

**ELECTROPHYSIOLOGICAL STUDY OF THE MECHANISM OF  
MODULATION OF GABAERGIC SYNAPTIC CURRENTS BY  
HYDROGEN IONS**

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Hydrogen ions are the most ubiquitous modulators in biological systems. The pH level in the intra- and extracellular milieu is known to be modulated by several mechanisms including enzymes (e.g. carbonic anhydrase), pumps, exchangers and ionic channels. It has been pointed out that the  $\text{HCO}_3^-$  ions, which are transported through the GABAA receptors, are able to significantly change the pH level in the closest vicinity of the GABAA receptor macromolecule. It is known that hydrogen ions are strong modulators of GABAA receptors, but the underlying mechanism has not yet been clarified. Moreover, a systematic characterisation of the pH effect on the GABAergic synaptic currents is essentially lacking. In the present study we addressed the issue of hydrogen ion effect on the miniature inhibitory synaptic currents (mIPSCs) in cultured hippocampal neurons. The mechanism of modulation was described in terms of modulation of the postsynaptic GABAA receptors. The patch-clamp technique in the whole-cell configuration was used to record the mIPSCs. Both acidic (pH=6) and basic pH (8) diminished the amplitude of mIPSCs. Additionally, at acidic pH the averaged duration of mIPSCs was shortened. In order to explore the mechanism of the pH effect on the GABAA receptor kinetics, current responses to ultrafast GABA applications were recorded in the excised-patch configuration. The exchange time of a perfusion system was sufficient to reliably assess the receptor kinetics (complete exchange at ca. 50-80  $\mu\text{s}$ ). An analysis of current responses revealed that an increase in pH regulates both the receptor affinity and desensitization. Thus a decrease in amplitude at acidic pH is due to a decrease in the GABAA receptor affinity. On the contrary, at basic pH more receptors reach the fully bound state due to higher affinity, but a larger proportion of channels desensitize, giving rise to a smaller current response. Moreover, our kinetic studies indicate that the fastest process in the GABAA receptor gating scheme is not the opening (as assumed in previous models), but the desensitization. In conclusion, our study demonstrates that the effect of pH changes on the mIPSCs is mediated by the allosteric modulation of the GABAA receptors by hydrogen ions.

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