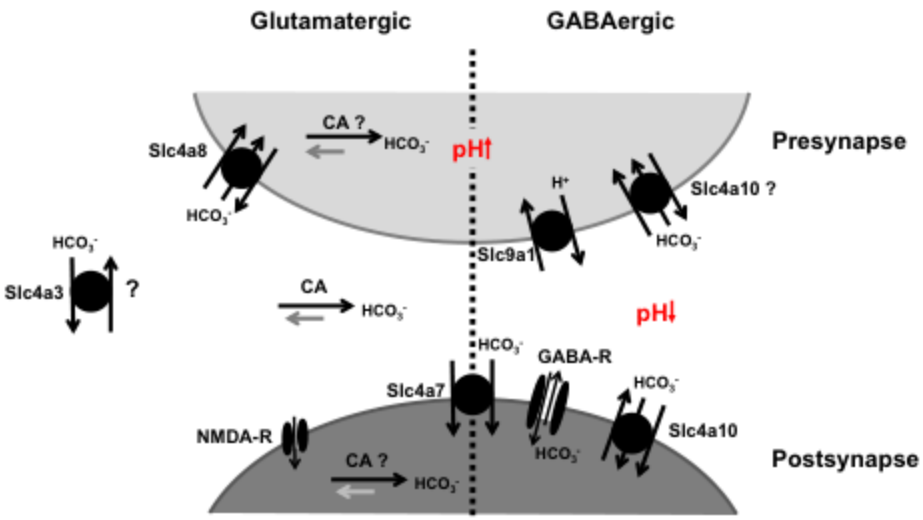

Fig. 2. Model displaying different regulators involved in the control of synaptic pH. Slc4a8 localizes to glutamatergic presynapses and modulates the release of glutamate vesicles in a pH-dependent manner [41]. Slc9a1 appears to play an important role for pH regulation at GABAergic nerve terminals [69]. Slc4a10 is likely to be expressed on both sides of GABAergic synapses [88]. Slc4a7 localizes to the postsynaptic site [94]. Extracellular and probably also intracellular CAs increase the buffering capacity of the different compartments [8]. Whether or not Slc4a3 and other pH regulators modulate synaptic activity remains unclear.

acid extrusion most likely also plays a role under physiological conditions. It was speculated that this depolarization induced alkalinization may be an adaptation to preempt untoward acidification from large intracellular Ca^{2+} loads, while maintaining or accelerating the rate of glucose utilization through the glycolytic pathway. Interestingly, a parallel Cl^--dependent mechanism also contributed to this depolarization induced alkalinization, but its molecular correlate is yet unclear [97].

8. Carbonic anhydrases

Carbonic anhydrases (CA) catalyse the rapid interconversion of carbon dioxide and water to bicarbonate and protons and vice versa. CA activity was first described in red blood cells [98] and later became evident in many other organs. In the mammalian brain at least 10 catalytically active isoforms or CAs have been described, which differ in cellular [99,100] and sub-cellular [101,102] localization. Evidence on the role of carbonic anhydrases for synaptic transmission has largely been deduced from studies with pharmacological blockers of CA. Experiments with membrane-permeant and membrane-impermeant blockers of carbonic anhydrases revealed that extracellular CA are involved in the regulation of the interstitial pH in the brain [8,103]. The extracellular and membrane-bound carbonic anhydrases CA4 and CA14 are abundantly expressed by neurons [101,104] and have been implicated in buffering extracellular alkaline shifts following neuronal activity [8,105]. Furthermore, functional coupling of CA activity of AE3-mediated bicarbonate transport was described in hippocampal neurons [106]. Intracellular CAs have been shown to be essential for synchronous firing of hippocampal neurons by enabling tonic GABAergic excitation [107].

CA inhibitors are widely used as anticovulsant drugs [108]. Hence, closer analysis of synaptic expression CAs and a better understanding of their functional role could greatly impact on future clinical applications.

9. Conclusion

There is ample evidence that synaptic function critically depends on intracellular and extracellular pH gradients and that synaptic activity also causes local pH gradients. Hence, it comes as no surprise that several proteins involved in local pH control localize to synaptic structures. The use of high resolution microscopy with better pH sensitive probes may allow measuring the pH in different synaptic compartments and how the pH changes with synaptic activity. These techniques will also help us to address the role of the different proteins involved in pH homeostasis more precisely (Fig. 2). A better understanding of these processes could also help to identify new pharmacological targets to treat epilepsy or pathological conditions involving synaptic transmission.

References

[1] Forgac, M. (2007) Vacuolar ATPases: rotary proton pumps in physiology and pathophysiology. Nat. Rev. Mol. Cell Biol. 8, 917–929.
[2] Krishtal, O., Osipchuk, Y., Shelest, T. and Smirnoff, S. (1987) Rapid extracellular pH transients related to synaptic transmission in rat hippocampal slices. Brain Res. 436, 352–356.
[3] DeVries, S. (2001) Exocytosed protons feedback to suppress the Ca^{2+} current in mammalian cone photoreceptors. Neuron 32, 1107–1117.
[4] Zhang, Z., Nguyen, K.T., Barrett, E.F. and David, G. (2010) Vesicular ATPase inserted into the plasma membrane of motor terminals by exocytosis alkalinizes cytosolic pH and facilitates endocytosis. Neuron 68, 1097–1108.
[5] Chesler, M. (2003) Regulation and modulation of pH in the brain. Physiol. Rev. 83, 1183–1221.
[6] Gottfried, J.A. and Chesler, M. (1996) Temporal resolution of activity-dependent pH shifts in rat hippocampal slices. J. Neurophysiol. 76, 2804–2807.
[7] Tong, C.K., Chen, K. and Chesler, M. (2006) Kinetics of activity-evoked pH transients and extracellular pH buffering in rat hippocampal slices. J. Neurophysiol. 95, 3686–3697.
[8] Chen, J. and Chesler, M. (1992) PH transients evoked by excitatory synaptic transmission are increased by inhibition of extracellular carbonic anhydrase. Proc. Natl. Acad. Sci. USA 89, 7786–7790.
[9] Schwiening, C.J., Kennedy, H.J. and Thomas, R.C. (1993) Calcium-hydrogen exchange by the plasma membrane Ca-ATPase of voltage-clamped snail neurons. Proc. Biol. Sci. 253, 285–289.
[10] Makani, S. and Chesler, M. (2010) Rapid rise of extracellular pH evoked by neural activity is generated by the plasma membrane calcium ATPase. J. Neurophysiol. 103, 667–676.
[11] Kaila, K. and Voipio, J. (1987) Postsynaptic fall in intracellular pH induced by GABA-activated bicarbonate conductance. Nature 330, 163–165.
[12] Caspers, H. and Speckmann, E.J. (1972) Cerebral pO_2, pCO_2 and pH: changes during convulsive activity and their significance for spontaneous arrest of seizures. Epilepsia 13, 699–725.
[13] Voipio, J. and Kaila, K. (1993) Interstitial PCO_2 and pH in rat hippocampal slices measured by means of a novel fast CO_2/H(+)-sensitive microelectrode based on a PVC-gelled membrane. Pflugers Arch. 423, 193–201.
[14] Fedirko, N., Svichar, N. and Chesler, M. (2006) Fabrication and use of high-speed, concentric H^+- and Ca^{2+}-selective microelectrodes suitable for in vitro extracellular recording. J. Neurophysiol. 96, 919–924.

[15] Chesler, M. and Kaila, K. (1992) Modulation of pH by neuronal activity. Trends Neurosci. 15, 396–402.
[16] Schwiening, C. and Willoughby, D. (2002) Depolarization-induced pH microdomains and their relationship to calcium transients in isolated snail neurones. J. Physiol. 538, 371–382.
[17] Nachshen, D.A. and Drapeau, P. (1988) The regulation of cytosolic pH in isolated presynaptic nerve terminals from rat brain. J. Gen. Physiol. 91, 289–303.
[18] Rossano, A.J., Chouhan, A.K. and Macleod, G.T. (2013) Genetically encoded pH-indicators reveal activity-dependent cytosolic acidification of Drosophila motor nerve termini in vivo. J. Physiol. 591, 1691–1706.
[19] Saggau, P., Gray, R. and Dani, J.A. (1999) Optical measurements of calcium signals in mammalian presynaptic terminals. Methods Enzymol. 294, 3–19.
[20] Arosio, D., Ricci, F., Marchetti, L., Gualdani, R., Albertazzi, L. and Beltram, F. (2010) Simultaneous intracellular chloride and pH measurements using a GFP-based sensor. Nat. Methods 7, 516–518.
[21] Diering, G.H., Mills, F., Bamji, S.X. and Numata, M. (2011) Regulation of dendritic spine growth through activity-dependent recruitment of the brain-enriched Na^+/H^+ exchanger NHE5. Mol. Biol. Cell 22, 2246–2257.
[22] Palmer, M.J., Hull, C., Vigh, J. and von Gersdorff, H. (2003) Synaptic cleft acidification and modulation of short-term depression by exocytosed protons in retinal bipolar cells. J. Neurosci. 23, 11332–11341.
[23] Hirasawa, H., Yamada, M. and Kaneko, A. (2012) Acidification of the synaptic cleft of cone photoreceptor terminal controls the amount of transmitter release, thereby forming the receptive field surround in the vertebrate retina. J. Physiol. Sci. 62, 359–375.
[24] Waldmann, R., Champigny, G., Bassilana, F., Heurteaux, C. and Lazdunski, M. (1997) A proton-gated cation channel involved in acid-sensing. Nature 386, 173–177.
[25] Zha, X.M., Wemmie, J.A., Green, S.H. and Welsh, M.J. (2006) Acid-sensing ion channel 1a is a postsynaptic proton receptor that affects the density of dendritic spines. Proc. Natl. Acad. Sci. USA 103, 16556–16561.
[26] Wemmie, J.A. et al. (2002) The acid-activated ion channel ASIC contributes to synaptic plasticity, learning, and memory. Neuron 34, 463–477.
[27] Wemmie, J.A., Price, M.P. and Welsh, M.J. (2006) Acid-sensing ion channels: advances, questions and therapeutic opportunities. Trends Neurosci. 29, 578–586.
[28] Ziemann, A.E., Schnizler, M.K., Albert, G.W., Severson, M.A., Howard, M.A., Welsh, M.J. and Wemmie, J.A. (2008) Seizure termination by acidosis depends on ASIC1a. Nat. Neurosci. 11, 816–822.
[29] Blakely, R.D. and Edwards, R.H. (2012) Vesicular and plasma membrane transporters for neurotransmitters. Cold Spring Harb. Perspect. Biol. 4.
[30] Billups, B. and Attwell, D. (1996) Modulation of non-vesicular glutamate release by pH. Nature 379, 171–174.
[31] Südhof, T.C. (1995) The synaptic vesicle cycle: a cascade of protein–protein interactions. Nature 375, 645–653.
[32] Schneggenburger, R. and Neher, E. (2000) Intracellular calcium dependence of transmitter release rates at a fast central synapse. Nature 406, 889–893.
[33] Tombaugh, G. and Somjen, G. (1997) Differential sensitivity to intracellular pH among high- and low-threshold Ca^{2+} currents in isolated rat CA1 neurons. J. Neurophysiol. 77, 639–653.
[34] Chen, X., Bezprozvanny, I. and Tsien, R. (1996) Molecular basis of proton block of L-type Ca^{2+} channels. J. Gen. Physiol. 108, 363–374.
[35] Prod'hom, B., Pietrobon, D. and Hess, P. (1987) Direct measurement of proton transfer rates to a group controlling the dihydropyridine-sensitive Ca^{2+} channel. Nature 329, 243–246.
[36] Klöckner, U. and Isenberg, G. (1994) Calcium channel current of vascular smooth muscle cells: extracellular protons modulate gating and single channel conductance. J. Gen. Physiol. 103, 665–678.
[37] Zhou, W. and Jones, S. (1996) The effects of external pH on calcium channel currents in bullfrog sympathetic neurons. Biophys. J. 70, 1326–1334.
[38] Tsukioka, M., Iino, M. and Endo, M. (1994) PH dependence of inositol 1,4,5-trisphosphate-induced Ca^{2+} release in permeabilized smooth muscle cells of the guinea-pig. J. Physiol. 475, 369–375.
[39] Ma, J., Fill, M., Knudson, C., Campbell, K. and Coronado, R. (1988) Ryanodine receptor of skeletal muscle is a gap junction-type channel. Science 242, 99–102.
[40] Lee, J., Taira, T., Pihlaja, P., Ransom, B. and Kaila, K. (1996) Effects of CO_2 on excitatory transmission apparently caused by changes in intracellular pH in the rat hippocampal slice. Brain Res. 706, 210–216.
[41] Sinning, A., Liebmann, L., Kougioumtzes, A., Westermann, M., Bruehl, C. and Hübner, C.A. (2011) Synaptic glutamate release is modulated by the Na^+-driven Cl^-/HCO_3^- exchanger Slc4a8. J. Neurosci. 31, 7300–7311.
[42] Traynelis, S. and Cull-Candy, S. (1990) Proton inhibition of N-methyl-D-aspartate receptors in cerebellar neurons. Nature 345, 347–350.
[43] Tang, C.M., Dichter, M. and Morad, M. (1990) Modulation of the N-methyl-D-aspartate channel by extracellular H^+. Proc. Natl. Acad. Sci. USA 87, 6445–6449.
[44] Chen, Y.H., Wu, M.L. and Fu, W.M. (1998) Regulation of presynaptic NMDA responses by external and intracellular pH changes at developing neuromuscular synapses. J. Neurosci. 18, 2982–2990.
[45] Makani, S. and Chesler, M. (2007) Endogenous alkaline transients boost postsynaptic NMDA receptor responses in hippocampal CA1 pyramidal neurons. J. Neurosci. 27, 7438–7446.
[46] Lei, S., Orser, B.A., Thatcher, G.R., Reynolds, J.N. and MacDonald, J.F. (2001) Positive allosteric modulators of AMPA receptors reduce proton-induced receptor desensitization in rat hippocampal neurons. J. Neurophysiol. 85, 2030–2038.
[47] Krishek, B.J., Amato, A., Connolly, C.N., Moss, S.J. and Smart, T.G. (1996) Proton sensitivity of the GABA(A) receptor is associated with the receptor subunit composition. J. Physiol. 492 (Pt 2), 431–443.
[48] Pasternack, M., Smirnov, S. and Kaila, K. (1996) Proton modulation of functionally distinct $GABA_A$ receptors in acutely isolated pyramidal neurons of rat hippocampus. Neuropharmacology 35, 1279–1288.
[49] Mozrzymas, J.W., Zarnowska, E.D., Pytel, M., Mercik, K. and Zarmowska, E.D. (2003) Modulation of GABA(A) receptors by hydrogen ions reveals synaptic GABA transient and a crucial role of the desensitization process. J. Neurosci. 23, 7981–7992.
[50] Hübner, C., Stein, V., Hermans-Borgmeyer, I., Meyer, T., Ballanyi, K. and Jentsch, T. (2001) Disruption of KCC2 reveals an essential role of K–Cl cotransport already in early synaptic inhibition. Neuron 30, 515–524.
[51] Ben-Ari, Y. (2002) Excitatory actions of gaba during development: the nature of the nurture. Nat. Rev. Neurosci. 3, 728–739.
[52] Dzhala, V.I. et al. (2005) NKCC1 transporter facilitates seizures in the developing brain. Nat. Med. 11, 1205–1213.
[53] Urbanics, R., Leniger-Follert, E. and Lübbers, D.W. (1978) Time course of changes of extracellular H^+ and K^+ activities during and after direct electrical stimulation of the brain cortex. Pflugers Arch. 378, 47–53.
[54] Dietrich, C.J. and Morad, M. (2010) Synaptic acidification enhances $GABA_A$ signaling. J. Neurosci. 30, 16044–16052.
[55] Alper, S. (2009) Molecular physiology and genetics of Na^+-independent SLC4 anion exchangers. J. Exp. Biol. 212, 1672–1683.
[56] Luo, J. and Sun, D. (2007) Physiology and pathophysiology of Na(+)/H(+) exchange isoform 1 in the central nervous system. Curr. Neurovasc. Res. 4, 205–215.
[57] Boron, W., Chen, L. and Parker, M. (2009) Modular structure of sodium-coupled bicarbonate transporters. J. Exp. Biol. 212, 1697–1706.
[58] Romero, M.F., Chen, A.P., Parker, M.D. and Boron, W.F. (2013) The SLC4 family of bicarbonate ($HCO_3(-)$) transporters. Mol. Aspects Med. 34, 159–182.
[59] Sterling, D. and Casey, J.R. (2002) Bicarbonate transport proteins. Biochem. Cell Biol. 80, 483–497.
[60] Dorwart, M.R., Shcheynikov, N., Yang, D. and Muallem, S. (2008) The solute carrier 26 family of proteins in epithelial ion transport. Physiology (Bethesda) 23, 104–114.
[61] Hertz, L. and Dienel, G. (2005) Lactate transport and transporters: general principles and functional roles in brain cells. J. Neurosci. Res. 79, 11–18.
[62] Halestrap, A.P. and Meredith, D. (2004) The SLC16 gene family-from monocarboxylate transporters (MCTs) to aromatic amino acid transporters and beyond. Pflugers Arch. 447, 619–628.
[63] Jensen, T.P., Filoteo, A.G., Knopfel, T. and Empson, R.M. (2007) Presynaptic plasma membrane Ca^{2+} ATPase isoform 2a regulates excitatory synaptic transmission in rat hippocampal CA3. J. Physiol. 579, 85–99.
[64] Supuran, C. (2008) Carbonic anhydrases – an overview, Curr. Pharm. Des. 14, 603–614.
[65] Casey, J., Grinstein, S. and Orlowski, J. (2010) Sensors and regulators of intracellular pH. Nat. Rev. Mol. Cell Biol. 11, 50–61.
[66] Obara, M., Szeliga, M. and Albrecht, J. (2008) Regulation of pH in the mammalian central nervous system under normal and pathological conditions: facts and hypotheses. Neurochem. Int. 52, 905–919.
[67] Jean, T., Frelin, C., Vigne, P., Barbry, P. and Lazdunski, M. (1985) Biochemical properties of the Na^+/H^+ exchange system in rat brain synaptosomes. Interdependence of internal and external pH control of the exchange activity. J. Biol. Chem. 260, 9678–9684.
[68] Luo, J. and Sun, D. (2007) Physiology and pathophysiology of Na(+)/H(+) exchange isoform 1 in the central nervous system. Curr. Neurovasc. Res. 4, 205–215.
[69] Malo, M.E. and Fliegel, L. (2006) Physiological role and regulation of the Na^+/H^+ exchanger. Can. J. Physiol. Pharmacol. 84, 1081–1095.
[70] Jang, I., Brodwick, M., Wang, Z., Jeong, H., Choi, B. and Akaike, N. (2006) The Na(+)/H(+) exchanger is a major pH regulator in GABAergic presynaptic nerve terminals synapsing onto rat CA3 pyramidal neurons. J. Neurochem. 99, 1224–1236.
[71] Trudeau, L.E., Parpura, V. and Haydon, P.G. (1999) Activation of neurotransmitter release in hippocampal nerve terminals during recovery from intracellular acidification. J. Neurophysiol. 81, 2627–2635.
[72] Cox, G. et al. (1997) Sodium/hydrogen exchanger gene defect in slow-wave epilepsy mutant mice. Cell 91, 139–148.
[73] Bell, S.M., Schreiner, C.M., Schultheis, P.J., Miller, M.L., Evans, R.L., Vorhees, C.V., Shull, G.E. and Scott, W.J. (1999) Targeted disruption of the murine Nhe1 locus induces ataxia, growth retardation, and seizures. Am. J. Physiol. 276, C788–C795.
[74] Yao, H., Ma, E., Gu, X. and Haddad, G. (1999) Intracellular pH regulation of CA1 neurons in Na(+)/H(+) isoform 1 mutant mice. J. Clin. Invest. 104, 637–645.
[75] Gu, X., Yao, H. and Haddad, G. (2001) Increased neuronal excitability and seizures in the Na(+)/H(+) exchanger null mutant mouse. Am. J. Physiol. Cell Physiol. 281, C496–C503.
[76] Xia, Y., Zhao, P., Xue, J., Gu, X., Sun, X., Yao, H. and Haddad, G. (2003) Na+ channel expression and neuronal function in the Na^+/H^+ exchanger 1 null mutant mouse. J. Neurophysiol. 89, 229–236.

[77] Boron, W. and De Weer, P. (1976) Intracellular pH transients in squid giant axons caused by CO_2, NH_3, and metabolic inhibitors. J. Gen. Physiol. 67, 91–112.
[78] Thomas, R. (1976) The effect of carbon dioxide on the intracellular pH and buffering power of snail neurones. J. Physiol. 255, 715–735.
[79] Schwiening, C. and Boron, W. (1994) Regulation of intracellular pH in pyramidal neurones from the rat hippocampus by Na(+)-dependent Cl(−)–HCO_3^- exchange. J. Physiol. 475, 59–67.
[80] Baxter, K. and Church, J. (1996) Characterization of acid extrusion mechanisms in cultured fetal rat hippocampal neurones. J. Physiol. 493 (Pt 2), 457–470.
[81] Bevensee, M., Cummins, T., Haddad, G., Boron, W. and Boyarsky, G. (1996) PH regulation in single CA1 neurons acutely isolated from the hippocampi of immature and mature rats. J. Physiol. 494 (Pt 2), 315–328.
[82] Bonnet, U., Leniger, T. and Wiemann, M. (2000) Alteration of intracellular pH and activity of CA3-pyramidal cells in guinea pig hippocampal slices by inhibition of transmembrane acid extrusion. Brain Res. 872, 116–124.
[83] Romero, M., Henry, D., Nelson, S., Harte, P., Dillon, A. and Sciortino, C. (2000) Cloning and characterization of a Na^+-driven anion exchanger (NDAE1). A new bicarbonate transporter. J. Biol. Chem. 275, 24552–24559.
[84] Wang, C., Yano, H., Nagashima, K. and Seino, S. (2000) The Na^+-driven Cl^-/HCO_3^- exchanger. Cloning, tissue distribution, and functional characterization. J. Biol. Chem. 275, 35486–35490.
[85] Grichtchenko, I., Choi, I., Zhong, X., Bray-Ward, P., Russell, J. and Boron, W. (2001) Cloning, characterization, and chromosomal mapping of a human electroneutral Na(+)-driven Cl^-HCO_3 exchanger. J. Biol. Chem. 276, 8358–8363.
[86] Damkier, H., Aalkjaer, C. and Praetorius, J. (2010) Na^+-dependent HCO_3^- import by the slc4a10 gene product involves Cl^- export. J. Biol. Chem. 285, 26998–27007.
[87] Parker, M., Bouyer, P., Daly, C. and Boron, W. (2008) Cloning and characterization of novel human SLC4A8 gene products encoding Na^+-driven $Cl^-/HCO_3(-)$ exchanger variants NDCBE-A, -C, and -D. Physiol. Genomics 34, 265–276.
[88] Leviel, F. et al. (2010) The Na^+-dependent chloride-bicarbonate exchanger SLC4A8 mediates an electroneutral Na^+ reabsorption process in the renal cortical collecting ducts of mice. J. Clin. Invest. 120, 1627–1635.
[89] Jacobs, S. et al. (2008) Mice with targeted Slc4a10 gene disruption have small brain ventricles and show reduced neuronal excitability. Proc. Natl. Acad. Sci. USA 105, 311–316.
[90] Hilgen, G. et al. (2012) Lack of the sodium-driven chloride bicarbonate exchanger NCBE impairs visual function in the mouse retina. PLoS ONE 7, e46155.
[91] Burette, A.C., Weinberg, R.J., Sassani, P., Abuladze, N., Kao, L. and Kurtz, I. (2011) The sodium-driven chloride/bicarbonate exchanger in presynaptic terminals. J. Comp. Neurol. 520, 1481–1492.
[92] Hentschke, M. et al. (2006) Mice with a targeted disruption of the Cl^-/HCO_3^- exchanger AE3 display a reduced seizure threshold. Mol. Cell. Biol. 26, 182–191.
[93] Cooper, D., Saxena, N., Yang, H., Lee, H., Moring, A., Lee, A. and Choi, I. (2005) Molecular and functional characterization of the electroneutral Na/HCO_3 cotransporter NBCn1 in rat hippocampal neurons. J. Biol. Chem. 280, 17823–17830.
[94] Bevensee, M.O., Schmitt, B.M., Choi, I., Romero, M.F. and Boron, W.F. (2000) An electrogenic Na(+)–HCO(−)(3) cotransporter (NBC) with a novel COOH-terminus, cloned from rat brain. Am. J. Physiol. Cell Physiol. 278, C1200–C1211.
[95] Park, H. et al. (2010) Neuronal expression of sodium/bicarbonate cotransporter NBCn1 (SLC4A7) and its response to chronic metabolic acidosis. Am. J. Physiol. Cell Physiol. 298, C1018–C1028.
[96] Bok, D. et al. (2003) Blindness and auditory impairment caused by loss of the sodium bicarbonate cotransporter NBC3. Nat. Genet. 34, 313–319.
[97] Svichar, N., Esquenazi, S., Chen, H.Y. and Chesler, M. (2011) Preemptive regulation of intracellular pH in hippocampal neurons by a dual mechanism of depolarization-induced alkalinization. J. Neurosci. 31, 6997–7004.
[98] Nyman, P.O. (1961) Purification and properties of carbonic anhydrase from human erythrocytes. Biochim. Biophys. Acta 52, 1–12.
[99] Parkkila, S., Parkkila, A.K., Rajaniemi, H., Shah, G.N., Grubb, J.H., Waheed, A. and Sly, W.S. (2001) Expression of membrane-associated carbonic anhydrase XIV on neurons and axons in mouse and human brain. Proc. Natl. Acad. Sci. USA 98, 1918–1923.
[100] Agnati, L.F., Tinner, B., Staines, W.A., Väänänen, K. and Fuxe, K. (1995) On the cellular localization and distribution of carbonic anhydrase II immunoreactivity in the rat brain. Brain Res. 676, 10–24.
[101] Tong, C.K., Brion, L.P., Suarez, C. and Chesler, M. (2000) Interstitial carbonic anhydrase (CA) activity in brain is attributable to membrane-bound CA type IV. J. Neurosci. 20, 8247–8253.
[102] Langley, O.K., Ghandour, M.S., Vincendon, G. and Gombos, G. (1980) Carbonic anhydrase: an ultrastructural study in rat cerebellum. Histochem. J. 12, 473–483.
[103] Kaila, K., Paalasmaa, P., Taira, T. and Voipio, J. (1992) pH transients due to monosynaptic activation of GABAA receptors in rat hippocampal slices. NeuroReport 3, 105–108.
[104] Parkkila, S., Parkkila, A.K., Rajaniemi, H., Shah, G.N., Grubb, J.H., Waheed, A. and Sly, W.S. (2001) Expression of membrane-associated carbonic anhydrase XIV on neurons and axons in mouse and human brain. Proc. Natl. Acad. Sci. USA 98, 1918–1923.
[105] Shah, G.N., Ulmasov, B., Waheed, A., Becker, T., Makani, S., Svichar, N., Chesler, M. and Sly, W.S. (2005) Carbonic anhydrase IV and XIV knockout mice: roles of the respective carbonic anhydrases in buffering the extracellular space in brain. Proc. Natl. Acad. Sci. USA 102, 16771–19776.
[106] Svichar, N., Waheed, A., Sly, W., Hennings, J., Hübner, C. and Chesler, M. (2009) Carbonic anhydrases CA4 and CA14 both enhance AE3-mediated $Cl^-HCO_3^-$ exchange in hippocampal neurons. J. Neurosci. 29, 3252–3258.
[107] Ruusuvuori, E., Li, H., Huttu, K., Palva, J.M., Smirnov, S., Rivera, C., Kaila, K. and Voipio, J. (2004) Carbonic anhydrase isoform VII acts as a molecular switch in the development of synchronous gamma-frequency firing of hippocampal CA1 pyramidal cells. J. Neurosci. 24, 2699–2707.
[108] Thiry, A., Dogné, J.M., Supuran, C.T. and Masereel, B. (2008) Anticonvulsant sulfonamides/sulfamates/sulfamides with carbonic anhydrase inhibitory activity: drug design and mechanism of action. Curr. Pharm. Des. 14, 661–671.