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## Endogenous H<sup>+</sup> modulation of NMDA receptor-mediated EPSCs revealed by carbonic anhydrase inhibition in rat hippocampus

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1. The occurrence of extracellular alkaline transients during excitatory synaptic transmission suggests that the NMDA receptor H<sup>+</sup>-modulatory site may have a physiological role. Here we amplify these pH shifts using benzolamide (a carbonic anhydrase inhibitor) and describe concomitant effects on EPSCs in whole-cell clamped CA1 neurones in rat hippocampal slices.
2. In CO<sub>2</sub>-HCO<sub>3</sub><sup>-</sup>-buffered media, benzolamide increased the time to 50% decay (*t*<sub>50</sub>) of the EPSCs by 78 ± 14% (*P* < 0.01, *n* = 10). This occurred simultaneously with amplification of the extracellular alkaline shift (154 ± 14%).
3. In CO<sub>2</sub>-HCO<sub>3</sub><sup>-</sup>-buffered media containing DL-2-amino-5-phosphonovalerate (APV), the EPSC *t*<sub>50</sub> was unaltered by benzolamide, while the extracellular alkaline shifts were increased (111 ± 23%, *n* = 8).
4. In HEPES-buffered media, neither the EPSC *t*<sub>50</sub> nor the extracellular alkaline shift was altered by benzolamide (*n* = 9).
5. These data demonstrate that NMDA receptor activity is dependent on the buffering kinetics of the brain extracellular space. The results suggest that endogenous pH shifts can modulate NMDA receptor function in a physiologically relevant time frame.

Excitatory synaptic transmission in the central nervous system has been associated with a rapid alkalization of the extracellular space (Chesler, 1990; Chesler & Kaila, 1992). Studies with pH-sensitive microelectrodes have established that these alkaline transients begin within 100 ms of postsynaptic current flow (Chesler & Chan, 1988). Although the transmembrane pathway of the acid-base fluxes has not been established, their participation in synaptic modulation has been suggested, in view of their rapid onset and link to glutamatergic transmission (Chen & Chesler, 1992b).

Among the ionotropic glutamate receptors, the NMDA receptor displays a unique sensitivity to extracellular pH. Exogenous elevations in pH have been shown to increase the probability of NMDA-channel opening in cultured cells (Tang, Dichter & Morad, 1990; Traynelis & Cull-Candy, 1990, 1991; Vyklicky, Vlachova & Krusek, 1990). Whether the NMDA receptor is modulated by endogenous pH shifts in a physiological context is unknown.

To determine whether endogenous alkaline transients can modulate NMDA receptor function, we manipulated the buffering capacity of the extracellular space using a

carbonic anhydrase (CA) inhibitor. Extracellular CA catalyses the hydration of CO<sub>2</sub>, which provides rapid buffering and normally limits the size of the stimulus-evoked alkaline shift (Chen & Chesler, 1992c). Inhibition of extracellular CA reduces CO<sub>2</sub> hydration to the uncatalysed rate, and thereby amplifies the extracellular alkalizations. In the present report, benzolamide, a charged, poorly permeant CA inhibitor (Travis, Wiley, Bohdan & Maren, 1964), was used to amplify synaptically evoked alkaline transients in rat hippocampal slices. We demonstrate an immediate, buffer-dependent augmentation of NMDA receptor-mediated currents. Our data suggest that the NMDA receptor may be modulated by endogenous pH transients on a physiologically relevant time scale.

### METHODS

Long-Evans rat pups (8–12 days old) were anaesthetized with methoxyflurane and killed by decapitation. Procedures were carried out with approval of the N.Y.U. Medical Center Institutional Animal Care and Use Committee. Hippocampal slices (300 μm) were prepared on a vibratome in ice-cold Ringer solution. Slices were incubated at room temperature in

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Ringer solution which contained (mM): NaCl, 124; NaHCO<sub>3</sub>, 26; KCl, 3; CaCl<sub>2</sub>, 2; Na<sub>2</sub>PO<sub>4</sub>, 1; glucose, 10; gassed with 95% O<sub>2</sub>, 5% CO<sub>2</sub> (pH 7.4). Ringer solution buffered with 26 mM Hepes (*N*-hydroxyethylpiperazine-*N'*-2-ethanesulphonic acid) had NaHCO<sub>3</sub> omitted and was titrated to pH 7.5 with NaOH. Experiments were all conducted in saline containing 100 μM picrotoxin (Sigma Chemical Co., USA) and no added Mg<sup>2+</sup> in a submersion-style slice chamber at 32 °C. APV (DL-2-amino-5-phosphonovalerate) was purchased from Tocris Neuramin. Benzolamide was a gift from Lederle Laboratories, Pearl River, NY, USA.

Excitatory postsynaptic currents (EPSCs), evoked by bipolar stimulation of the Schaffer collateral fibres (0.05 Hz), were recorded in the whole-cell patch-clamp configuration from neurones in the CA1 pyramidal layer (Blanton, Lo Turco & Kriegstein, 1989). Patch electrodes (2–4 MΩ) contained (mM): CsF, 130; KCl, 10; EGTA, 10; Hepes, 10; titrated to pH 7.2 with CsOH. EPSCs were filtered at 1–2 kHz (4-pole Bessel, –3 dB). Series resistance was compensated and was monitored throughout experiments. The time to 50% decay (*t*<sub>50</sub>) of the EPSC for a given cell was obtained by averaging the *t*<sub>50</sub> of equal numbers of EPSCs (10–20) prior to and following addition of benzolamide. Input resistance was determined from 5 mV hyperpolarizing steps (10 ms duration) given 200 ms prior to each afferent stimulus. Reversal potentials were obtained by linear regression of response amplitudes obtained between –60 and +20 mV, in 10 mV steps. To monitor baseline extracellular pH and the relative amplitude of evoked pH transients, double-barrelled extracellular pH microelectrodes were placed in the vicinity of the patch pipettes within area CA1. The pH microelectrodes used a liquid-sensor pH-sensitive cocktail (Fluka 95291) and were fabricated by standard methods (Chesler & Chan, 1988). Extracellular DC potential was constantly monitored and subtracted from the signal on the pH-sensitive barrel. The unfiltered pH signals were recorded on a strip chart recorder and traced. Statistics are presented as

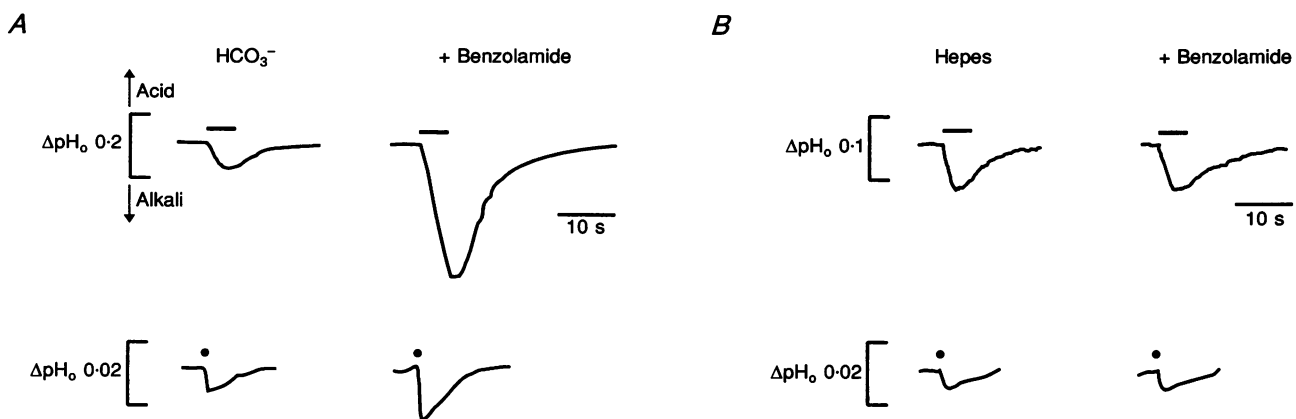
means ± s.e.m. Comparisons were made by a paired, two-tailed *t* test.

## RESULTS

### Stimulus-evoked alkaline shifts in HCO<sub>3</sub><sup>–</sup>- and Hepes-buffered media

An extracellular alkaline transient evoked by repetitive stimulation of the Schaffer collaterals is illustrated in Fig. 1*A* (top). Application of benzolamide (1 μM) increased its amplitude about 5-fold, as noted previously (Chen & Chesler, 1992*a*). At increased gain, responses to single stimuli were noted which were also amplified by benzolamide (Fig. 1*A*, bottom). These small alkalizations peaked within a few hundred milliseconds. This was far faster than the response time of the pH microelectrodes (1–5 s) (Ammann, Lanter, Steiner, Schulthess, Shijo & Simon, 1981), suggesting that the single-shock pH responses were highly attenuated and filtered (see Discussion). Although CA inhibitors can cause a baseline extracellular acidification at high concentrations (Chen & Chesler, 1992*a*), benzolamide at 1 μM had no effect on the baseline extracellular pH, which averaged 7.25 in HCO<sub>3</sub><sup>–</sup>-buffered media (Table 1).

When Hepes served as the extracellular buffer, similar pH responses could be elicited (Chesler & Chan, 1988). However, these pH shifts were not enhanced by the addition of benzolamide (Fig. 1*B*), because the rapid buffering by the Hepes was independent of CA. The baseline extracellular pH in Hepes-buffered media was 7.28, which is comparable to the value in HCO<sub>3</sub><sup>–</sup>-buffered solutions (Table 1).



**Figure 1.** Amplification of evoked alkaline shifts by benzolamide is HCO<sub>3</sub><sup>–</sup> dependent. *A*, in HCO<sub>3</sub><sup>–</sup> Ringer solution (26 mM), repetitive stimulation (bars, 20 Hz) or single shocks (dots) to the Schaffer collateral fibres evoked rapid extracellular alkaline shifts in area CA1. Benzolamide (1 μM) increased the pH transients 2- to 5-fold. *B*, in Hepes-buffered media (26 mM), benzolamide had no effect on the alkaline shifts. Extracellular pH was 7.2–7.3 in all cases. Alkaline shifts are indicated by downward deflections in all figures.

**Table 1.** Input resistance ( $R_N$ ), EPSC reversal potential ( $E_{rev}$ ) and baseline extracellular pH ( $pH_o$ )

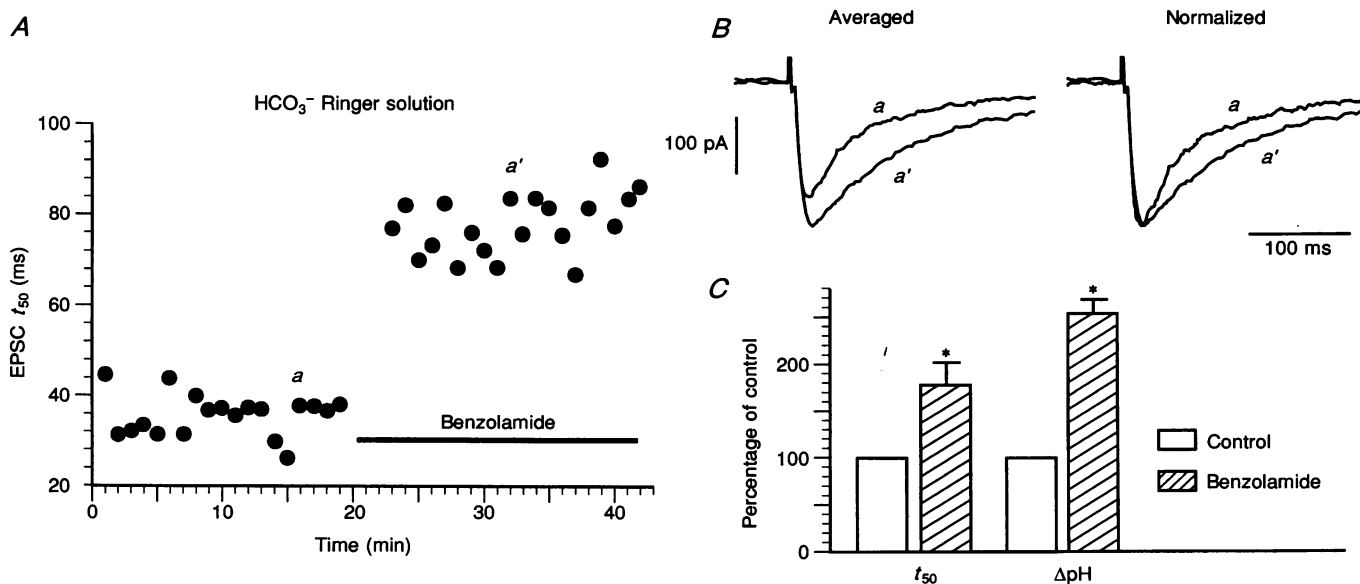
Solution	$R_N$	$E_{rev}$	$pH_o$
$HCO_3^-$	$454 \pm 106$	$+1.4 \pm 2.2$	$7.25 \pm 0.02$
+ Benzolamide	$486 \pm 91$	$-1.2 \pm 2.0$	
$HCO_3^-$ -APV	$384 \pm 41$	$+1.9 \pm 2.0$	$7.25 \pm 0.01$
+ Benzolamide	$416 \pm 51$	$-1.8 \pm 1.4$	
Hepes	$239 \pm 42$	$0.0 \pm 1.5$	$7.28 \pm 0.02$
+ Benzolamide	$290 \pm 44$	$-1.2 \pm 0.8$	

Values are means  $\pm$  s.e.m.

### Effect of benzolamide on EPSCs evoked by Schaffer collateral stimulation

In  $HCO_3^-$  Ringer solution, benzolamide ( $1 \mu M$ ) caused an immediate prolongation of the EPSCs and a simultaneous increase in the peak amplitude of the alkaline transients evoked by each stimulus. The increase in time to 50% decay ( $t_{50}$ ) far exceeded the normal scatter of this parameter, as shown in the experiment depicted in Fig. 2A. The first stimulus in which the alkaline shift was increased always corresponded to the first stimulus in which the EPSC time course was prolonged. Averages of original and normalized EPSCs at times before (*a*) and after (*a'*) benzolamide are superimposed in Fig. 2B,

highlighting the increase in decay time course. For ten cells held at  $-80$  mV, the  $t_{50}$  ( $33 \pm 7$  ms, range 14–74 ms) was prolonged by  $78 \pm 24\%$  following addition of benzolamide ( $P < 0.01$ ), with a maximum increase of 225%. This was associated with an increase in the amplitude of the alkaline transients of  $154 \pm 14\%$  (Fig. 2C). Benzolamide had no significant effect on the relaxation half-time of the alkaline shifts, which averaged  $2.7 \pm 0.3$  s before, and  $3.1 \pm 0.3$  s after, application of the drug ( $P = 0.24$ ). Similar results were observed at a holding potential of  $-40$  mV ( $n = 6$  cells), with an increase in the EPSC  $t_{50}$  of  $92 \pm 48\%$  ( $P < 0.05$ ). Neither input resistance nor EPSC reversal potential was notably affected by benzolamide (Table 1). Benzolamide caused increases in



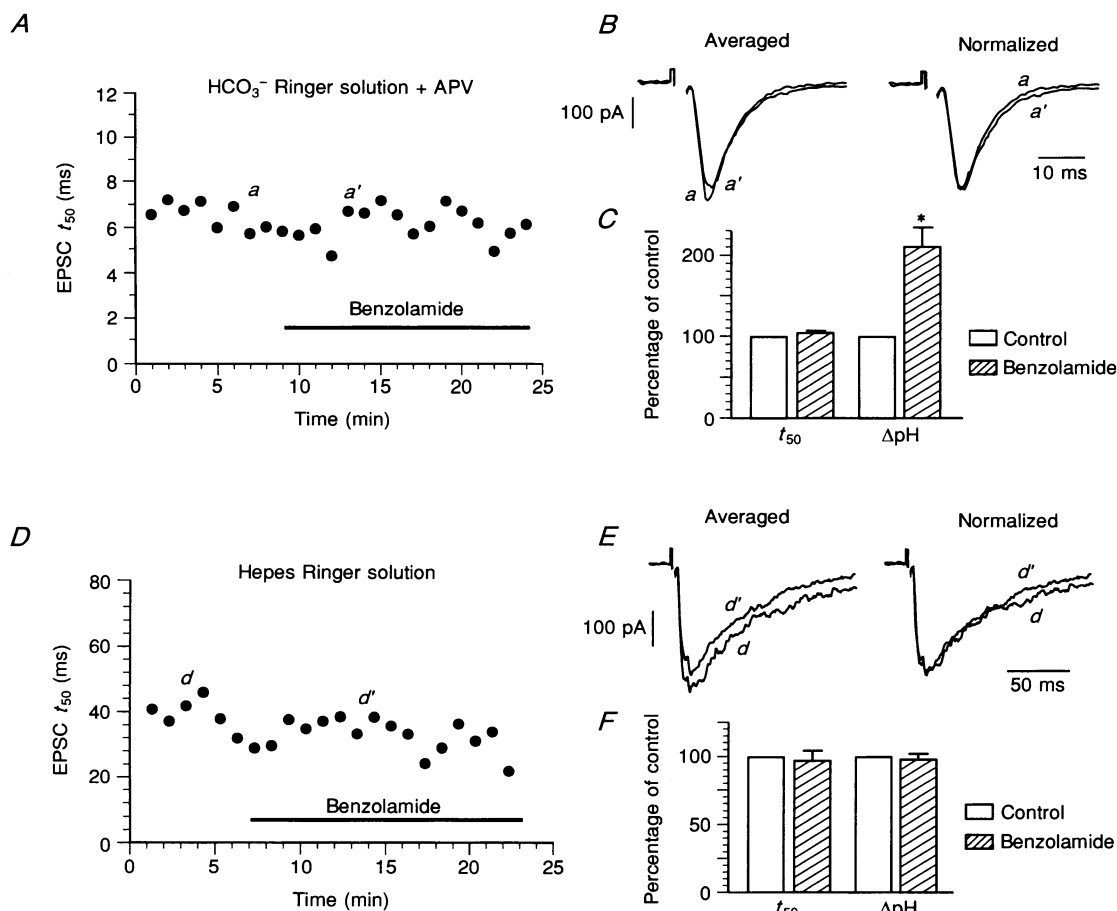
**Figure 2.** Benzolamide prolongs the EPSC decay time

A, experiment in which benzolamide ( $1 \mu M$ ) approximately doubled the EPSC  $t_{50}$ . Each dot represents the  $t_{50}$  from the mean of 3 consecutive EPSCs, evoked at 20 s intervals. Holding potential was  $-80$  mV. B, EPSCs are shown averaged ( $n = 10$ ) and normalized, corresponding to data indicated (*a*, *a'*) in A. The normalized records illustrate the prolongation of the decay time course. C, average effect of benzolamide on alkaline shifts and EPSC decay. Data are from 10 cells held at  $-80$  mV. Asterisk indicates significance ( $P < 0.01$ ).

EPSC amplitude (7 of 10 cells), as well as decreases (3 of 10 cells), with an overall change of  $+36 \pm 17\%$  that was not statistically significant ( $P = 0.13$ ). In a previous report, benzolamide had similar effects on the amplitude of extracellular field potentials, increasing them in six of eleven experiments (Taira, Smirnov, Voipio & Kaila, 1993). These inconsistent effects of benzolamide are most probably due to a mixture of pre- and postsynaptic actions. Therefore, the decay time was emphasized in the analysis of the EPSCs. In the case of NMDA receptor-mediated currents, this is strictly a postsynaptic measure that reflects the channel kinetics (Lester, Clements, Westbrook & Jahr, 1990; Hestrin, Sah & Nicoll, 1990).

The effect of benzolamide on EPSC decay was dependent on NMDA receptor activation. In the presence of the

NMDA receptor antagonist APV, the EPSCs displayed rapid, single-exponential decays, with a  $t_{50}$  of  $7.2 \pm 0.7$  ms (corresponding to a time constant of  $10.0 \pm 1.0$  ms), consistent with studies of the  $\alpha$ -amino-3-hydroxy-5-methyl-4-isoxazole propionic acid (AMPA) receptor-mediated component of the EPSC in adult CA1 neurones (Hestrin, Nicoll, Perkel & Sah, 1990). The EPSC decay time was unaffected by benzolamide (Fig. 3A and B), although the alkaline shifts were still enhanced. In eight cells, the  $t_{50}$  changed by only  $4.0 \pm 2.9\%$  ( $P = 0.17$ ), while the alkaline shifts were amplified by  $111 \pm 23\%$  (Fig. 3C). Benzolamide caused increases in the EPSC amplitude ( $n = 3$ ), as well as decreases ( $n = 5$ ), with a mean amplitude change of  $+2 \pm 15\%$  ( $P = 0.51$ ). This inconsistent effect on amplitude in the presence of APV further suggests a mixed action of



**Figure 3.** The effect of benzolamide on the EPSC time course is abolished in APV or Hepes-buffered Ringer solution

A, benzolamide ( $1 \mu\text{M}$ ) had no effect on the EPSC  $t_{50}$  in the presence of  $75 \mu\text{M}$  APV. Holding potential  $-80$  mV. B, EPSCs are shown averaged ( $n = 3$ ) and normalized, corresponding to data indicated (a, a') in A. C, average effect of benzolamide on EPSC decay and alkaline shifts in APV ( $25$ – $75 \mu\text{M}$ ). Data are from 8 cells held at  $-80$  mV. D, benzolamide ( $1 \mu\text{M}$ ) had no effect on the EPSC  $t_{50}$  in Hepes-buffered Ringer solution. E, EPSCs in Hepes Ringer solution are shown averaged ( $n = 6$ ) and normalized, corresponding to data indicated (d, d') in D. F, average effect of benzolamide on EPSC decay and alkaline shifts in Hepes. Data are from 9 cells held at  $-80$  mV.

benzolamide on pre- and postsynaptic processes. The drug had no notable effect on input resistance or EPSC reversal potential in the presence of APV (Table 1).

To ascertain whether benzolamide acted directly on the NMDA receptor or indirectly (via modification of extracellular buffering), experiments were repeated in Ringer solution buffered by 26 mM Hepes. As shown in Fig. 3*D* and *E*, benzolamide had no effect on the EPSC time course in Hepes solutions. In nine cells, the  $t_{50}$  ( $45 \pm 9$  ms) fell by  $-3 \pm 7\%$ , which was not significant ( $P = 0.31$ ). Likewise, the evoked alkaline shifts in Hepes were unaffected by benzolamide, with a change of  $-2 \pm 4\%$  (Fig. 3*F*). In Hepes media, benzolamide had no notable effect on input resistance, EPSC reversal potential or baseline extracellular pH (Table 1).

## DISCUSSION

Our data indicate that the time course of NMDA receptor-mediated synaptic currents is significantly influenced by the buffering capacity of the extracellular fluid. By contrast, the AMPA receptor-mediated component of the EPSC was unaffected. This indicated that the effect of benzolamide had a postsynaptic origin, and could not be attributed to persistent transmitter action or to changes in adequacy of the space clamp. In addition, the prolongation of the synaptic currents was not due to a direct action on the NMDA receptor, since EPSC duration was not increased in Hepes media.

It is notable that benzolamide had a similar effect on the EPSC time course at a holding potential of  $-40$  mV. Its action was therefore unrelated to any residual  $Mg^{2+}$ -dependent block of the NMDA receptor channels (Nowak, Bregestovski, Ascher, Herbet & Prochiantz, 1984) at a holding potential of  $-80$  mV. Indirect effects mediated by changes in baseline pH were also unlikely. Extracellular pH was comparable in all solutions (Table 1) and was unaffected by benzolamide. In addition, while intracellular pH was not measured, the NMDA receptor is rather insensitive to internal pH changes (Tang *et al.* 1990).

The effect of benzolamide did not require  $HCO_3^-$  *per se*, since considerable endogenous  $HCO_3^-$  is still present in Hepes-buffered media. This is because the extracellular acidosis of brain slices is almost exclusively due to the generation of carbon dioxide (Voipio & Kaila, 1993); accordingly, an extracellular tissue pH of 7.25 in Hepes-buffered media (pH 7.50) would correspond to a tissue  $CO_2$  tension of 10 mmHg and an extracellular bicarbonate concentration of 5 mM. Therefore, the failure of benzolamide to prolong the EPSCs in Hepes media cannot be attributed to the absence of  $HCO_3^-$ , but rather is due to the fast (CA-independent) buffering provided by the Hepes.

In view of the external pH dependence of the NMDA receptor (Tang *et al.* 1990; Traynelis & Cull-Candy, 1990, 1991; Vyklicky *et al.* 1990), our results are consistent with a

significant subsynaptic alkalization with a rise time of milliseconds. We emphasize that because the pH micro-electrodes have a response time of seconds (Ammann *et al.* 1981), and sample from a relatively large extracellular space, they are not capable of resolving rapid subsynaptic pH changes. Indeed, means of directly measuring pH transients in this micro-domain do not exist. The small pH shifts evoked by single shocks to the Schaffer collaterals should be viewed as highly filtered representations of the underlying extracellular pH changes, which are likely to be considerably larger. Nonetheless, the increase in amplitude of the pH transients and the prolongation of the NMDA receptor-mediated currents were well correlated. Both were augmented by benzolamide in  $HCO_3^-$ -buffered media but were unaffected in Hepes. It should be noted that the relaxation time of the alkaline shifts, having a far longer duration, was unrelated to the time course of the EPSCs. The recovery from alkalization, which required several seconds, was most probably due to slow diffusion of buffer between the interstitial spaces and the bath.

The data are consistent with a role for CA in NMDA receptor-mediated synaptic transmission. If localized to the synapse, the enzyme could govern the size and duration of postsynaptic responses. However, while extracellular CA activity has been demonstrated in brain slices (Chen & Chesler, 1992*c*; Kaila, Paalasmaa, Taira & Voipio, 1992), its concentration, isoform and distribution in brain extracellular space are not known. We note that in the absence of CA inhibition, the activity-dependent alkaline shifts would be smaller. However, alkaline transients would still be capable of modulating the NMDA receptor. This is evident during repetitive activity, where pH micro-electrodes can record extracellular alkaline shifts as large as 0.1–0.2 pH units (Chesler & Kaila, 1992).

Modulation of excitatory transmission by endogenous pH shifts may be especially pertinent to forms of long-term potentiation (LTP) that have been linked to NMDA receptor activation (Bliss & Collingridge, 1993). Experimentally, LTP is often induced by trains of high-frequency stimulation. The pH shifts so generated may influence the relationship between LTP induction and stimulus frequency. In a recent report, field potentials in hippocampal slices were potentiated several minutes after application of benzolamide (Taira *et al.* 1993), suggesting a gradual induction of LTP. The immediate augmentation of individual NMDA responses described herein may underlie these observations. Indeed, the rapid effect of benzolamide on single EPSCs suggests that  $H^+$  might serve as an extracellular signal, in a time frame relevant to synaptic transmission. The NMDA receptor, with a mid-point of its pH dependence at physiological extracellular pH (Tang *et al.* 1990; Traynelis & Cull-Candy, 1990, 1991; Vyklicky *et al.* 1990), appears well-suited to respond to such signals.

## REFERENCES

- AMMANN, D., LANTER, F., STEINER, R. A., SCHULTHESS, P., SHIJO, Y. & SIMON, W. (1981). Neutral carrier based hydrogen ion selective microelectrode for extra- and intracellular studies. *Analytical Chemistry* **53**, 2267–2269.
- BLANTON, M. G., LO TURCO, J. J. & KRIEGSTEIN, A. R. (1989). Whole cell recording from neurons in slices of reptilian and mammalian cerebral cortex. *Journal of Neuroscience Methods* **30**, 203–210.
- BLISS, T. V. P. & COLLINGRIDGE, G. L. (1993). A synaptic model of memory: long-term potentiation in the hippocampus. *Nature* **361**, 31–39.
- CHEN, J. C. T. & CHESLER, M. (1992a). Modulation of extracellular pH by glutamate and GABA in rat hippocampal slices. *Journal of Neurophysiology* **67**, 29–36.
- CHEN, J. C. T. & CHESLER, M. (1992b). Extracellular alkaline shifts in rat hippocampal slice are mediated by NMDA and non-NMDA receptors. *Journal of Neurophysiology* **68**, 342–344.
- CHEN, J. C. T. & CHESLER, M. (1992c). pH transients evoked by excitatory synaptic transmission are increased by inhibition of extracellular carbonic anhydrase. *Proceedings of the National Academy of Sciences of the USA* **89**, 7786–7790.
- CHESLER, M. (1990). The regulation and modulation of pH in the nervous system. *Progress in Neurobiology* **34**, 401–427.
- CHESLER, M. & CHAN, C. Y. (1988). Stimulus-induced extracellular pH transients in the *in vitro* turtle cerebellum. *Neuroscience* **27**, 941–948.
- CHESLER, M. & KAILA, K. (1992). Modulation of pH by neuronal activity. *Trends in Neurosciences* **15**, 396–402.
- HESTRIN, S., NICOLL, R. A., PERKEL, D. J. & SAH, P. (1990). Analysis of excitatory synaptic action in pyramidal cells using whole-cell recording from rat hippocampal slices. *Journal of Physiology* **422**, 203–226.
- HESTRIN, S., SAH, P. & NICOLL, R. (1990). Mechanisms generating the time course of dual component excitatory synaptic currents recorded in hippocampal slices. *Neuron* **5**, 247–253.
- KAILA, K., PAALASMAA, P., TAIRA, T. & VOIPIO, J. (1992). pH transients due to monosynaptic activation of GABA<sub>A</sub> receptors in rat hippocampal slices. *NeuroReport* **3**, 105–108.
- LESTER, R. A., CLEMENTS, J. D., WESTBROOK, G. L. & JAHR, C. E. (1990). Channel kinetics determine the time course of NMDA receptor-mediated synaptic currents. *Nature* **346**, 565–566.
- NOWAK, L., BREGESTOVSKI, P., ASCHER, P., HERBET, A. & PROCIANTZ, A. (1984). Magnesium gates glutamate-activated channels in mouse central neurones. *Nature* **307**, 462–465.
- TAIRA, T., SMIRNOV, S., VOIPIO, J. & KAILA, K. (1993). Intrinsic proton modulation of excitatory transmission in rat hippocampal slices. *NeuroReport* **4**, 93–96.
- TANG, C. M., DICHTER, M. & MORAD, M. (1990). Modulation of the *N*-methyl-D-aspartate channel by extracellular H<sup>+</sup>. *Proceedings of the National Academy of Sciences of the USA* **87**, 6445–6449.
- TRAVIS, D. M., WILEY, C., BOHDAN, R. N. & MAREN, T. H. (1964). Selective renal carbonic anhydrase inhibition without respiratory effect: pharmacology of 2-benzenesulfonamido-1,3,4-thiadiazole-5-sulfonamide (CL 11,366). *Journal of Pharmacology and Experimental Therapeutics* **143**, 383–394.
- TRAYNELIS, S. F. & CULL-CANDY, S. G. (1990). Proton inhibition of *N*-methyl-D-aspartate receptors in cerebellar neurons. *Nature* **345**, 347–350.
- TRAYNELIS, S. F. & CULL-CANDY, S. G. (1991). Pharmacological properties and H<sup>+</sup> sensitivity of excitatory amino acid receptor channels in rat cerebellar granule neurones. *Journal of Physiology* **433**, 727–763.
- VOIPIO, J. & KAILA, K. (1993). Interstitial P<sub>CO<sub>2</sub></sub> and pH in rat hippocampal slices measured by means of a novel fast CO<sub>2</sub>/H<sup>+</sup>-sensitive microelectrode based on a PVC-gelled membrane. *Pflügers Archiv* **423**, 193–201.
- VYKLYCKY, L., VLACHOVA, V. & KRUSEK, J. (1990). The effect of external pH changes on responses to excitatory amino acids in mouse hippocampal neurones. *Journal of Physiology* **430**, 497–517.

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