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Synaptic response patterns of neurons in the cortex of rat inferior colliculus

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Abstract

The present study examined synaptic potentials of neurons in inferior colliculus (IC) cortex slice and the roles of GABA and glutamate receptors in generating these potentials. Multipolar (82%) and elongated (18%) cells were observed with intracellular biocytin staining. Electrical stimulation of the IC commissure (CoIC) elicited only inhibitory postsynaptic potentials (IPSPs) (10% of cells), only excitatory postsynaptic potentials (EPSPs) (51%), or both (38%). IPSPs were elicited at lower thresholds and shorter latencies than EPSPs (mean: 1.6 ± 1.2 ms) and IPSPs were observed in all neurons following membrane depolarization. Short-latency EPSPs were blocked by non-NMDA receptor antagonists, and longer-latency EPSPs were blocked by NMDA antagonists. CoIC stimulation evoked short-latency IPSPs (mean: 0.55 ± 0.33 ms) in 48% of neurons, and the IPSPs persisted despite glutamate receptor blockade, which implies monosynaptic inhibitory input. A $GABA_A$ antagonist blocked IPSPs and paired pulse inhibition of EPSPs, suggesting $GABA_A$ receptor mediation. A $GABA_B$ antagonist reduced paired pulse inhibition of IPSPs, suggesting $GABA_B$ receptor modulation. Thus, GABA-mediated inhibition plays a critical role in shaping synaptic responses of IC cortex neurons. Normal GABAergic function in IC has been shown to be important in acoustic coding, and reduced efficacy of GABA function in IC neurons is critical in IC pathophysiology in presbycusis, tinnitus and audiogenic seizures.

Key words: Inferior colliculus; Neuron; Slice; Synaptic response

1. Introduction

The inferior colliculus (IC) is a critical structure for processing auditory information and receives a robust array of ascending and descending synaptic auditory projections (Herbert et al., 1991; Saldaña and Merchán, 1992; Covey et al., 1996; Kuwada et al., 1997). The IC is subdivided into the central nucleus (ICc) and cortices that surround the ICc (Faye-Lund and Osen, 1985; Oliver and Shneiderman, 1991). The ICc receives the major ascending auditory input to the IC and is important in processing and coding auditory information (Caird, 1991; Palombi and Caspary, 1996a). The IC cortices are part of the extralemniscal auditory system. The major ascending inputs to IC cortex neurons arise from cells in the ICc and the contralateral IC (Coleman and Clerici, 1987; Saldaña and Merchán, 1992), and descending inputs from auditory cortex are also well described (Coleman and Clerici, 1987; Herbert et al., 1991; Feliciano and Potashner, 1995), with some inputs projecting through the commissure of the IC (CoIC) (Faye-Lund, 1985; Oliver and Huerta, 1992). The interconnections between the IC subdivisions appear to be extensive, both ipsilaterally and contralaterally via commissural connections (Coleman and Clerici, 1987; Saldaña and Merchán, 1992).

Numerous papers have investigated the neurophysiology of IC in the intact animal, primarily using extracellular recording techniques, and much has been learnt about the function of these neurons (Nelson and Erulkar, 1963; Caird, 1991; Covey et al., 1996; Palombi and Caspary, 1996a,b; Kuwada et al., 1997). In vitro

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approaches are being used to study the details of the physiology of IC neurons that are difficult to examine in vivo where intracellular techniques are possible but difficult to perform (Kuwada et al., 1997; Pedemonte et al., 1997). Previous in vitro studies have provided details about the intrinsic properties of IC neurons in the cortices (Smith, 1992; Li et al., 1998) and the ICc (Wagner, 1994; Hosomi et al., 1995; Li et al., 1998) in slice and in culture (Hosomi et al., 1997). The synaptic properties of IC neurons have been examined to a lesser extent (Smith, 1992; Wagner, 1996) and the roles played by specific neurotransmitters in generation of these properties have only begun to be examined systematically (Yamauchi and Amatsu, 1989; Pierson et al., 1989; Smith, 1992; Hosomi et al., 1997; Moore et al., 1998).

A wide variety of approaches have strongly implicated GABA as the major inhibitory neurotransmitter in the IC (Glendenning and Baker, 1988; Faingold et al., 1989a, 1991a,b; Oliver et al., 1994; Goldsmith et al., 1995; Milbrandt et al., 1996). A number of reports have also implicated glutamate as a major excitatory neurotransmitter in IC neurons (Banay-Schwartz et al., 1989; Najlerahim et al., 1990; Faingold et al., 1991a; Feldman and Knudsen, 1994; Feliciano and Potashner, 1995; Gaza and Ribak, 1997; Saint Marie, 1996; Caicedo et al., 1998). Pharmacological modifications of the actions of these neurotransmitters significantly alter acoustic responses of IC neurons in vivo (Faingold et al., 1989a, b; Feldman and Knudsen, 1994; Klug et al., 1995; Fuzessery and Hall, 1996; Le Beau et al., 1996; Palombi and Caspary, 1996b; Mori, 1997). The actions of these amino acid neurotransmitters on IC neurons appear to be a major target for acoustically-related pathological conditions. The purpose of the present study was to extend the evaluation of the synaptic properties of IC cortex neurons and examine the roles of GABA and glutamate receptors in the generation of these synaptic potentials using in vitro brain slice techniques.

2. Materials and methods

Adult Sprague-Dawley rats weighing 250–350 g were deeply anesthetized with halothane and decapitated. The brain was quickly removed and immersed in cold artificial rat cerebrospinal fluid (ACSF) bubbled with 95% O_2 and 5% CO_2. A block of tissue containing IC was made with coronal cuts just rostral to the superior colliculus and in the midportion of the cerebellum. This block was mounted with cyanoacrylate glue, cerebellum end up, and sliced into 400 μm thick coronal sections using a vibrating tissue slicer (Frederick Haer). Slices were kept in an incubation chamber at room temperature for at least 30 min prior to transfer to a recording chamber. In the incubation and recording chambers, slices were bathed in ACSF with the composition (in mM) NaCl 117.4, KCl 2.0, $MgSO_4$ 1.4, $CaCl_2$ 2.5, KH_2PO_4 1.0, $NaHCO_3$ 26.2, glucose 11.0. The ACSF was continually bubbled with 95% O_2 and 5% CO_2. The volume of the recording chamber was approximately 0.6 ml and the ACSF flow rate was 4 ml/min. The brain slice was completely submerged in ACSF. The temperature in the recording chamber was maintained at 35°C. Drugs were applied by superfusion, with the necessary solution changes being made by means of three-way valves so that the perfusion rate did not change. The 6-cyano-7-nitroquinoxaline-2,3-dione (CNQX, 0.1–30 μM) and 6,7-dinitroquinoxaline-2,3-dione (DNQX, 0.1–30 μM) were obtained from Research Biochemical, phaclofen (10–30 μM) from Tocris Cookson, and *N*-methyl-D-aspartate (NMDA, 10–30 μM), bicuculline (10–30 μM) and electrolytes from Sigma.

The IC cortex was located visually according to the atlas of Paxinos and Watson (1986) and descriptions of Faye-Lund and Osen (1985) (Fig. 1). The three IC subdivisions: dorsal cortex (ICd), external cortex (ICx) and the ICc, are visible in the unstained transilluminated slice. Intracellular recording electrodes were pulled from glass pipettes with an outer diameter of 1.0 mm and inner diameter of 0.58 mm (A and M systems) on a Flaming/Brown P87 micropipette puller (Sutter Instrument) and filled with 4.0 M potassium acetate. Electrode impedances ranged from 80 to 110 MΩ. Intracellular potentials were recorded with an Axoclamp 2A preamplifier (Axon Instruments), low-pass filtered at 3–10 kHz, and digitized for storage on a microcomputer, or displayed on a storage oscilloscope and photographed. Bridge balance was carefully monitored and adjusted when necessary. The intracellular electrodes passed ±1.0 nA of current without significant rectification. Each electrode was tested before use and was discarded if its electrical characteristics were unsuitable. Capacity compensation was adjusted to just below the maximum obtainable without oscillation. Synaptic responses were elicited by placing a bipolar stimulating electrode (10NE-200×50 mm, Rhodes Electronics) in the fibers of the CoIC at the midline. Stimuli were 0.1 ms duration constant-voltage square waves (Grass S44 stimulator and SIU5 stimulus isolation unit). The pClamp series of programs (Axon Instruments) were used for data acquisition and off-line analysis of digitized data.

The morphology of some neurons was studied using intracellular biocytin. For these experiments, the intracellular electrode was filled with a solution containing potassium acetate and biocytin (2%, Sigma). After examining neuronal response characteristics, biocytin was iontophoresed into the neuron by passing positive cur-

rent (0.2–1.0 nA current pulses, 200 ms on and 200 ms off for 5 to 10 min). The slice was allowed to sit in the recording chamber for 30 min to 2 h so that biocytin could completely fill the cell. Only one cell was labeled in each slice. The position of the injection was marked on a sketch of the IC slice. The slice was then fixed overnight in 4% paraformaldehyde with 0.2% picric acid. After fixation, the slices were embedded in a 2.5% solution of agar dissolved in 10% formaldehyde and cut into 60 μm sections. The sections were rinsed in 0.1 M phosphate buffer and incubated in 0.5% hydrogen peroxide for 20 min followed by incubation in Triton-X 100 for 1 h and incubated overnight with an avidin-conjugated horseradish peroxidase solution (Vector ABC immunolabeling kit). They were rinsed in phosphate buffer and reacted with diaminobenzidine using nickel-cobalt intensification. After completion of the staining procedure, the diluted diaminobenzidine working solution was neutralized in distilled water with an equal volume of 3% potassium permanganate and 2% sodium carbonate. The sections were cleared and mounted without further processing, and the location of the cells was within the external and dorsal cortices, as seen in Fig. 1.

The care and use of animals in this study were approved by the Laboratory Animal Care and Use Committee of Southern Illinois University School of Medicine and the National Institutes of Health.

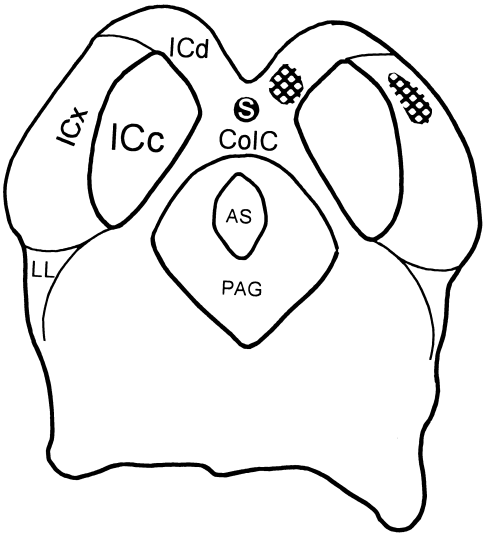

Fig. 1. IC slice. This diagram indicates the major landmarks visible in transilluminated slice, including the ICc, ICx, ICd and CoIC as well as the lateral lemniscus (LL), periaqueductal gray (PAG), and aqueduct of Sylvius (AS). The location of our bipolar stimulating electrode in the CoIC is indicated (circled 's'). The cross-hatched areas in the diagram are the approximate areas from which we recorded.

3. Results

3.1. Recording locations in the IC slice

A diagram of the IC (Fig. 1) shows the transverse slice of the IC and the landmarks that are visible in the transilluminated preparation. The range of recording sites in the ICx, ICd and the stimulation site in the CoIC are shown. The cross-hatched areas in the diagram are the approximate areas from which we recorded.

3.2. Cellular morphology

The neurons studied had two different morphologies. Most had a multipolar shape ($n = 23$, Fig. 2A, B) but a few were elongated cells ($n = 5$, Fig. 2C, D). Multipolar cells had several dendrites branching from the soma, and these had secondary branches that extended over a large area of the IC cortex, several hundred microns in diameter, but contained within a single subdivision. The mean ($n = 23$) soma dimensions were 15.4 by 23.2 μm. Neurons with a multipolar morphology were found in both external and dorsal cortices. The morphology of elongated cells was different. Elongated cells had only two or three major dendrites and these were oriented in a dorsal-ventral direction perpendicular to the fibers of the CoIC. Their mean ($n = 5$) soma dimensions were 14.5 by 25 μm ($n = 3$). This cell type was found only in the ICd.

3.3. Synaptic responses to CoIC stimulation

We found that the synaptic properties of neurons in ICd ($n = 56$) and ICx ($n = 48$) were similar, including response latencies (dorsal, 1.69 ± 0.46 S.E.M., $n = 10$; external 1.78 ± 0.54, $n = 10$). Therefore, the data from these two regions have been combined for analysis. Electrical stimulation of the CoIC elicited short-latency hyperpolarizing inhibitory postsynaptic potentials (IPSPs) and excitatory depolarizing postsynaptic potentials (EPSPs) in IC cortex neurons. Response latencies were measured from the stimulus onset to the initial deflection of each potential of the response as the potential became depolarized or hyperpolarized with respect to the resting membrane potential. When synaptic potentials were elicited at resting membrane potential, several different synaptic response patterns were seen. Because the inputs to these neurons in the in vitro preparation are artificially limited due to the brain slicing procedure, the classification of response is only to the input from the intact CoIC fibers and cannot be considered characteristic of the underlying response patterns of these neurons in vivo. Some neurons exhibited both inhibitory responses and excitatory synaptic responses to CoIC stimulation (I/E pattern, Fig. 3) and

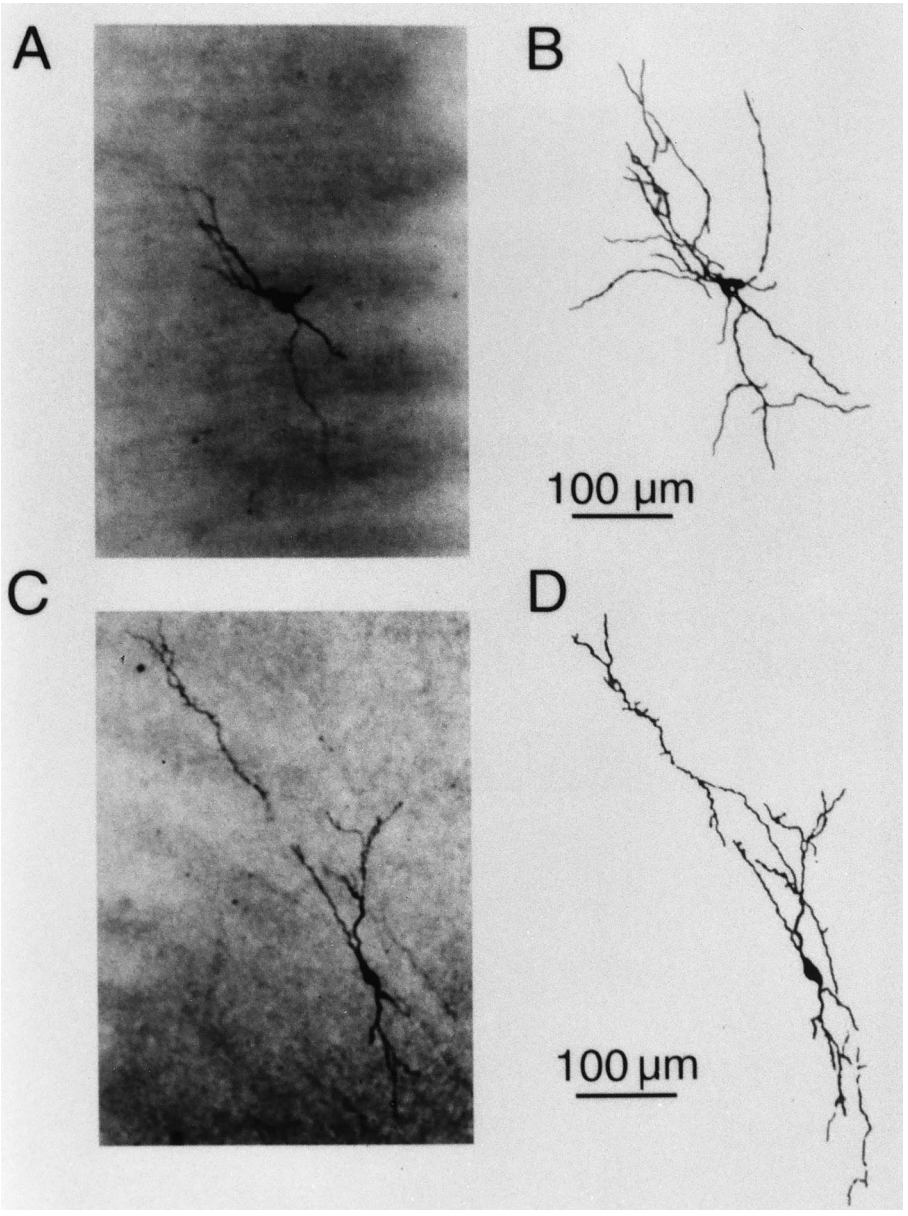

Fig. 2. Biocytin-stained IC cortex neurons had either a multipolar or an elongated morphology. A: Most (82%) neurons in dorsal and external cortices had a multipolar shape. These neurons have several dendrites with secondary branches extending over a large area of the IC cortex but contained within a single IC subdivision. B: The camera lucida drawing of this neuron is shown. C: The remainder of neurons (18%) were elongated, having two or three major dendrites oriented in a dorsal-ventral direction perpendicular to the fibers of the CoIC. D: The camera lucida drawing of this neuron is shown. The latter cell type was observed only in dorsal cortex.

we have referred to each element as an IPSP or EPSP rather than the hyperpolarizing or depolarizing post synaptic response for simplicity. Other neurons had only EPSPs (E pattern, Fig. 4A, B) and some had only IPSPs (I pattern, Fig. 4C, D). Of 68 neurons studied, 35 (51%) had only EPSPs and 7 (10%) had only IPSPs in response to CoIC stimulation at the resting membrane potential. The remaining 26 neurons (38%) had both EPSPs and IPSPs. Cells with E, I and I/E synaptic patterns to CoIC stimulation were found in both multipolar and elongated neurons.

3.3.1. I/E responses to CoIC stimulation

In most neurons that exhibited both IPSPs and EPSPs in response to CoIC stimulation at resting membrane potential, IPSPs usually were elicited with a lower threshold and shorter latency than EPSPs. In the 26 I/E type neurons the IPSP had a lower threshold in 14 neurons (54%) and the EPSP a lower threshold in 10 (38%). In the other two neurons (8%), the thresholds were approximately the same.

Examples of neurons with an I/E pattern are shown in Fig. 3A–D. The form of the postsynaptic poten-

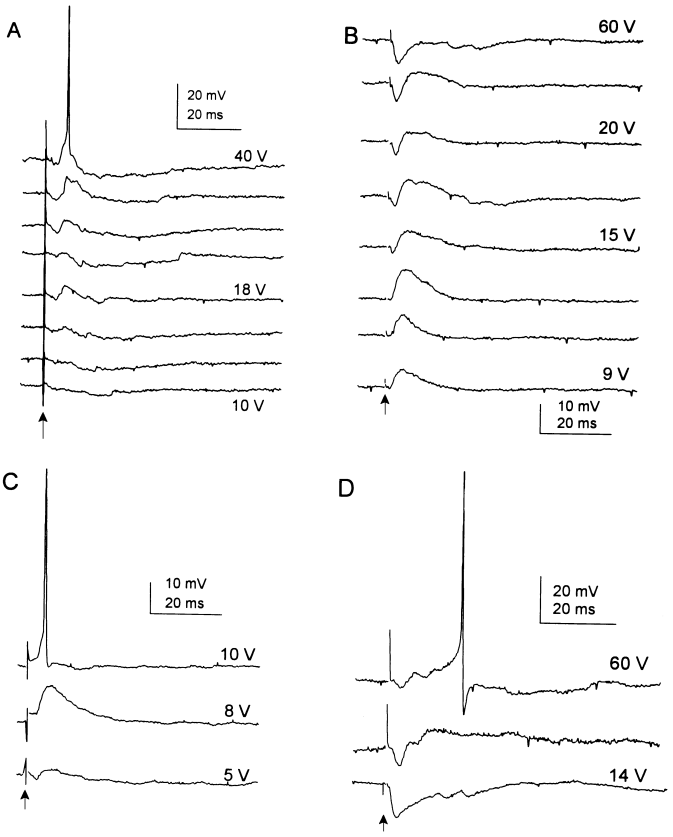

Fig. 3. I/E pattern neurons of IC cortex display varying combinations of IPSP and EPSP: Typical examples. A: This neuron had an I/E pattern, and the EPSP was dominant at higher stimulus intensities. At low stimulation intensities only an IPSP was evoked. With higher voltages a short-latency EPSP became apparent and with still higher intensities the EPSP led to an action potential. B: This neuron had a prominent EPSP at low stimulus intensities, but with higher intensities an IPSP appeared and gradually became dominant, so that the cell would not fire an action potential even with the strongest stimuli. C: In this I/E neuron, both an IPSP and an EPSP were apparent at low intensities, but the EPSP dominated at higher intensities. D: In this neuron an IPSP was prominent at low stimulus intensities, but with larger intensities, a long-latency EPSP appeared and at higher intensities an action potential was observed. Arrows in this and the following figures indicate the time of CoIC stimulation. (RMP: A, −70 mV; B, −62 mV; C, −64mV; D, −56 mV).

tial varied with the stimulation intensity. In the typical example in Fig. 3A, low-intensity stimulation produced an IPSP with a latency of 0.8 ms from the stimulus. At higher stimulus levels, an EPSP with a latency of 2.2 ms became apparent. As intensity levels were increased, the EPSP dominated the IPSP (I/E pattern, E dominant, $n = 16$ of 26), so that with strong stimuli, a single action potential could be evoked in most cells. In some I/E neurons multiple action potentials could be evoked with strong synaptic stimuli ($n = 6$ of 26).

In other I/E neurons, the EPSP occurred at a lower stimulation intensity than the IPSP. The example in Fig. 3B illustrates a neuron with a short-latency EPSP at low stimulus levels. As the stimulus level was increased, a shorter latency IPSP appeared and dominated the response at the highest level tested (I/E pattern, I dominant; $n = 10$). For this cell no action potential could be elicited even with the highest stimulus strengths.

Both IPSPs and EPSPs occurred with only a short delay from the stimulation, but in some neurons ($n = 6$) a short-latency IPSP was followed by a much longer-latency EPSP, as illustrated in Fig. 3D. At low levels of stimulation, a single shock evoked an IPSP with a latency of 1.0 ms. At higher intensities of stimulation, an EPSP emerged with a latency of about 5.0 ms. At relatively high levels of stimulation, the long-latency EPSP dominated and resulted in a single action potential followed by a fast afterhyperpolarization.

3.3.2. E pattern and I responses to CoIC stimulation

The EPSPs induced by CoIC stimulation were often rapid and large enough to cause the neuron to reach spike threshold. The most common type of responses were of the E type. In over 50% of cells an EPSP (E pattern) was apparent without any evidence of an IPSP, if the response was tested with the cell at resting membrane potential. The typical example in Fig. 4A exhibited a graded EPSP in response to increasing stimulus

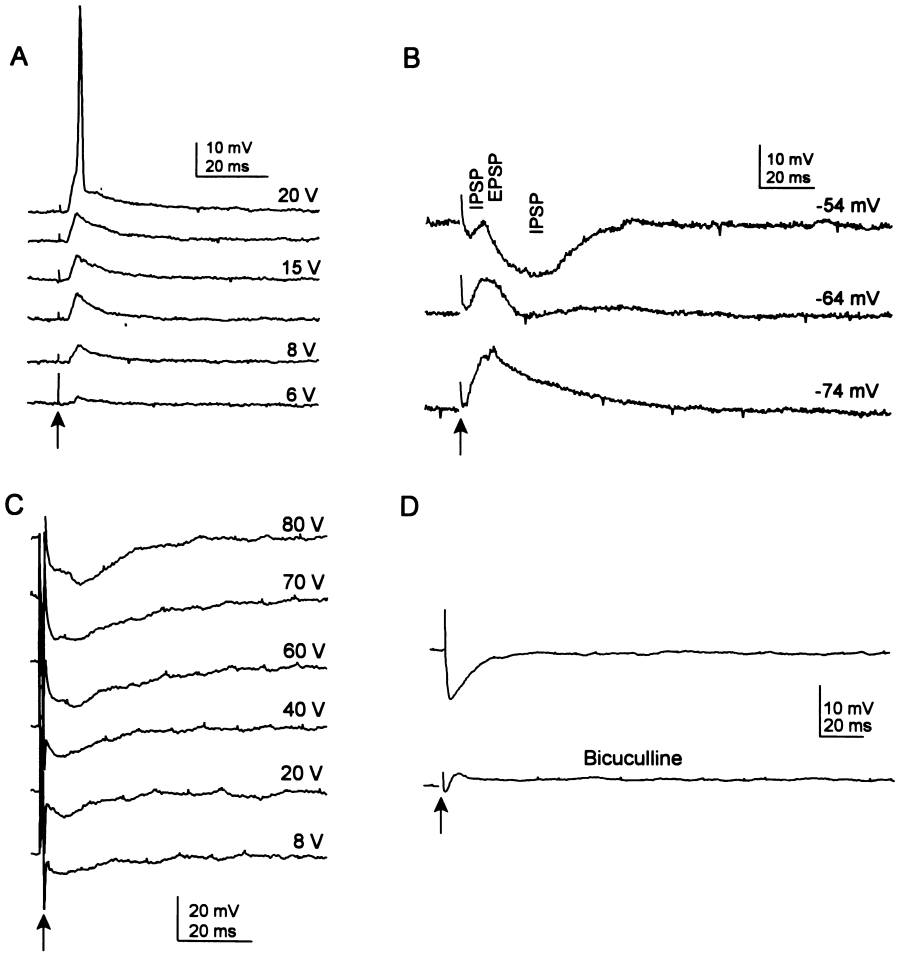

Fig. 4. Examples of E and I pattern neurons. A: This E pattern neuron had only an EPSP if synaptic potentials were elicited with the cell at resting membrane potential. If the stimulation strength was great enough, an action potential fired, but an IPSP was not apparent. B: Another E pattern neuron had only an EPSP at resting membrane potential (−74 mV), but if the cell was depolarized with steady current injection, an IPSP became apparent. At depolarized levels, the postsynaptic potential had the triphasic form (IPSP-EPSP-IPSP) seen in I/E pattern neurons. C: This I pattern neuron had only IPSPs. Even with the most intense stimuli no action potential was evoked. D: This neuron also was classified as an I pattern neuron. If the IPSP was blocked with the $GABA_A$ antagonist bicuculline (30 μM), a very small EPSP became apparent. (RMP: A, −62 mV; B, −74 mV; C, −60 mV; D, −65 mV).

intensity. In this neuron, the largest stimulus caused only a single action potential to fire, but in other neurons ($n = 6$), large stimuli caused multiple action potentials to fire.

Although responses with apparently pure EPSPs accounted for a relatively large percentage of our sample, several lines of evidence demonstrate that one can not conclude from this result that these cells receive no inhibitory input from the CoIC. In some E type neurons ($n = 6$) low-level stimulation produced an EPSP and evoked an action potential. However, further increase in stimulus intensity caused loss of action potential firing, even though only EPSPs were apparent (Fig. 5). This suggested the presence of synaptic inhibition at higher stimulus intensities. This was supported by the effects of bath application of the $GABA_A$ antagonist, bicuculline, which abolished the inhibition (Fig. 5D) and allowed an action potential to be evoked. This finding suggests that GABAergic synaptic transmission is a mechanism underlying this synaptic response pattern.

IPSPs could be evoked by CoIC stimulation in all neurons if the membrane was depolarized with an intracellular current injection. At potentials more positive than resting membrane potential, IPSPs became apparent, and a typical IPSP-EPSP-IPSP response pattern was usually recorded (Fig. 4B, see also Smith, 1992). Although we found no definite examples of neurons that completely lacked synaptic inhibition, we found several neurons that appeared to lack synaptic excitation. In some neurons (e.g. Fig. 4C) only an IPSP was seen, even with high intensities of stimulation (I pattern, $n = 7$). No action potentials could be induced in these neurons by synaptic stimulation.

3.4. Synaptic pharmacology

3.4.1. Excitatory postsynaptic potentials

Synaptic potentials in response to CoIC stimulation often contained a short-latency IPSP and a longer-latency EPSP. The EPSP contained two components, a short-latency component that could be blocked by a non-NMDA quinoxalone excitatory amino acid receptor antagonist, CNQX or DNQX, and a longer-latency component that could be blocked by the NMDA antagonist, AP5. In all neurons tested, the early portion of the EPSP was blocked by the quinoxalone derivative (10 of 10 neurons), and the late portion was blocked by AP5 (10 of 10 neurons). In the example shown in Fig. 6A, a long-lasting EPSP with a latency of 1.1 ms was elicited by CoIC stimulation. AP5 blocked the late portion of the EPSP without influencing the early portion (Fig. 6B), which was then blocked by perfusion of DNQX (Fig. 6C). The IPSP elicited by CoIC stimulation persisted even in the presence of both antagonists.

In another example, shown in Fig. 6E–G, bicuculline and AP5 were perfused to isolate the fast EPSP (Fig. 6F) or CNQX to isolate the later EPSP (Fig. 6G). Note that in these neurons the delayed NMDA receptor-mediated EPSP was not prominent at the resting membrane potential. Part of the reason was that the EPSP was partially masked by a $GABA_A$ receptor-mediated IPSP (see Kanter et al., 1996). In these neurons an IPSP became apparent when the EPSP was blocked by perfusion with DNQX and AP5 (Fig. 6C). Since the IPSPs persisted in the presence of these excitatory amino acid receptor antagonists, this suggests that inhibitory afferents in CoIC end directly on the IC neurons recorded from, rather than acting only through excitation of local inhibitory interneurons.

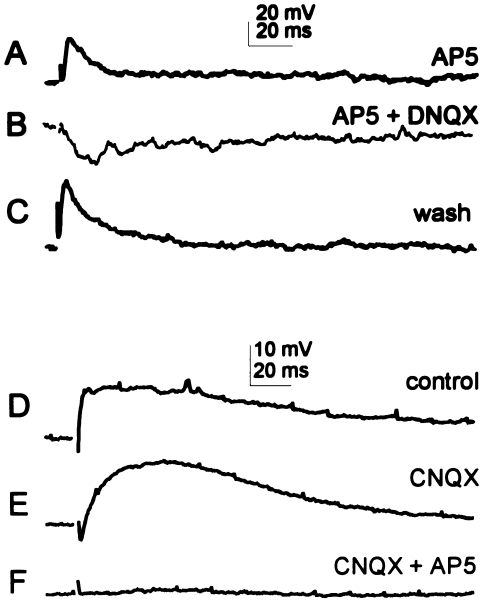

Fig. 6. Postsynaptic potentials in response to stimulation of CoIC are mediated by non-NMDA, NMDA, and/or $GABA_A$ receptors. A: The late phase of the EPSP was attenuated by application of the NMDA receptor antagonist, AP5 (50 μM). B: A combination of non-NMDA receptor blockade with DNQX (20 μM) and NMDA receptor blockade with AP5 eliminated the EPSP and revealed a latent IPSP. C: The effects of the excitatory amino acid antagonists were reversible. D: An EPSP with a fast and a slow component was apparent in a second example neuron. E: The fast component was blocked by CNQX (20 μM) and the slow component remained. F: The remaining slow component was blocked by AP5 (50 μM). (RMP: A–C −66 mV; D–F −68 mV).

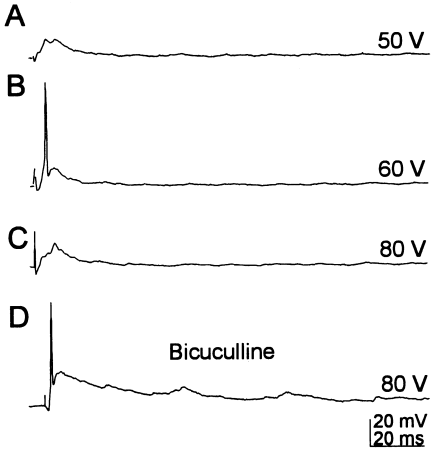

Fig. 5. Evidence for the presence of synaptic inhibition in an E pattern neuron. This neuron was classified as an E pattern neuron because it had only an EPSP even with the highest stimulus intensities. A: At low stimulus intensities, an EPSP was seen. B: With moderate stimulus intensities, the EPSP caused action potential firing. C: With higher intensities, action potential firing was lost. This suggests the presence of synaptic inhibition occurring at higher stimulus intensities, in an amount sufficient to inhibit spike firing, but insufficient to produce an obvious postsynaptic potential. D: This latent inhibition appears to be $GABA_A$ mediated because application of bicuculline (30 μM) was able to restore action potential firing (RMP: −72 mV).

In the presence of bicuculline with either CNQX or AP5, the amplitude of the EPSP was higher and the later EPSP lasted much longer than without bicuculline. These data indicate that short-latency EPSPs in IC cortex are likely mediated by non-NMDA receptors, and long-lasting EPSPs are likely mediated by NMDA receptors.

3.4.2. Inhibitory postsynaptic potentials

When recorded at resting membrane potential, 48% of neurons in the IC cortex exhibited an IPSP in response to CoIC stimulation. The mean latency from the electrical shock stimulation to the onset of the postsynaptic potentials was very short. For EPSPs the latency was 1.6 ± 1.2 (S.E.M.) ms ($n = 38$), and for IPSPs it was 0.55 ± 0.33 ms ($n = 15$), consistent with a monosynaptic connection (Berry and Pentreath, 1976) for both excitatory and inhibitory potentials.

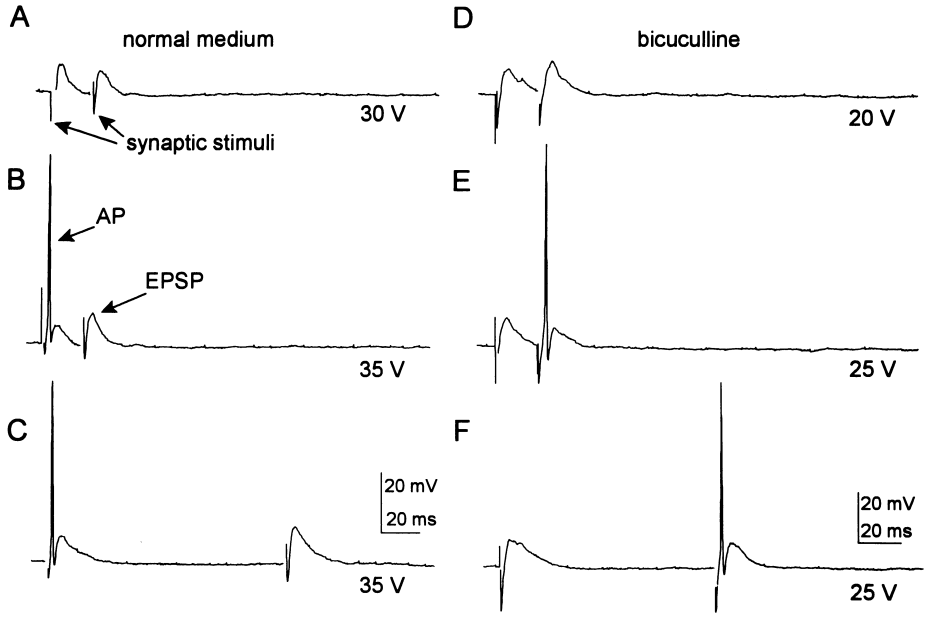

Fig. 7. Paired pulse inhibition in IC cortex neurons is changed into paired pulse excitation by $GABA_A$ receptor blockade. A: In normal perfusion medium, paired pulse stimuli (20 ms interval) to the CoIC resulted in inhibition of the EPSP evoked by the second pulse. B: With further increases in stimulus intensity, action potential firing (AP) was evoked by the first stimulus, but not with the second. C: This inhibition was long-lasting, in this case lasting at least 120 ms. D: The same neuron in A was studied in the presence of bicuculline (5 μM), to partially block $GABA_A$-mediated inhibition. With $GABA_A$ receptor blockade, EPSPs exhibited paired pulse excitation rather than inhibition. E: With stronger stimuli, action potential firing occurred in response to the second stimulus of the pair rather than the first. F: This effect was long-lasting. (RMP: −74 mV).

In order to record monosynaptic IPSPs without overlapping EPSPs, the EPSPs were blocked with perfusion of CNQX and AP5. The mean reversal potential of the monosynaptic IPSPs was -70 ± 12 mV ($n = 6$), slightly negative to the mean resting membrane potential (-64 ± 12 mV, $n = 6$). E type neurons did not have a hyperpolarizing IPSP when tested at resting membrane potential, but an IPSP could be demonstrated if the early EPSP was blocked by application of excitatory amino acid receptor antagonists, or if the cell was depolarized by steady current injection (Fig. 4B, $n = 15$ of 15 neurons).

IPSPs elicited by CoIC stimulation were completely blocked by bicuculline at low concentrations (≤30 μM, 12 of 12 neurons). Fig. 4D shows the blocking effect of bicuculline on the IPSP evoked by CoIC stimulation in an IC cortex neuron. In normal ACSF, a low intensity CoIC stimulation elicited an IPSP with a latency of 0.9 ms. Application of bicuculline completely blocked the IPSP so that no response was apparent at lower levels of stimulation. Higher levels of stimulation elicited an EPSP with a latency of 1.3 ms, but no action potential could be induced in this cell. These results suggest that the IPSPs in the IC cortex neurons are mediated by $GABA_A$ receptors. Postsynaptic non-NMDA, NMDA and $GABA_A$ receptors appear to account for the entire postsynaptic potential in IC cortex neurons evoked by CoIC stimulation, because a combination of CNQX, AP5 and bicuculline eliminated essentially all postsynaptic potentials (Fig. 8B, bottom).

3.5. Paired pulse responses

Pairs of closely-spaced shocks of identical strength were applied to the CoIC to examine whether pairing stimuli modulated synaptic efficacy. Inhibition of the second synaptic response invariably was observed (paired pulse inhibition, e.g. Fig. 7). If the first stimulus was sufficiently intense to elicit action potential firing, firing was commonly inhibited by the second stimulus (11 of 17 recorded neurons). In some cases, the paired pulse inhibition could last for over 100 ms (Fig. 7C, bottom). Perfusion with the $GABA_A$ antagonist, bicuculline (5 μM) in 6 of 6 neurons examined, changed paired pulse inhibition to paired pulse facilitation (Fig. 7D). This indicates that $GABA_A$ receptor-mediated synaptic transmission is necessary for paired pulse inhibition.

The IC cortex shows dense immunolabeling for glutamic acid decarboxylase, and $GABA_A$ and $GABA_B$ receptor binding sites are found, especially in the ICd (Moore and Moore, 1987; Roberts and Ribak, 1987; Glendenning and Baker, 1988; Milbrandt et al., 1996). In certain other brain areas, $GABA_B$ receptors are responsible for auto-inhibition of GABAergic IPSPs during repetitive stimuli (Thompson and Gähwiler, 1989).

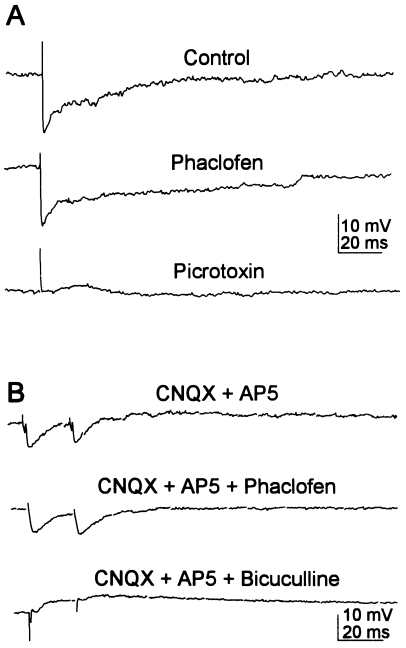

Fig. 8. $GABA_B$ receptors modulate synaptic inhibition. A: (top) This neuron exhibited only an IPSP, without a discernible EPSP at this stimulation intensity. (middle) Application of the $GABA_B$ antagonist, phaclofen (20 μM), caused prolongation of the IPSP. (bottom) Subsequent application of the $GABA_A$ channel antagonist, picrotoxin (40 μM), blocked the IPSP. B: (top) In the presence of CNQX and AP5 this neuron exhibited IPSPs in response to paired pulse stimulation and the second IPSP was diminished in amplitude (paired pulse inhibition of the IPSP). (middle) Addition of phaclofen (20 μM) blocked the paired pulse inhibition of the second IPSP. (bottom) Subsequent application of bicuculline (30 μM) blocked both IPSPs. (RMP: A, −63 mV; B, −68 mV). (Note: The initial deflections in each trace are stimulus artifacts.)

In the present study application of the $GABA_B$ antagonist, phaclofen (20 μM), could prolong the duration of IPSPs in all IC cortex neurons tested (Fig. 8A, middle trace). This is consistent with blockade of inhibitory $GABA_B$ autoreceptors by phaclofen. To investigate whether $GABA_B$ receptors play a role in repetitive synaptic stimulation in IC cortex neurons, we examined the effect of paired stimuli on synaptic responses in the presence of CNQX and AP5. There is a persistent depolarization after the perfusion of these excitatory amino acid receptor antagonists, which may suggest excitatory neurotransmitter may be released with commissural stimulation, such as acetylcholine (Farley et al., 1983). Paired pulse inhibition of the IPSP was normally seen. This reduction of the IPSP was significantly diminished after perfusion of phaclofen (20 μM) (Fig. 8B; 5 of 5 neurons tested). This result suggested that $GABA_B$ receptors can modulate synaptic responses presynaptically via a negative feedback mechanism.

Although IPSPs were sufficiently robust in most cells to produce paired pulse inhibition of the synaptic response, the response of E pattern neurons was sometimes different. The neuron shown in Fig. 9A–D was an E type neuron but an IPSP could be demonstrated by depolarization (compare Fig. 9A and C). At resting membrane potential only a two-phase EPSP was apparent. At resting potential, an action potential could be evoked on the first phase of the EPSP, but at a depolarized potential, the IPSP was enhanced, preventing the initial AP, but allowing an action potential to be evoked on the second phase of the EPSP. During repetitive stimulation, temporal summation of the EPSP occurred, leading to action potential firing late in the train (Fig. 9E) if lower intensity stimuli were given, and with each stimulus if larger stimuli were given (Fig. 9F,G).

3.6. Response to repetitive synaptic stimulation

We examined the responses of IC cortex neurons to repetitive stimulation of the CoIC (Fig. 10, $n = 10$). Each of the neurons tested was capable of firing an action potential after a single pulse of electrical stimulation. However, when tested with repetitive stimuli they were not able to consistently exhibit action potentials at rates greater than 10–50 Hz. At higher rates of stimulation the probability of responding dropped precipitously. During trains of stimuli the amplitude of IPSPs was markedly reduced (Fig. 10A and B) but not abolished. Although GABAergic inhibition appears to be markedly reduced during such a train of stimuli, that which remains can contribute to limitation of firing rate. In three neurons the effects of repetition rate were determined after bath application of bicuculline. In the example shown in Fig. 10C, 10 μM bicuculline was perfused, and the neuron was able to follow the 50 Hz stimulus. These data imply that synaptic responses to repetitive stimulation of the CoIC are, in part, regulated by $GABA_A$ receptor mediated transmission.

4. Discussion

The results of the present study illustrate the complexity of the excitatory and inhibitory influences from commissural sources onto single neurons, which appears to be a common feature of neurons in the IC cortex. Thus, the categories of postsynaptic response in IC cortex neurons to CoIC stimulation included exclusive E patterns or I patterns and I/E patterns were also commonly observed. Either the EPSP or IPSP may be dominant in E/I cells with intense stimuli. Differential changes in EPSP and/or IPSP amplitude were produced by gradually increasing the intensity of stimulation. Similarly, complex synaptic responses were also observed with lemniscal and commissural stimulation in a previous in vitro study (Smith, 1992). Considering

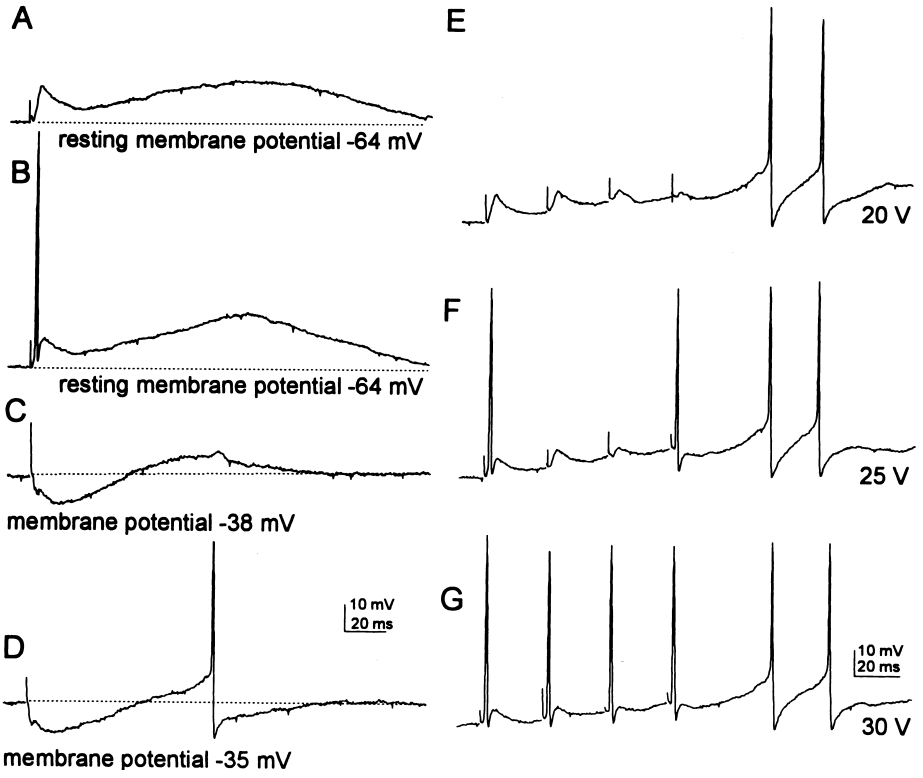

Fig. 9. Prolonged EPSPs may contribute to paired pulse excitation. A: Occasionally, IC cortex neurons had both a short-latency as well as a prolonged secondary EPSP, as seen in this example. B: At resting membrane potential, action potentials were elicited from the first EPSP by lower stimulus intensities than from the second. C: When the membrane was depolarized with steady current injection, a prominent IPSP was seen that overcame the depolarization caused by the short-latency EPSP. D: With further membrane depolarization action potential firing was elicited by lower intensity stimuli from the long-latency EPSP, because the IPSP overlapped the first EPSP. E–G: The long latency EPSP contributes to action potential firing in a short train of stimuli. A–G are from the same neuron and the initial upward going deflections represent stimulus artifacts. (RMP: −64 mV).

that the CoIC is only one of the major inputs into IC neurons, a more complex pattern of excitatory and inhibitory response patterns in response to acoustic stimuli undoubtedly exists in these neurons in vivo.

Inhibition appears to play a critical role in shaping synaptic responses of IC cortex neurons to CoIC stimulation. Although some IC cortex cells without EPSPs in response to CoIC stimulation were noted in the present study, all neurons examined exhibited IPSPs. Thus, even IC cortex cells that were characterized as E type would exhibit an IPSP following depolarization. IPSPs evoked by CoIC stimulation in the present study appear to be mediated by $GABA_A$ receptors, as demonstrated by the blockade of these potentials by bath application of a selective $GABA_A$ antagonist. GABA is the major inhibitory transmitter that shapes the responses of IC neurons, as noted above. Inhibitory responses to acoustic stimuli in vivo have been shown to involve GABA prominently, and in the present study all synaptic potentials evoked by CoIC stimulation were blocked by combined perfusion with $GABA_A$ and glutamate receptor antagonists. However, IC neurons receive synaptic input from other sources, including glycinergic inputs (Suneja et al., 1998), as noted above, and potential contributions of other transmitters are not precluded (Farley et al., 1983; Faingold et al., 1991b; Ikeuchi and Nishizaki, 1995; Le Beau et al., 1996). Considerably more data had been obtained studying the action of GABA in the ICc, but the data that are available on IC cortex are quite comparable (Smith, 1992; Li et al., 1994).

In the present and a previous study, IC cortex neurons exhibited paired pulse inhibition of EPSPs, but perfusion with a $GABA_A$ receptor antagonist in the present study resulted in conversion of the inhibition to paired pulse excitation. Hence, $GABA_A$ receptors appear to mediate the inhibitory effect. The paired pulse excitation that is unmasked may be due to residual calcium in presynaptic excitatory terminals, as seen in other areas of the nervous system (Tanabe and Kaneko, 1996). $GABA_B$ binding sites have also been found in the IC cortex (Glendenning and Baker, 1988; Milbrandt et al., 1996) but their role is not clear. Paired pulse inhibition of IPSPs in IC cortex neurons was observed previously (Li et al., 1994) and in the present study this inhibition was blocked by perfusion of a

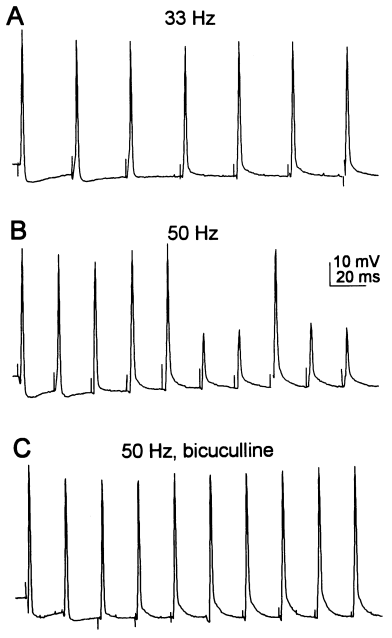

Fig. 10. $GABA_A$ mediated synaptic inhibition modulates action potential firing during trains of stimuli. A: In this neuron, stimulus strength was adjusted to produce action potential firing with each stimulus in a 33 Hz train. A robust IPSP was evoked by the first stimulus, but the amplitude of these IPSPs were markedly attenuated later in the train. B: Despite fading of synaptic inhibition during the train, action potentials were not evoked consistently at 50 Hz, although EPSPs could be seen. C: Application of the $GABA_A$ antagonist, bicuculline (10 μM), reduced synaptic inhibition and allowed the action potentials to be evoked consistently at 50 Hz. (RMP: −76 mV).

$GABA_B$ antagonist, phaclofen. This finding suggests the possibility of auto-inhibition of GABAergic terminals. Thus, paired pulse inhibition of EPSPs appears to be due to $GABA_A$ inhibition and paired pulse inhibition of IPSPs may be due to $GABA_B$ inhibition of GABA release. The present results provide evidence that one possible active site of $GABA_B$ receptors in IC may be presynaptic to modulate GABA release via negative feedback, as seen in other CNS neurons (Lambert and Wilson, 1994; Tanabe and Kaneko, 1996).

The present results further confirm the presence of significant $GABA_A$ synaptic responses in IC cortex neurons and that contralateral inhibition from the CoIC is, at least partially, GABAergic. The finding of short-latency inhibition in the IC neurons is consistent with extracellular recording studies in vivo that show that some IC neurons discharge to acoustic stimulation at shorter latencies in the presence of inhibitory amino acid receptor antagonists (Faingold et al., 1991a,b; Klug et al., 1995).

In the present study GABA-mediated inhibition was prominent in I neurons and E/I neurons in response to CoIC stimulation and inhibition could also be readily unmasked in E neurons by application of NMDA and/or non-NMDA receptor antagonists. Blockade of GABAergic IPSPs induced in IC cortex neurons by stimulation at CoIC also unmasked a long-lasting EPSP in response to synaptic stimulation (Li et al., 1994; Smith, 1992).

The present study provides evidence that CoIC contains direct monosynaptic inhibitory projections to cells in the cortex of IC. This is supported by the finding that IPSPs are still evoked in the presence of a complete excitatory amino acid receptor blockade, which suggests that feed-forward and feed-back inhibition are not required for the inhibition. The presence of neurons with pure IPSPs is also supportive of this conclusion, but these data do not preclude the existence of feed-forward inhibition. The presence of this monosynaptic inhibitory connection may account for the unusually short latency of the IPSP, often resulting in the uncommon IPSP-EPSP-IPSP configuration. Evidence for short-latency IPSPs evoked by acoustic stimulation has been observed in vivo (Kuwada et al., 1997; Covey et al., 1996).

Stimulation of the CoIC produces both EPSPs and IPSPs, frequently in the same IC cortex neuron. A number of possible sources of these potentials are known, which include input from the contralateral IC (Faye-Lund, 1985; Coleman and Clerici, 1987; Herbert et al., 1991; Feliciano and Potashner, 1995), and inputs via the lateral lemniscus are also well known (Oliver and Shneiderman, 1991). The contralateral IC is one of the major sources of axons that cross the midline to innervate ICd and ICx (Faye-Lund and Osen, 1985; Coleman and Clerici, 1987; Druga and Syka, 1984; Saldaña and Merchán, 1992). Neurons in the ICc and cortex are immunoreactive for GABA and the GABA synthesizing enzyme, glutamic acid decarboxylase, and many terminals in the IC cortex have flattened or pleomorphic vesicles, characteristic of inhibitory synapses seen elsewhere in the CNS (Moore and Moore, 1987; Roberts and Ribak, 1987; Caspary et al., 1990; Oliver et al., 1994; Goldsmith et al., 1995; Chakravarty and Faingold, 1996).

Direct GABAergic modulation of IC neuron synaptic responses observed in the present study confirms previous in vivo extracellular studies of GABAergic modulation of inhibitory responses to acoustic stimulation (as discussed above). Thus, non-monotonic response and offset inhibition observed in IC neuronal responses to acoustic stimulation could reflect the I/E pattern observed in vitro. These acoustically evoked forms of inhibition were reduced by application of a $GABA_A$ antagonist (Faingold et al., 1991b). IC cortex neurons tend to habituate to repeated stimulation as compared to the ICc (Chakravarty and Faingold, 1996). E/I neurons with prolonged periods of post-excitatory inhibi-

tion, which appear to be GABA-mediated, were commonly observed in the present study and may provide a partial explanation for this habituation, since blockade of $GABA_A$ receptors by bicuculline can increase the response rate, as shown in the present study (e.g. Fig. 10).

The IC cortex, in contrast to the ICc, has significant interactions with regions not directly associated with auditory function (Tokunaga et al., 1984; Coleman and Clerici, 1987; Olazabal and Moore, 1989; Oliver and Shneiderman, 1991). Thus, the IC cortex provides a critical link in the acousticomotor reflex pathway. A number of studies have implicated the IC cortex as a critical site in the neuronal network for acoustically-evoked epileptic seizure generalization and a site of convergence of auditory outflow from the ICc to the motor pathway (McCown et al., 1984, 1987; Browning, 1994; Ribak et al., 1994; Albeck and Konishi, 1995; Faingold and Randall, 1999; Chakravarty and Faingold, 1996; Faingold, 1999). The present study illuminates some of the details of the complex role that GABAergic transmission serves in shaping IC synaptic response patterns. In animals susceptible to acoustically evoked seizures, reduced efficacy of GABA function in the IC leads to diminished amounts of several forms of GABA-mediated inhibition (Faingold et al., 1986a,b; Faingold and Boersma Anderson, 1991). The extensive ability of the $GABA_A$ antagonist, bicuculline, to block IPSPs in IC neurons in the present study is consistent with the ability of bicuculline to produce acoustically-evoked seizure susceptibility in normal animals when injected focally into the IC (Millan et al., 1986). Further support for this hypothesis is provided by recent preliminary studies, indicating that GABA-mediated IPSPs of IC cortex neurons from AGS-susceptible rats are blocked at lower than normal concentrations of bicuculline (Faingold et al., 1998). Alterations of GABAergic inhibition in IC neurons appear to play a critical role in deafferentation-related forms of IC pathophysiology, including presbycusis, tinnitus and audiogenic seizures (Roberts et al., 1985; Faingold et al., 1986b; Caspary et al., 1990; Caspary et al., 1995; Bledsoe et al., 1995; Gerken, 1996; N'Gouemo et al., 1996; Szczepaniak and Moller, 1996; Chakravarty and Faingold, 1997; Milbrandt et al., 1997; Potashner et al., 1997; Tang et al., 1997; Faingold et al., 1998), and normal GABAergic function in the IC in vivo is important to coding of acoustic stimuli.

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References

Albeck, Y., Konishi, M., 1995. Responses of neurons in the auditory pathway of the barn owl to partially correlated binaural signals. J. Neurophysiol. 74, 1689–1700.

Banay-Schwartz, M., Lajtha, A., Palkovits, M., 1989. Changes with aging in the levels of amino acids in rat CNS structural elements I. Glutamate and related amino acids. Neurochem. Res. 14, 555–562.

Berry, M.S., Pentreath, V.W., 1976. Criteria for distinguishing between monosynaptic and polysynaptic transmission. Brain Res. 105, 1–20.

Bledsoe, S.C., Jr., Nagase, S., Miller, J.M., Altschuler, R.A., 1995. Deafness-induced plasticity in the mature central auditory system. NeuroReport 7, 225–229.

Browning, R.A., 1994. Anatomy of generalized convulsive seizures. In: Malafosse, A., Genton, P., Hirsch, E., Marescaux, C., Broglin, D., Bernasconi, R. (Eds.), Idiopathic Generalized Epilepsies: Clinical, Experimental and Genetic Aspects. John Libbey and Company, New York, pp. 399–413.

Caicedo, A., Kungel, M., Pujol, R., Friauf, E., 1998. Glutamate-induced Co^{2+} uptake in rat auditory brainstem neurons reveals developmental changes in Ca^{2+} permeability of glutamate receptors. Eur. J. Neurosci. 10, 941–954.

Caird, D., 1991. Processing in the colliculi. In: Altschuler, R.A. et al. (Eds.), Neurobiology of Hearing: The Central Auditory System. Raven Press, New York, pp. 253–292.

Caspary, D.M., Raza, A., Armour, B., Pippin, J., Arneric, S.P., 1990. Immunocytochemical and neurochemical evidence for age-related loss of GABA in the inferior colliculus: Implications for neural presbycusis. J. Neurosci. 10, 2363–2372.

Caspary, D.M., Milbrandt, J.C., Helfert, R.H., 1995. Central auditory aging: GABA changes in the inferior colliculus. Exp. Gerontol. 30, 349–360.

Chakravarty, D.N., Faingold, C.L., 1996. Increased responsiveness and failure of habituation in neurons of the external nucleus of inferior colliculus associated with audiogenic seizures of the genetically epilepsy-prone rat. Exp. Neurol. 141, 280–286.

Chakravarty, D.N., Faingold, C.L., 1997. Aberrant neuronal responsiveness in the genetically epilepsy-prone rat: Acoustic responses and influences of the central nucleus upon the external nucleus of inferior colliculus. Brain Res. 761, 263–270.

Coleman, J.R., Clerici, W.J., 1987. Sources of projections to subdivisions of the inferior colliculus in the rat. J. Comp. Neurol. 262, 215–226.

Covey, E., Kauer, J.A., Casseday, J.H., 1996. Whole-cell patch-clamp recording reveals subthreshold sound-evoked postsynaptic currents in the inferior colliculus of awake bats. J. Neurosci. 16, 3009–3018.

Druga, R., Syka, J., 1984. Ascending and descending projections to the inferior colliculus in the rat. Physiol. Bohemoslov. 33, 31–42.

Faingold, C.L., 1999. Neuronal networks in the genetically epilepsy-prone rat. In: Delgado-Escueta, A. Olsen, R., Wilson, M. (Eds.), Jasper's Basic Mechanisms of the Epilepsies, Lippincott-Raven, NY, pp. 311–321.

Faingold, C.L., Boersma Anderson, C.A., 1991. Loss of intensity-induced inhibition in inferior colliculus neurons leads to audiogenic seizure susceptibility in behaving genetically epilepsy-prone rats. Exp. Neurol. 113, 354–363.

Faingold, C.L., Randall, M.E., 1999. Neurons in the deep layers of superior colliculus play a critical role in the neuronal network for audiogenic seizures: mechanisms of production of wild running behavior. Brain Res. 815 (2), 250–258.

Faingold, C.L., Travis, M.A., Gehlbach, G., Hoffmann, W.E., Jobe, P.C., Laird II, H.E., Caspary, D.M., 1986a. Neuronal response abnormalities in the inferior colliculus of the genetically epilepsy-prone rat. Electroencephalogr. Clin. Neurophysiol. 63, 296–305.

Faingold, C.L., Gehlbach, G., Caspary, D.M., 1986b. Decreased effectiveness of GABA-mediated inhibition in the inferior colliculus of the genetically epilepsy-prone rat. Exp. Neurol. 93, 145–159.

Faingold, C.L., Gehlbach, G., Caspary, D.M., 1989a. On the role of GABA as an inhibitory neurotransmitter in inferior colliculus neurons: Iontophoretic studies. Brain Res. 500, 302–312.

Faingold, C.L., Hoffmann, W.E., Caspary, D.M., 1989b. Effects of excitant amino acids on acoustic responses of inferior colliculus neurons. Hear. Res. 40, 127–136.

Faingold, C.L., Boersma Anderson, C.A., Caspary, D.M., 1991a. Involvement of GABA in acoustically-evoked inhibition in inferior colliculus neurons. Hear. Res. 52, 201–216.

Faingold, C.L., Gehlbach, G., Caspary, D.M., 1991b. Functional pharmacology of inferior colliculus neurons. In: Altschuler, R.A., Bobbin, R.P., Clopton, B.M., Hoffman, D.W. (Eds.), Neurobiology of Hearing-The Central Auditory System. Raven Press, New York, pp. 223–251.

Faingold, C.L., Li, Y., Evans, M.S., 1998. Diminished GABA-A receptor-mediated inhibition in the inferior colliculus may subserve audiogenic seizure susceptibility. Epilepsia 39 (56), 37.

Farley, G.R., Morley, B.J., Javel, E., Gorga, M.P., 1983. Single-unit responses to cholinergic agents in the rat inferior colliculus. Hear. Res. 11, 73–91.

Faye-Lund, H., 1985. The neocortical projection to the inferior colliculus in the albino rat. Anat. Embryol. 173, 53–70.

Faye-Lund, H., Osen, K.K., 1985. Anatomy of the inferior colliculus in rat. Anat. Embryol. 171, 1–20.

Feldman, D.E., Knudsen, E.I., 1994. NMDA and non-NMDA glutamate receptors in auditory transmission in the barn owl inferior colliculus. J. Neurosci. 14, 5939–5958.

Feliciano, M., Potashner, S.J., 1995. Evidence for a glutamatergic pathway from the guinea pig auditory cortex to the inferior colliculus. J. Neurochem. 65, 1348–1357.

Fuzessery, Z.M., Hall, J.C., 1996. Role of GABA in shaping frequency tuning and creating FM sweep selectivity in the inferior colliculus. J. Neurophysiol. 76, 1059–1073.

Gaza, W.C., Ribak, C.E., 1997. Immunocytochemical localization of AMPA receptors in the rat inferior colliculus. Brain Res. 774, 175–183.

Gerken, G.M., 1996. Central tinnitus and lateral inhibition: an auditory brainstem model. Hear. Res. 97, 75–83.

Glendenning, K.K., Baker, B.N., 1988. Neuroanatomical distribution of receptors for three potential inhibitory neurotransmitters in the brainstem auditory nuclei of the cat. J. Comp. Neurol. 275, 288–308.

Goldsmith, J.D., Kujawa, S.G., McLaren, J.D., Bledsoe, S.C., Jr., 1995. In vivo release of neuroactive amino acids from the inferior colliculus of the guinea pig using brain microdialysis. Hear. Res. 83, 80–88.

Herbert, H., Aschoff, A., Ostwald, J., 1991. Topography of projections from the auditory cortex to the inferior colliculus in the rat. J. Comp. Neurol. 304, 103–122.

Hosomi, H., Hirai, H., Okada, Y., Amatsu, M., 1995. Long-term potentiation of neurotransmission in the inferior colliculus of the rat. Neurosci. Lett. 195, 175–178.

Hosomi, H., Mori, M., Amatsu, M., Okada, Y., 1997. GABA-Activated conductance in cultured rat inferior colliculus neurons. J. Neurophysiol. 77, 994–1002.

Ikeuchi, Y., Nishizaki, T., 1995. The P2Y purinoceptor-operated potassium channel is possibly regulated by the beta gamma subunits of a pertussis toxin-insensitive G-protein in cultured rat inferior colliculus neurons. Biochem. Biophys. Res. Commun. 214, 589–596.

Kanter, E.D., Kapur, A., Haberly, L.B., 1996. A dendritic $GABA_A$-mediated IPSP regulates facilitation of NMDA-mediated responses to burst stimulation of afferent fibers in piriform cortex. J. Neurosci. 16, 307–312.

Klug, A., Park, T.J., Pollak, G.D., 1995. Glycine and GABA influence binaural processing in the inferior colliculus of the mustache bat. J. Neurophysiol. 74, 1701–1713.

Kuwada, S., Batra, R., Yin, T.C., Oliver, D.L., Haberly, L.B., Stanford, T.R., 1997. Intracellular recordings in response to monaural and binaural stimulation of neurons in the inferior colliculus of the cat. J. Neurosci. 17, 7565–7581.

Lambert, N.A., Wilson, W.A., 1994. Temporally distinct mechanisms of use-dependent depression at inhibitory synapses in the rat hippocampus in vitro. J. Neurophysiol. 72, 121–130.

Le Beau, F.E., Rees, A., Malmierca, M.S., 1996. Contribution of GABA- and glycine-mediated inhibition to the monaural temporal response properties of neurons in the inferior colliculus. J. Neurophysiol. 75, 902–919.

Li, Y., Evans, M.S., Faingold, C.L., 1994. Inferior colliculus neuronal membrane and synaptic properties in genetically epilepsy-prone rats. Brain Res. 660, 232–240.

Li, Y., Evans, M.S., Faingold, C.L., 1998. In vitro electrophysiology of neurons in subnuclei of rat inferior colliculus in vitro. Hear. Res. 121, 1–10.

McCown, T.J., Greenwood, R.S., Frye, G.D., Breese, G.R., 1984. Electrically elicited seizures from the inferior colliculus: A potential site for the genesis of epilepsy? Exp. Neurol. 86, 527–542.

McCown, T.J., Greenwood, R.S., Breese, G.R., 1987. Inferior collicular interactions with limbic seizure activity. Epilepsia 28, 234–241.

Milbrandt, J.C., Albin, R.L., Turgeon, S.M., Caspary, D.M., 1996. $GABA_A$ receptor binding in the aging rat inferior colliculus. Neuroscience 73, 449–458.

Milbrandt, J.C., Hunter, C., Caspary, D.M., 1997. Alterations of $GABA_A$ receptor subunit mRNA levels in the aging Fischer 344 rat inferior colliculus. J. Comp. Neurol. 379, 455–465.

Millan, M.H., Meldrum, B.S., Faingold, C.L., 1986. Induction of audiogenic seizure susceptibility by focal infusion of excitant amino acid or bicuculline into the inferior colliculus of normal rats. Exp. Neurol. 91, 634–639.

Moore, D.R., Kotak, V.C., Sanes, D.H., 1998. Commissural and lemniscal synaptic input to the gerbil inferior colliculus. J. Neurophysiol. 80, 2229–2236.

Moore, J.K., Moore, R.Y., 1987. Glutamic acid decarboxylase-like immunoreactivity in brainstem auditory nuclei of the rat. J. Comp. Neurol. 260, 157–174.

Mori, K., 1997. Across-frequency nonlinear inhibition by GABA in processing of interaural time difference. Hear. Res. 111, 22–30.

Najlerahim, A., Harrison, P.J., Barton, A.J.L., Hefferman, J., Pearson, R.C.A., 1990. Distribution of messenger RNAs encoding the enzymes glutaminase, aspartate aminotransferase and glutamic acid decarboxylase in rat brain. Mol. Brain Res. 7, 317–333.

Nelson, P.G., Erulkar, S.D., 1963. Synaptic mechanisms of excitation and inhibition in the central auditory pathway. J. Neurophysiol. 26, 908–923.

N'Gouemo, P., Caspary, D.M., Faingold, C.L., 1996. Decreased GABA effectiveness in the inferior colliculus neurons during ethanol withdrawal in rats susceptible to audiogenic seizures. Brain Res. 724, 200–204.

Olazabal, U.E., Moore, J.K., 1989. Nigrotectal projection to the inferior colliculus: horseradish peroxidase transport and tyrosine hydroxylase immunohistochemical studies in rats, cats and bats. J. Comp. Neurol. 282, 98–118.

Oliver, D.L., Shneiderman, A., 1991. The Anatomy of the inferior colliculus: a cellular basis for integration of monaural and binaural information. In: Altschuler, R.A., et al. (Eds.), Neurobiology

of Hearing: The Central Auditory System. Raven Press, New York, pp. 195–222.
Oliver, D.L., Huerta, M.F., 1992. Inferior and Superior Colliculi. In: Webster, D.B., Popper, A.N., Fay, R.R. (Eds.), The Mammalian Auditory Pathway: Neuroanatomy. Springer-Verlag, New York, pp. 168–221.
Oliver, D.L., Winer, J.A., Beckius, G.E., Saint Marie, R.L., 1994. Morphology of GABAergic neurons in the inferior colliculus of the cat. J. Comp. Neurol. 340, 27–42.
Palombi, P.S., Caspary, D.M., 1996a. Physiology of the young adult Fischer 344 rat inferior colliculus: Responses to contralateral monaural stimuli. Hear. Res. 100, 41–58.
Palombi, P.S., Caspary, D.M., 1996b. GABA inputs control discharge rate primarily within frequency receptive fields of inferior colliculus neurons. J. Neurophysiol. 75, 2211–2219.
Paxinos, G., Watson, C., 1986. The Rat Brain in Stereotaxic Coordinates. Academic Press, Sydney.
Pedemonte, M., Torterolo, P., Velluti, R.A., 1997. In vivo intracellular characteristics of inferior colliculus neurons in guinea pigs. Brain Res. 759, 24–31.
Pierson, M.G., Smith, K.L., Swann, J.W., 1989. A slow NMDA-mediated synaptic potential underlies seizures originating from the midbrain. Brain Res. 486, 381–386.
Potashner, S.J., Suneja, S.K., Benson, C.G., 1997. Regulation of D-aspartate release and uptake in adult brain stem auditory nuclei after unilateral middle ear ossicle removal and cochlear ablation. Exp. Neurol. 148, 222–235.
Ribak, C.E., Khurana, V., Lien, N.T., 1994. The effect of midbrain follicular knife cuts on audiogenic seizure severity in the genetically epilepsy-prone rat. J. Hirnforsch. 35, 303–311.
Roberts, R.C., Ribak, C.E., 1987. GABAergic neurons and axon terminals in the brainstem auditory nuclei of the gerbil. J. Comp. Neurol. 258, 267–280.
Roberts, R.C., Ribak, C.E., Oertel, W.H., 1985. Increased numbers of GABAergic neurons occur in the inferior colliculus of an audiogenic model of genetic epilepsy. Brain Res. 361, 324–338.
Saint Marie, R.L., 1996. Glutamatergic connections of the auditory midbrain: Selective uptake and axonal transport of D-[^{3}H]aspartate. J. Comp. Neurol. 373, 255–270.
Saldaña, E., Merchán, M.A., 1992. Intrinsic and commissural connections of the rat inferior colliculus. J. Comp. Neurol. 319, 417–437.
Smith, P.H., 1992. Anatomy and physiology of multipolar cells in the rat inferior collicular cortex using the in vitro brain slice technique. J. Neurosci. 12, 3700–3715.
Suneja, S.K., Potashner, S.J., Benson, C.G., 1998. Plastic changes in glycine and GABA release and uptake in adult brain stem auditory nuclei after unilateral middle ear ossicle removal and cochlear ablation. Exp. Neurol. 151, 273–288.
Szczepaniak, W.S., Moller, A.R., 1996. Effects of (-)-baclofen, clonazepam, and diazepam on tone exposure-induced hyperexcitability of the inferior colliculus in the rat: Possible therapeutic implications for pharmacological management of tinnitus and hyperacusis. Hear. Res. 97, 46–53.
Tanabe, M., Kaneko, T., 1996. Paired pulse facilitation of GABAergic IPSCs in ventral horn neurons in neonatal spinal cord. Brain Res. 716, 101–106.
Tang, E., Yip, P.K., Chapman, A.G., Jane, D.E., Meldrum, B.S., 1997. Prolonged anticonvulsant action of glutamate metabotropic receptor agonists in inferior colliculus of genetically epilepsy-prone rats. Eur. J. Pharmacol. 327, 109–115.
Thompson, S.M., Gähwiler, B.H., 1989. Activity-dependent disinhibition III. Desensitization and GABAB receptor-mediated presynaptic inhibition in the hippocampus in vitro. J. Neurophysiol. 61, 524–533.
Tokunaga, A., Sugita, S., Otani, K., 1984. Auditory and non-auditory subcortical afferents to the inferior colliculus of the rat. J. Hirnforsch. 25, 461–472.
Wagner, T., 1994. Intrinsic properties of identified neurones in the central nucleus of mouse inferior colliculus. NeuroReport 6, 89–93.
Wagner, T., 1996. Lemniscal input to identified neurons of the central nucleus of mouse inferior colliculus: an intracellular brain slice study. Eur. J. Neurosci. 8, 1231–1239.
Yamauchi, R., Amatsu, M., Okada, Y., 1989. Effect of GABA (gamma-aminobutyric acid) on neurotransmission in inferior colliculus slices from guinea pigs. Neurosci. Res. 6, 446–455.