Endogenous GABA Activates Small-Conductance $K^+$ Channels Underlying Slow IPSCs in Rat Hippocampal Neurons

YVES DE KONINCK 1 AND ISTVAN MODY 2

1 Department of Pharmacology and Therapeutics, McGill University, Montreal, Quebec H3G 1Y6, Canada; and 2 Departments of Neurology and Physiology, UCLA School of Medicine, Los Angeles, California 90095

De Koninck, Yves and Istvan Mody. Endogenous GABA activates small-conductance $K^+$ channels underlying slow IPSCs in rat hippocampal neurons. J. Neurophysiol. 77: 2202–2208, 1997. The objective of this study was to determine the properties of $K^+$ channels activated by endogenously released transmitter under synaptic conditions. First, the levels of $\gamma$-aminobutyric acid (GABA) were depleted in hippocampal nerve endings to establish the relative contribution of endogenously released GABA to the activation of GABA A receptors mediating slow inhibitory postsynaptic currents (IPSCs). Inhibition of glutamic acid decarboxylase and GABA reuptake effectively depleted $>85\%$ of the releasable GABA pool, producing parallel reductions of GABA A and GABA B receptor-mediated IPSCs, indicating that both classes of receptors are activated synthetically by endogenously released GABA. Whole cell patch clamp recordings of stimulus-evoked slow IPSCs at potentials hyperpolarized from the potassium reversal potential were consistent with the activation of a nonrectifying $(n = 3)$ or slightly outwardly rectifying $(n = 4)$ $K^+$ conductance by the endogenously released GABA. Spectral analysis of the decay phase of GABA A/IPSCs revealed several time constants indicating complex underlying channel kinetics. Nonstationary variance analysis yielded a small unitary conductance in the range of 5–13 pS, consistent with a large number of channels activated during evoked currents. These results indicate that in granule cells of the dentate gyrus, GABA released synaptically from interneuron terminals activates an unusually small $K^+$ conductance, with no or slight outward rectification. This conductance is therefore unlike those typically reported for neuronal G protein-coupled $K^+$ channels or those activated by exogenously applied baclofen with larger, inwardly rectifying conductances.

INTRODUCTION

Several experiments suggest that $\gamma$-aminobutyric acid (GABA) is the endogenous mediator of slow, GABA A receptor-mediated inhibitory postsynaptic currents (IPSCs). For example, exogenously applied GABA activates a $K^+$ conductance with a pharmacological profile similar to that of the slow IPSC (for reviews, see Misgeld et al. 1995; Thompson 1994) and GABA uptake inhibitors enhance the amplitude and prolong the duration of GABA A inhibitory postsynaptic potentials/IPSCs (Isaacson et al. 1993; Thompson and Gähwiler 1992). Yet, activation of postsynaptic GABA A receptors generally requires stimulation of the presynaptic terminals with a stimulus intensity or frequency greater than that necessary to elicit GABA A responses (Destexhe and Sejnowski 1995; Misgeld et al. 1995; Mody et al. 1994; Thompson 1994). The larger stimulus intensities or faster stimulus frequencies may be required to $J$ activate extrasynaptic GABA A receptors (spillover of GABA) (Isaacson et al. 1993; 2) produce a sufficiently long activation of these receptors to raise the intracellular concentration of G proteins to the level required for activation of the $K^+$ channels (Destexhe and Sejnowski 1995), or 3) cause the release of another neurotransmitter or cofactor necessary for the activation of these receptors (Mody et al. 1994). To determine the relative contribution of endogenous GABA versus that of other possible endogenous agents, we studied the effects of depleting the releasable pool of GABA from interneuron terminals. This was accomplished by inhibiting the GABA synthesizing enzyme glutamic acid decarboxylase (GAD) with isonicotinic acid hydrazide (isoniazid, INH) in combination with a reuptake blocker, nipecotic acid (NIP) (Geddes and Wood 1984; Maynert and Kaji 1962; Wood and Abrahams 1971; Wood et al. 1988).

To date, little is known about the properties of the $K^+$ channels activated by endogenously released GABA, i.e., under synaptic conditions. The GABA A receptors activated by their endogenous agonist may engage channels distinct from those activated by synthetic GABA A agonists such as baclofen or may favor distinct conductance or kinetic states for the same channel (for review, see Misgeld et al. 1995). Our previous findings on synaptically elicited GABA A IPSCs have shown the activation of an outwardly rectifying $K^+$ channel (Otis et al. 1993), in contrast to inwardly rectifying conductances activated by exogenous agonists (Misgeld et al. 1995). Therefore, to determine the unitary conductance of the $K^+$ channels linked to GABA A receptors during their synaptic activation, we used nonstationary fluctuation analysis of synaptic currents (De Koninck and Mody 1994; Traynelis et al. 1993). A similar approach was used recently to derive the unit conductance of Ca$^{2+}$-dependent $K^+$ channels underlying the afterhyperpolarization of vagal motoneurons (Sah 1995). Preliminary results of this study have been published in abstract form (De Koninck and Mody 1995).

METHODS

Slice preparation

Recordings were made from granule cells of the dentate gyrus in coronal half-brain slices (400 $\mu$m thick) obtained from adult male Wistar rats (postnatal day $\approx$60; 200–400 g). Methods for the slice preparation and recordings have been described in detail previously (De Koninck and Mody 1994; Otis et al. 1993). Briefly, after pentobarbital sodium anesthesia (60 mg/kg ip), animals were decapitated, and the brain was quickly dissected and immersed for 1–2 min in cold (4°C) artificial cerebrospinal fluid (ACSF) solution. Slices were prepared with a Vibratome (Lancer Series 1000), were hemisected, and stored oxygenated at 32°C in a storage chamber until individually transferred to the recording chamber (34–35°C) at the start of an experiment.
The ACSF contained (in mM) 126 NaCl, 2.5 KCl, 2 CaCl$_2$, 2 MgCl$_2$, 26 NaHCO$_3$, 1.25 NaH$_2$PO$_4$, 10 glucose, 0.04 d-2-amino-5-phosphonooxalic acid (d-AP5), and 0.01 6-cyano-7-nitroquinoline-2,3-dione (CNQX). For noise analysis experiments, in which only monosynaptic GABA$_B$ IPSCs were isolated, 10 mM picrotoxin was added (Davies et al. 1990). The remaining current could be completely and reversibly blocked by addition of CGP 35348 (200–800 µM; Ciba-Geigy), as previously described (Otis et al. 1993). The solutions were continuously bubbled with 95% O$_2$-5% CO$_2$ (pH 7.35 ± 0.05, mean ± SE). Drugs such as picrotoxin (10 µM), bicuculline (10–50 µM), INH (10 mM), and NIP (2 mM) were added to the ACSF. All chemicals were purchased from Sigma except CNQX and d-AP5 (Tocris Neuramin).

INH has been used to inhibit GAD activity in the brain and from synaptosomal preparations (Casey and Wood 1973; Geddes and Wood 1984; Maynert and Kaji 1962; Wood and Kurylo 1984; Wood and Peesker 1972; Wood et al. 1988), preferentially to other GAD synthesis inhibitors (Wood and Abrahams 1971). INH specifically reduced GABA levels from synaptosomes with accompanying increase in glutamate levels, as expected from block of GAD activity, and had no effect on the levels of other amino acids (aspartate, glutamine, taurine) (Geddes and Wood 1984; Wood and Kurylo 1984). Finally, the convulsant action of INH was shown to be specifically due to its effect on GABA metabolism and not on that of glutamate or any other amino acids (Geddes and Wood 1984).

NIP was used to prevent the reuptake of GABA that would have impaired the ability of INH to deplete GABA from the terminals (Wood et al. 1988).

Recordings
Whole cell voltage-clamp recordings were obtained with the use of borosilicate glass capillaries with an inner filament (KG-33, 1.12 mm ID, 1.5 mm OD, Garner Glass) pulled to 1.5- to 3-µm outer tip diameters (0.5–1 µm diameter of the lumen) with the use of a two-stage vertical Narishige PP-83 puller. Intracellular solution consisted of (in mM) 140 potassium gluconate, 10 N-2-hydroxyethylpiperazine-N'-2-ethanesulfonic acid, 2 MgCl$_2$, 2 MgATP, and 0.2 tris (hydroxymethyl) aminomethane–guanosine 5'-triphosphate, pH titrated to 7.2 with KOH, total osmolality 255–285 mosM.

Postsynaptic currents were evoked with the use of bipolar stimulating electrodes that were placed in the middle of the molecular layer of the dentate gyrus, 1–2 mm from the recording site. Constant current stimulus intensities ranging from 100 to 400 µA and 50–200 µs in duration reliably evoked GABA$_A$ currents in most granule cells. As reported previously (Otis et al. 1993), no differences in the kinetics of the GABA$_A$ current or in its sensitivity to antagonists were seen for different stimulus intensities.

Data analysis and curve fitting
All analog DC signals were digitized at 44 or 88 kHz (Neurocorder, NeuroData) and stored on a videotape for later analysis. Off-line, the recordings were low-pass filtered at 2 kHz (~3 dB, 8-pole Bessel, Frequency Devices 9002), sampled at 10 kHz, and analyzed with the use of software developed by Y. De Koninck as described previously (De Koninck and Mody 1994; Otis et al. 1993).

For experiments comparing the effects of INH plus NIP on the GABA$_A$ versus GABA$_B$ responses, to be able to quantify the effect of GABA depletion on both components in the same cell, we used a subtraction protocol (Fig. 1). Because a delay occurs before the rising phase of the GABA$_A$ IPSC (Otis et al. 1993), a single exponential was fit to the initial decay phase of the response to estimate the decay kinetics of the GABA$_A$ component. The extrapolated decay was subtracted from the remaining part of the response to isolate the late GABA$_B$ component (Fig. 1C). Similarly, the GABA$_A$ component was fitted with a fourth-power exponential activation and double-exponential decay kinetics (Otis et al. 1993) of the form

$$I(t) = A \cdot (1 - e^{-t/\tau_a})^\nu \cdot (A_i \cdot w_{h,i} \cdot e^{-t/\tau_{h,i}} + A_i \cdot w_{r,i} \cdot e^{-t/\tau_{r,i}})$$

where $I(t)$ is current as a function of time, $w_{h,i}$ and $w_{r,i}$ (where $A = A_i \cdot w_{h,i} + A_i \cdot w_{r,i}$) are weighting factors, $\tau_a$ is the time constant of the activation parameter, and $\tau_{h,i}$ and $\tau_{r,i}$ are the time constants of the inactivation parameters (Otis et al. 1993). Curves were fitted by a least-square method based on a simplex algorithm (De Koninck and Mody 1994).

Nonstationary spectral and variance analysis
For noise analysis experiments, monosynaptic GABA$_A$ IPSCs were studied in isolation (10 µM picrotoxin added). The technique used for the nonstationary noise analysis has been described in detail previously (De Koninck and Mody 1994). Briefly, with each cell, 50–300 postsynaptic currents were used for the noise analysis. For the variance analysis, a segment of a fixed length was selected from the peak of the current to past the end of the decay. The amplitude of the current was divided into bins (30–40) of equal size on the amplitude scale. Within each bin, the variance of the IPSC around the scaled average was computed and plotted against the mean current value within the scaled average current bin. The data were fitted with the relationship $\sigma^2 = I_{m0} - \bar{I}_m^2/N$, where $\sigma^2$ is the variance, $I_m$ is the mean current, and $i$ is the unitary current. For the spectral analysis, the average current was subtracted from each IPSC after scaling. Segments of 1,024–2,048 points (depending on the sampling frequency; chosen to be 5–10 times longer than the decay time constant of the ensemble average) were Parzen windowed and the one-sided power spectrum was computed by fast Fourier transform (De Koninck and Mody 1994). The power spectrum was fitted with the sum of $n$ Lorentzian functions of the form

$$L(f) = \sum_{i=1}^{n} S_i (1 + (f/f_c)^2)$$

where $L(f)$ is the spectral density at frequency $f$, $S_i$ is the power of the spectrum at $f = 0$, and $f_c$ is the cutoff frequency at which the spectral power is half. The corresponding time constants ($\tau_i$) are derived with the relationship $\tau_i = 1/2\pi f_c$. The resulting power of the spectrum was not corrected for the power of the mean time course (Sigworth 1981), because the focus was only on evaluating the corner frequencies; therefore no attempt was made to derive the single-channel conductance from the power of the spectrum.

The goodness of fit was evaluated on the basis of fitting subsets of points drawn from the whole set of data points as well as from evaluation of the reduced $\chi^2$ ($\chi^2_{red}$)

$$\chi^2_{red} = \frac{\chi^2}{\nu}$$

where the factor $\nu = N - n$ is the number of degrees of freedom left after fitting $N$ data points to the $n$ parameters. The necessity of introducing additional Lorentzian components to the fits was judged first on the basis of visual inspection of the fitted curves superimposed onto the data. When the merit of additional components was not obvious, an $F$ test was used to assess how the additional component improved the value of the reduced $\chi^2$

$$F_{\chi^2} = \frac{\Delta \chi^2}{\nu}$$

The $F$ follows the $F$ distribution with $df_1 = 1$ and $df_2 = \nu$. The critical value for the merit of additional components was set at a low level ($P < 0.001$) to favor parsimony of the fitted function (De Koninck and Mody 1994).
FIG. 1. Isolation of γ-aminobutyric acid-A (GABA_A) and GABA_B components for quantitative measurements. A: in the presence of 6-cyano-7-nitroquinolinic acid-2,3-dione (CNQX) and d-2-amino-5-phosphonovaleric acid (d-AP5), electrical stimulation in the molecular layer of the dentate gyrus produces a mixed GABA_A and GABA_B receptor-mediated inhibitory postsynaptic current (IPSC). B: the 2 IPSCs can be distinguished on the basis of their time course and reversal potential. We used a subtraction protocol (Otis et al. 1993) to properly quantify the area corresponding to each component. C: according to our previous studies (Otis et al. 1993), there is a delay before in the rising phase of the GABA_B IPSC. Thus the initial phase of the IPSC consists only of a GABA_A component. We can therefore fit a single exponential to the early decay phase of the 1st component and subtract the extrapolated curve from the remaining part of the response to isolate the late GABA_B component. Similarly, the GABA_A component can be fit with a 4th-power exponential activation and double-exponential decay kinetics. Subtraction of the fitted curve from the original trace effectively isolates the GABA_A component.

RESULTS
Depletion of releasable GABA

INH (10 mM), an inhibitor of the pyridoxine cofactor of GAD (Wood et al. 1988), and NIP (2 mM), a GABA uptake blocker, were perfused onto the slices (Fig. 2). In all seven slices tested, multiple population spikes characteristic of a diminished GABAergic inhibition appeared within 45–60 min of perfusion (Fig. 2A). These epileptic field potentials were further enhanced by only ~15% on block of postsynap-
Depletion of the releasable GABA pools depresses both GABA<sub>A</sub>- and GABA<sub>B</sub>-mediated inhibitions. A: isoniazid and nipecotic acid depress GABA<sub>A</sub> receptor-mediated inhibition of field responses. As the drugs were perfused onto the slices, multiple population spikes characteristic of a diminished GABAergic inhibition appeared. The epileptic fields were enhanced by only 18 and 14% on block of postsynaptic GABA<sub>A</sub> receptors with picrotoxin (PTX) (10 μM) or bicuculline (BMI) (50 μM), respectively. The field responses were either quantified by measuring the area under the curve or with the use of a contour line (not shown) indicator. Similar results were obtained with both methods. B: isoniazid and nipecotic acid depress GABA<sub>A</sub>- and GABA<sub>B</sub>-mediated IPSCs, but not responses to exogenous baclofen. Each trace represents the average of 5 responses obtained by stimulating regularly at 30-s intervals. In the absence of drugs, this type of stimulation routinely yielded stable recordings lasting for several h without any rundown. The traces in Ba and Bb represent the subtracted GABA<sub>A</sub> and GABA<sub>B</sub> components, respectively (see Fig. 1C). Most of the evoked responses were typically abolished within <1 h, yet responses to exogenously applied baclofen and/or GABA always persisted, confirming that the neurons did not deteriorate and continued to respond to GABA<sub>B</sub> receptor activation. The holding potential was −60 mV. C: depressions of GABA<sub>A</sub> and GABA<sub>B</sub> IPSCs follow a parallel time course. Responses were quantified as the area under the curve for each isolated component (GABA<sub>A</sub> and GABA<sub>B</sub>, respectively) (points represent means ± SE, obtained from 4 slices).
tic GABA\textsubscript{A} receptors with picrotoxin (100 \textmu M) or bicuculine (100 \textmu M) \cite{Fig. 2, Ab and Ac}. Similar values were obtained whether measurements of area or contour line (not shown) were used, consistent with reduced GAD activity and GABA uptake effectively depleting \(>85\%\) of the releasable GABA pool.

We then investigated GABA\textsubscript{A} and GABA\textsubscript{B} receptor-mediated monosynaptic IPSCs in whole cell recordings during the depletion of GABA. Consistent with the above findings, INH plus NIP produced a parallel reduction in the size of GABA\textsubscript{A} IPSCs by 96\% and GABA\textsubscript{B} IPSCs by 81\% \cite{Fig. 2, B and C}. In separate experiments, CGP 35348 completely blocked GABA\textsubscript{B} IPSCs (not shown). Responses to exogenous application of baclofen persisted after ablation of the synaptic responses. Taken together, these results show that the rundown of the evoked GABA-mediated responses was due to effective depletion of GABA levels from presynaptic terminals and not to the toxicity of the drugs.

Conductance properties of K\textsuperscript{+} channels underlying the GABA\textsubscript{B} IPSCs

Experiments related to conductance measurements, current-voltage relationships, and nonstationary noise analysis were performed in control slices on pharmacologically isolated (10 \textmu M picrotoxin; 10 \textmu M CNQX; 40 \textmu M d-AP5) monosynaptic GABA\textsubscript{A} IPSCs.

As reported previously \cite{Otis et al. 1993}, current-voltage relationships were linear \((n=9)\) in the range of \(-45\) to \(-110\) mV when the extracellular K\textsuperscript{+} concentration was 2.5 mM. With an extracellular K\textsuperscript{+} concentration of 6.8 mM, a small outward rectification was observed in four of seven cells tested (no rectification in the remaining 3), with a slope conductance for the outward portion of 2.69 \pm 0.52 nS vs. 1.52 \pm 0.16 nS for linear current-voltage plots (not shown).

To obtain information on the underlying single-channel conductance and kinetics, we performed spectral and variance analysis on the decay phase of evoked GABA\textsubscript{B} IPSCs. Ensemble averages (100–300 events) were subtracted from individual traces \cite{Fig. 3A} and noise analysis was performed on the difference trace as previously described \cite{De Koninck and Mody 1994}.

Spectral analysis on the decay phase of evoked current \((n=3)\) revealed four major corner frequencies corresponding to time constants of 140 \pm 52 ms, 46.2 \pm 12.4 ms, 6.95 \pm 2.15 ms, and 0.85 \pm 0.24 ms at 35\°C \cite{Fig. 3B}. This analysis resolved more time constants than the simple exponential curve fitting of the decay phases of the currents. The shortest corner frequency corresponds to the shorter of the two decay time constants \((110.2 \pm 6.8\) ms). The very long decay time constant described in our previous study \((516.2 \pm 52.5\) ms; 16\% fraction) is likely to fall outside the resolution of the spectral analysis given the length of the samples used \cite{Fig. 3A}. Similarly, this will tend to yield an overestimation of the first corner frequency. Nevertheless, the additional corner frequencies revealed by the spectral analysis likely indicate complex intraburst kinetics of the underlying K\textsuperscript{+} channels such as those observed for the G protein-coupled single K\textsuperscript{+} channels in locus coeruleus neurons \cite{Grigg et al. 1996}.

Nonstationary variance analysis \((n=5)\) yielded an average estimate for the single-channel conductance \((\gamma)\) of 8.6 \pm 2.8 pS (range 5.2–12.6 pS) \cite{Fig. 3C}. Accordingly, a large number of these channels \((177 \pm 51)\) was estimated to contribute to the peak of the evoked GABA\textsubscript{A} IPSCs \cite{Fig. 3C}. Comparable estimates were obtained in two cells at room temperature (22\°C).

**DISCUSSION**

Endogenous GABA as a mediator of both GABA\textsubscript{A} and GABA\textsubscript{B} IPSCs

The results obtained after depletion of GABA from inhibitory terminals indicate that both GABA\textsubscript{A} and GABA\textsubscript{B} receptors are activated synthetically by endogenously released GABA. It establishes that GABA is a necessary transmitter in the response, yet it does not completely rule out the possibility that another cotransmitter (albeit not sufficient by itself) participates in GABA\textsubscript{B} responses.

In the depletion experiments, the attenuation of the GABA\textsubscript{A}-mediated IPSC paralleled, with a slower time course, the decline of the GABA\textsubscript{B}-mediated response. This may reflect a greater affinity of the GABA\textsubscript{B} receptor for GABA \cite{Isaacson et al. 1993}. Alternatively, it may be an indication that the GABA\textsubscript{B} response involves another endogenous agent. As stressed above, this would indicate that although GABA is a necessary transmitter, it may not be sufficient for the activation of synaptic GABA\textsubscript{B} receptors. The data are, however, insufficient to establish that this difference in time course is statistically significant.

Properties of K\textsuperscript{+} channels linked to synaptic GABA\textsubscript{B} receptors

The properties and the relative number of postsynaptic K\textsuperscript{+} channels linked to GABA\textsubscript{B} receptors activated during slow IPSCs were studied with the use of nonstationary fluctuation analysis. We have determined that these receptors are linked to small-conductance (5–12 pS) K\textsuperscript{+} channels with no or slight outward rectification. A large number of these channels is activated during an average-sized stimulus-evoked GABA\textsubscript{B} IPSC.

The GABA\textsubscript{B} receptors are coupled through peritussin toxin-sensitive G proteins to K\textsuperscript{+} channels \cite{Thalmann 1988}. The small-conductance channels reported here are unlike the more commonly reported G protein-linked K\textsuperscript{+} channels with larger, inwardly rectifying conductances \cite{Brown and Birnbaumer 1990; Doupnik et al. 1995}. Nevertheless, small-conductance (13 pS) G protein-activated K\textsuperscript{+} channels have been described in hippocampal neurons \cite{VanDongen et al. 1988}, and recently a small unit conductance has been resolved for apamin-insensitive Ca\textsuperscript{2+}-dependent K\textsuperscript{+} channels with the use of nonstationary noise analysis \cite{Sah 1995}.

Two previous studies have addressed the unitary conductance of K\textsuperscript{+} channels linked to GABA\textsubscript{B} receptors. Both used cell-attached recordings to measure single-channel activity in response to exogenous applications of baclofen \cite{Wagner and Dekin 1993} and/or GABA \cite{Premkumar et al. 1990} and reported the opening of large-conductance (70–100 pS) channels. Although the channels observed by Premkumar et al. \cite{1990} had a maximum conductance of \(\sim 70\) pS, they also displayed many lower conductance states that were integral multiples of \(5–6\) pS, and in several cells the single-channel
currents became progressively larger by multiples of an elementary current of 5–6 pS. This small elementary current may correspond to the main conductance state activated during the synaptic events recorded in our study.

In previous studies researchers used nonstationary noise analysis to estimate the conductance linked to non-N-methyl-D-aspartate receptors and GABA_A receptors activated at synapses. Interestingly, in those cases it was found that the larger of the multiple conductance states revealed in single-channel recordings appeared to carry most of the synaptic current (De Koninck and Mody 1994; Traynelis et al. 1993). On the basis of our results and those of Premkumar et al. (1990), it would appear that a small-subconductance state predominates during synaptic GABA_B currents. The possibility has to be considered, however, that the unit conductance was underestimated by the nonstationary noise analysis. Synaptic currents and estimates from noise analysis are attenuated by filtering due to the electrotonic properties of the cell (De Koninck and Mody 1994; Traynelis et al. 1993). Calculations based on electrotonic properties of cerebellar granule cells revealed that the values of single-channel conductance derived from noise analysis of filtered excitatory postsynaptic potentials may have been attenuated by up to 10%. In our recent study of GABA_A receptor-mediated miniature IPSCs, our worst case scenario of filtering for most distant IPSCs (Soltesz et al. 1995) yielded a possible attenuation of up to 25% in the unit conductance measured with noise analysis. Applying this factor to the present case would mean that our average value of 8.6 pS (range 5.2–12.6) would correspond to 11.4 pS (range 6.9–16.8). This value remains in the range of that reported for small G protein-activated K^+ conductances and is almost 1 order of magnitude smaller than for the more commonly reported G protein-linked K^+ channels. The small unit conductance linked to synaptically activated GABA_B receptors measured in our study would either indicate that channels activated...
in these conditions are different from those stimulated by exogenous agonists or that it is a subconductance state that preferentially participates in synaptic currents. Distinct K⁺ conductances may also be associated with GABAᵦ receptors in the different cell types studied.

In summary, GABAᵦ receptors activated by endogenous transmitter are linked to small-conductance K⁺ channels with no or small outward rectification. These channels appear to have different properties than those stimulated by exogenous agonists, thus raising the possibility that the receptors or the K⁺ channels activated by synaptically released GABA are distinct from those activated experimentally by exogenous applications of agonists or that a small-subconductance state is the predominant charge carrier during synaptic GABAᵦ currents.

We thank Dr. T. S. Otis for participating in some of the experiments. This work was supported by National Institutes of Health Grants NS-30549 to I. Mody, NS-34022 To Y. De Koninck, and MT-12942 to Y. De Koninck from the Canadian Medical Research Council. Y. De Koninck is a Scholar of the Canadian MRC.

Address for reprint requests: Y. De Koninck, Dept. of Pharmacology and Therapeutics, McGill University, 3655 Drummond St., Rm. 1242, Montreal, Quebec H3G 1Y6, Canada.

Received 8 August 1996; accepted in final form 9 December 1996.

REFERENCES


