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Partition of Transient and Sustained Inhibitory Glycinergic Input to Retinal Ganglion Cells

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Physiological and pharmacological properties of possible subtypes of the native glycine receptor were investigated in retinal neurons using whole-cell voltage-clamp techniques. Two discrete inhibitory glycine responses were identified in ganglion cells. The responses could be distinguished pharmacologically: one was sensitive to strychnine and the other to 5,7-dichlorokynurenic acid. The two responses had different kinetics: the former had a fast onset and fast desensitization, whereas the latter had a slower onset and was much more sustained. The physiological and pharmacological distinctions suggest that the responses are mediated by different receptors. These receptors transduce glycinergic synaptic signals to ganglion cells, where they serve as low- and high-pass filters, respectively, of EPSPs.

Key words: glycine receptors; inhibition; strychnine; 5,7-dichlorokynurenic acid; ganglion cells; retina

In the vertebrate retina, glycine and GABA share the task of mediating inhibition to ganglion cells. They contribute to the formation of trigger features, such as directional selectivity and edge detection (Caldwell et al., 1978). Several GABA receptor subtypes have been identified and linked to specific aspects of visual information processing (Werblin et al., 1988; Pan and Slaughter, 1991; Zhang and Slaughter, 1995). Although GABAergic and glycinergic neurons are equally populous in the inner retina, a similar diversity of glycinergic receptors has not been described.

There is reason to suspect that discrete glycine receptor subtypes exist in retina. Like the GABA receptor, the glycine receptor is a pentamer of α and β subunits in which there are multiple isoforms. In mammalian retina, three α subunit isoforms (α_1, α_2, α_3) have been localized to rat ganglion cells (Greferath et al., 1994). This molecular diversity implies functional and pharmacological variability (Becker, et al., 1988; Betz, 1991; Malosio, 1991a); however, it has proven difficult to translate molecular studies to properties of native glycine receptors or to determine the physiological significance of differential expression.

We examined native glycine receptors in isolated amphibian retinal neurons and found that glycine produced two currents: a large, fast, transient, desensitizing component and a smaller, slower, sustained component. Selective antagonists of each of these two currents were identified, implicating two subtypes of the glycine receptor. The agonist and antagonist sensitivities of these two putative receptors were characterized, and their role in synaptic transmission was identified. The results indicate that tonic and phasic glycinergic IPSPs result from two populations of receptor.

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MATERIALS AND METHODS

Animal experimental preparation. The isolated retinal cell preparation has been described (Bader et al., 1979; Pan and Slaughter, 1995). Briefly, the tiger salamander *Ambystoma tigrinum* (Kons Scientific, Germantown, WI) was decapitated and pithed, and the eyes were removed. The retina was isolated and incubated for ~30–60 min at room temperature (22°C) in 400 μl of enzyme solution containing 12 U/ml papain (Type IV, Sigma, St. Louis, MO) and 5 mM L-cysteine in amphibian Ringer's solution. At the end of the incubation, the retina was rinsed five times with amphibian Ringer's solution, transferred to calcium-free Ringer's solution, and shaken gently until the tissue dissociated. The cells were placed on a lectin-coated coverslip in Ringer's solution and stored in a 17°C incubator. Acutely dissociated cells were used in all experiments.

The retinal slice preparation has been described (Werblin, 1978; Wu, 1987). All surgical and experimental procedures were performed under infrared illumination, and recordings were made from neurons in the ganglion cell layer.

Electrophysiological recordings. The retinal slice or isolated neuron was superfused with amphibian Ringer's solution consisting of (in mM): 111 NaCl, 3 KCl, 2 $CaCl_2$, 1 $MgCl_2$, 10 dextrose, buffered with 5mM HEPES and NaOH to pH 7.8. The internal pipette solution contained (in mM): 110 K-gluconate, 5 NaCl, 1 $MgCl_2$, 5 EGTA, buffered with 5mM HEPES and KOH to pH 7.4. An ATP "regenerating system" (4 mM ATP, 20 mM phosphocreatine, 50 U/ml creatine phosphatase) was added to the internal solution. Pipette resistances were 4–7 MΩ. Access resistance was not compensated, but tip potentials were measured and corrected (Neher, 1992). During the electrophysiological recording, the oxygenated control Ringer's solution or this solution plus antagonist was superfused continuously. In all cases in which antagonists were tested, they were applied first, and then agonists and antagonists were coapplied. A rapid perfusion system (DAD-12, ALA Scientific Instruments) was used. High potassium was perfused onto neurons to measure the time course of drug application. The current increased with a time constant of ~20 msec. Third-order neurons in culture could be identified on the basis of a combination of morphological and physiological characteristics. Ganglion cells were tentatively identified on the basis of soma size and appearance and the amplitude of voltage-dependent sodium currents. Although this was not a positive identification, recordings were made from >100 third-order neurons. With respect to the topic of this paper, the properties of these third-order neurons were the same so that it can be concluded that ganglion cells possessed two glycine responses. In the slice preparation, cells in the ganglion cell layer were patched and assumed to be ganglion cells (Lukasiewicz and Werblin, 1988).

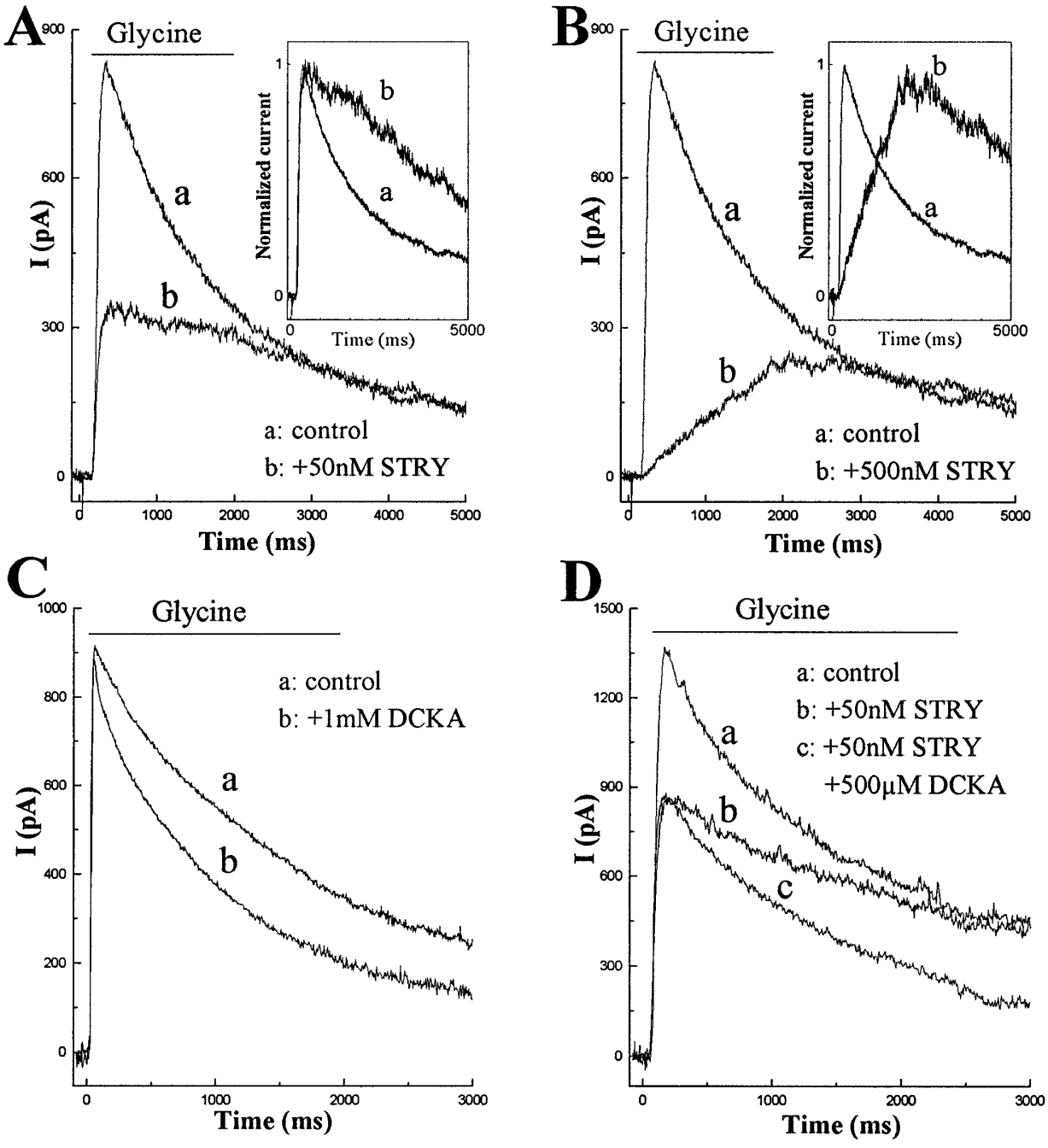

Figure 1. Strychnine and DCKA differentially affect the glycine response. Nanomolar strychnine blocked the fast component of the glycine response. Third-order neurons were voltage-clamped at −10 mV. *A*, Puff applications of 250 μM glycine produced a current with fast and slow components (response *a*); 50 nM strychnine reduced the fast but not the late phase of the glycine response (response *b*). The *inset* shows normalized currents, indicating that low doses of strychnine blocked about half the glycine current without changing the time course of the current. *B*, In the presence of 500 nM strychnine, the fast component of the glycine response was almost completely suppressed, whereas the late component remained. In contrast, DCKA blocked the slow component. *C*, Glycine was applied before (*control*) and during the application of DCKA (response *b*). The late component of the glycine current was preferentially blocked. *D*, Glycine was applied alone, in the presence of 50 nM strychnine, and in the presence of both 50 nM strychnine and 500 μM DCKA. Strychnine reduced the fast component, and DCKA reduced the late component of the glycine current.

Recordings were obtained with an Axopatch 2B amplifier using PCLAMP acquisition software and analysis with Origin software. Cumulative data are expressed as means ± SD. 5,7-Dichlorokynurenic acid (DCKA) was obtained from RBI (Natick, MA). Cyanotriphenylborate was a gift from Dr. J. Bormann (Max Planck Institute fur Hirnforschung, Frankfurt, Germany); all other chemicals were purchased from Sigma.

RESULTS

Glycine antagonists reveal two receptors

The action of glycine was examined on isolated third-order retinal neurons voltage-clamped at −10 mV, a potential that

provided alarge driving force for chloride influx. High concentrations of glycine were applied to promote desensitization, thereby permitting the two glycine receptors to be distinguished. Typical responses to glycine application (250 μM for 2 secs) are shown in Figure 1. Glycine induced an outward current that reached a peak within 200 msec, and then declined because of desensitization (Pan and Slaughter, 1995). Low doses of strychnine (50 nM) suppressed the peak current by a mean of 43% (from a peak current of 458 ± 185 to 196 ± 71 pA; $n = 13$) but did not reduce the late phase of the glycine current (Fig. 1*A*). Normalizing these currents illustrated that 50 nM strychnine did not slow the rise time of the current (Fig. 1*A*, *inset*). This indicated that 50 nM strychnine selectively suppressed a fast component of the glycine response and implied that two subtypes of ionotropic glycine receptor combined to generate the ligand-gated current. At tenfold higher concentrations, strychnine almost completely eliminated the fast component (Fig. 1*B*), leaving a slowly developing current (Fig. 1*B*, *inset*) that was relatively insensitive to strychnine ($n = 15$).

The glycine responses were not blocked by 20 μM SR95531, a GABA antagonist that completely blocked GABA-evoked currents in these cells. Therefore, the strychnine-insensitive glycine current response was not attributable to cross-over to the GABA receptor.

Although low doses of strychnine suppressed the fast current component, DCKA produced the opposite effect. DCKA, at low concentrations, is a selective blocker of the glycine recognition site at the NMDA receptor (IC_{50} = 560 nM) (Kemp et al., 1988), but we found that high concentrations (500 μM–1 mM) selectively blocked the slow, inhibitory glycine current. This is illustrated in Figure 1*C*, which displays the glycine current with and without DCKA. Note that DCKA did not block the fast phase of the glycine-evoked current but significantly reduced the slow current. The relative effects of strychnine and DCKA are shown in Figure 1*D*. Strychnine (50 nM) reduced the peak current but not the late current. Coapplication of DCKA resulted in no further reduction in the peak current, but the late phase of the glycine current was suppressed. This illustrates the diacritical kinetics of the strychnine-sensitive and DCKA-sensitive glycine currents.

DCKA did not produce its effect by increasing the decay rate (desensitization) of the fast component. The decay was measured by fitting the glycine current, from the time of the peak current to the 2 sec mark, with a single exponential. The decay time constant of the fast component in the presence of 500 μM DCKA was 950 ± 183 msec ($n = 13$). If the fast component was determined by isolating the strychnine-sensitive glycine current (control current minus the remaining current in the presence of 50 nM strychnine), this current had a decay time constant of 918 ± 120 msec ($n = 8$), very close to the value in the presence of DCKA. This suggests that the time course of the fast glycine current was not abbreviated by DCKA. To check the sensitivity of this experiment, we applied cyanotriphenylborate, which is a use-dependent chloride channel blocker known to shorten the time course of the glycine response (Rundström et al., 1994). The time constant of the fast component was decreased to 306 ± 86 msec ($n = 13$) in the presence of 20 μM cyanotriphenylborate.

These results suggest that the transient and sustained components of the glycine current arose from receptors with distinct kinetics and pharmacology: the strychnine-sensitive current had fast onset and fast desensitization rates, whereas the DCKA-sensitive current had slow onset and slow desensitization rates.

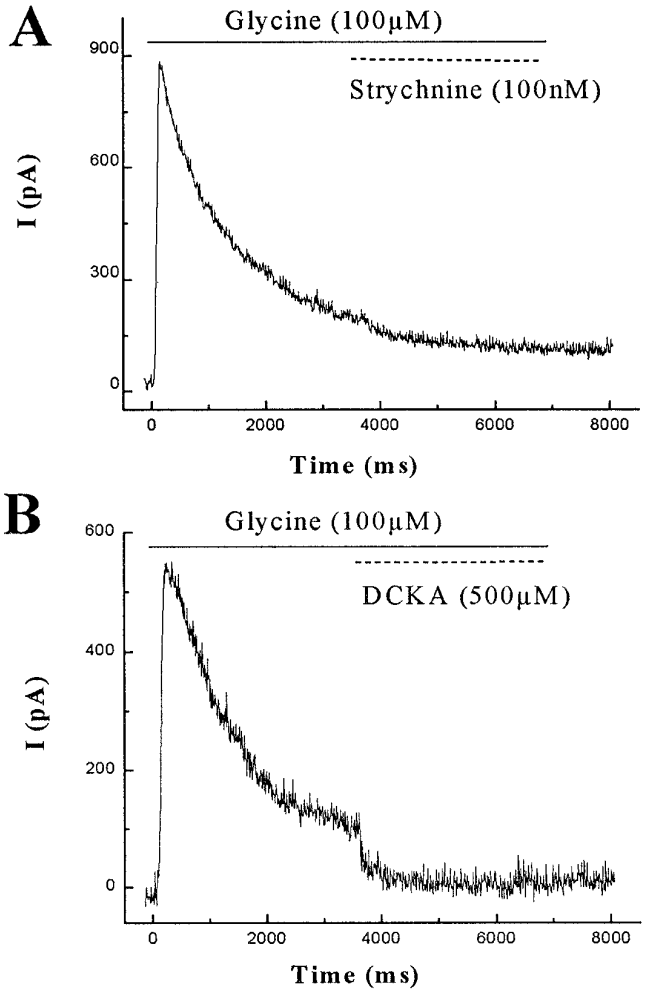

Figure 2. Selective suppression of the late phase of the glycine current. The late phase of the glycine current was suppressed by DCKA but not nanomolar strychnine. Glycine was applied for 7 sec. At the midpoint of glycine application, either 100 nM strychnine (*A*) or 500 μM DCKA (*B*) was coapplied.

The ratio of glycine currents induced by the two putative receptors changed with time. In the first 500 msec after glycine application, the strychnine-sensitive response predominated, whereas after 2 sec the current was caused almost exclusively by the DCKA-sensitive response. This was confirmed by applying antagonists during the late phase of the glycine current (Fig. 2). Glycine was applied for 3.5 sec and then antagonist was added. DCKA blocked this late phase of the glycine current (Fig. 2*B*), whereas low concentrations of strychnine were without effect (Fig. 3*A*).

Properties of the two glycine receptors

To characterize the pharmacology of the two glycine currents, the actions of agonists and antagonists were investigated. Glycine responses were determined in the presence of various strychnine concentrations. The fast current (measured as the peak current) and the slow current (measured after 2 sec of glycine application) were monitored. To pool results from different cells, the currents in the presence of strychnine were normalized to the peak glycine current in the absence of strychnine. The mean IC_{50} value of the fast component was almost two orders of magnitude lower than that of the slow component (Fig. 3). Even 10 μM strychnine did not completely block the slow current component.

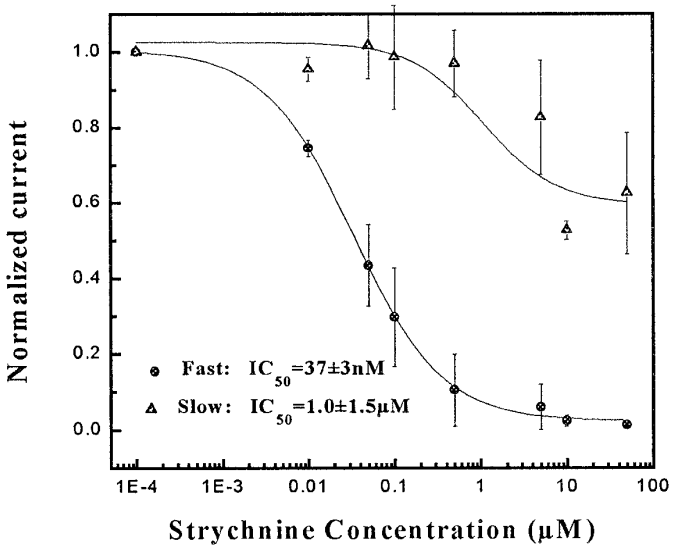

Figure 3. Strychnine potency was different for the fast and slow glycine currents. Glycine (250 μM) was applied in the presence of various concentrations of strychnine. The effects on the fast component (measured at the peak of the glycine current) and on the slow component (measured after 2 sec of glycine application) were plotted and the data fitted to:

$$I_{exp} = I_{max} - \frac{I_{max} - I_{min}}{1 + (IC_{50}/\text{strychnine concentration})^n},$$

where I_{max} is the maximum amplitude glycine-induced current and I_{min} is the smallest.

Second, we compared the glycine affinity of the two receptors. Different concentrations of glycine were puffed either in the absence or presence of 1 μM strychnine, thus distinguishing between the fast and slow components (see Fig. 6*A, B*). Figure 4*A* displays dose–response curves from one cell. Normalized curves from seven cells are shown in Figure 4*B*. The affinity of glycine was not changed by strychnine. It indicates that the two receptors have similar glycine affinities. Fitting the data to the logistic equation yields an apparent EC_{50} of 43 ± 1.3 μM and a Hill coefficient of 1.9 ± 0.1 (n = 7), which is comparable to published values for the inhibitory glycine receptor (K_D = 90 μM; n = 1.8 in rat hypothalamic neurons) (Akaike and Kaneda, 1989).

We did not find agonists that selectively activated only one component of the glycine response. L-serine, L-alanine, β-alanine, and taurine are all glycine analogs that are known to activate the retinal glycine receptor (Bolz et al., 1985; Tauck et al., 1988; Pan and Slaughter, 1995). All analogs were found to produce transient and sustained currents in which the transient component was blocked by low concentrations of strychnine (50 nM), similar to the action of glycine (Fig. 5.) The stereoisomers D-alanine and D-serine were without effect at 1 mM. There was some indication in the data that the ratio of fast to slow currents was smaller for L-serine and L-alanine than for glycine, taurine, or β-alanine, suggesting that the relative potencies of these analogs at the two sites may differ; however, these differences were slight and did not proffer selectivity.

Third, the reversal potentials of the two components were compared by puffing glycine while clamping neurons at potentials ranging from −30 to −90 mV. The peak glycine current in control Ringer's solution was used as a measure of the fast component (Fig. 6*A*). To measure the slow component, glycine was applied in the presence of 1 μM strychnine (Fig. 6*B*). The *I–V* curves from

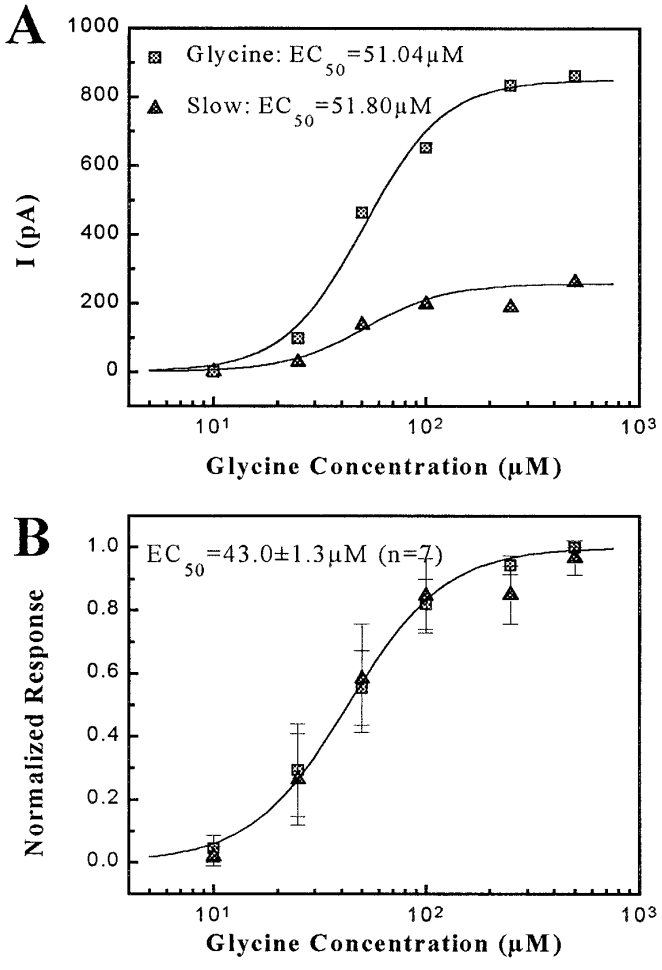

Figure 4. The potency of glycine was similar at both receptor sites. *A*, The fast glycine-induced current (measured as the fast, peak current) and the slow glycine-induced current (measured in the presence of 1 μM strychnine and at the 2 sec point during glycine application) were plotted for one cell. The fast component produced a larger current, but the EC_{50} values of both components were similar. *B*, Data on both components from seven neurons were normalized and plotted. The curves closely overlapped and were fitted to the following equation:

$$I_{exp} = \frac{I_{max} - I_{min}}{1 + (EC_{50}/\text{glycine concentration})^n} + I_{min},$$

where n is the Hill coefficient.

these two data sets are shown in Figure 6*C*, illustrating that the fast and slow components reversed at approximately the same voltage. The reversal potential for the fast current was −65 ± 7 mV (n = 12), and the slow component reversed at −63 ± 6 mV (n = 5). The reversal potential was close to the calculated chloride equilibrium potential (based on concentration) of −72 mV. Thus both receptors probably govern permeability to chloride. This result also indicated that the slow current was not attributable to an electrogenic carrier.

Synaptic glycinergic responses

The retinal slice was used to determine the effects of the two glycine receptors on the light-evoked synaptic responses. Recordings were obtained from neurons (n = 5) in the ganglion cell layer. These cells were clamped to −10 mV, which was slightly below the reversal potential of EPSCs. Picrotoxin (100 μM) was continuously bath-perfused to block GABAergic IPSCs. Therefore, light-

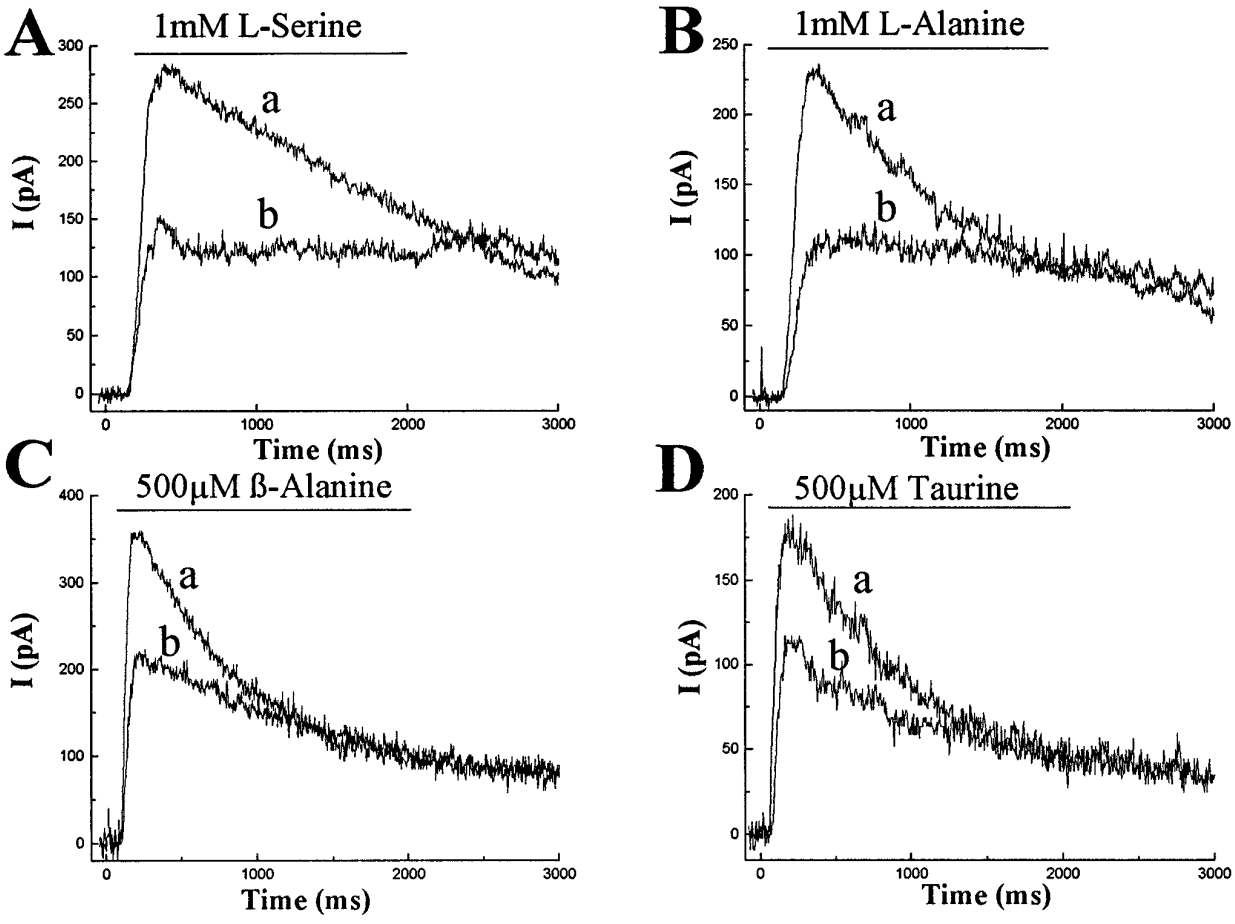

Figure 5. Selective agonists for the two receptors were not identified. L-Serine, L-alanine, β-alanine, and taurine each activated a fast current component that was suppressed by 50 nM strychnine and a slow current that was insensitive to nanomolar strychnine. The larger current in each panel (*a*) represents the agonist alone; the smaller current (*b*) is the agonist in the presence of 50 nM strychnine.

evoked outward currents represented glycinergic synaptic responses (Fig. 7). When 1 μM strychnine was applied, the fast component of outward current was suppressed, leaving a slow outward current (Fig. 7*B*). A fast inward current was also revealed, indicating that the strychnine-sensitive glycinergic IPSC opposed a fast EPSC (Fig. 7*B*). The remaining outward current was largely blocked by 10 μM strychnine (Fig. 7*D*). A more prolonged EPSC was revealed. Digital subtraction of the currents before and during strychnine application manifested the waveforms of the glycinergic IPSCs (Fig. 7*B*, *dotted lines*) generated by the fast response (Fig. 7*B*) and by both fast and slow acting responses (Fig. 7*D*). Thus, the 1 μM strychnine-sensitive, fast glycine current reduced a rapid excitatory current, whereas a more prolonged excitatory current was suppressed by the 10 μM strychnine-sensitive, slower glycine current. Similar results were obtained from four other neurons in the ganglion cell layer. We could not use long-duration light pulses that may have accentuated the influence of the slow current, because light adaptation is particularly pronounced in the retinal slice preparation. Also, because NMDA receptors are implicated in light responses of ganglion cells (Mittman et al., 1990; Cohen and Miller, 1994), we used high concentrations of strychnine, rather than DCKA, to suppress the slow component of the glycine response. Nevertheless, results from the slice preparation indicated that the two glycine responses both played a role in synaptic input to retinal ganglion cells.

DISCUSSION

Glycine receptor subtypes

These experiments demonstrate that there are two synaptic glycine responses in retinal ganglion cells. The responses have different kinetics and can be separated by antagonists, suggesting two glycine receptors. The different kinetics implies that the two putative glycine receptors can differentially suppress fast or slow excitatory currents, and this is supported by retinal slice studies on endogenous synaptic activity.

The temporal differences between the two responses could be described by a simple model that assumed two receptors, each activated (open channel) and inactived (desensitized) with first-order kinetics (Fig. 8). This is clearly an oversimplification of the microscopic events but provides a description of the macroscopic currents. Figure 8*A* displays the model and the equations used. Figure 8*B* illustrates how the fast and slow components were isolated and fitted to this descriptor. Fifty nanomolar strychnine partially blocked the fast glycine current (Fig. 8*B*, curve *d*), whereas the slow current was unaffected (Fig. 3). Therefore, subtraction from the control current (*a*–*d*) gave a scaled representation of the fast current (*b*). In the presence of 20 μM

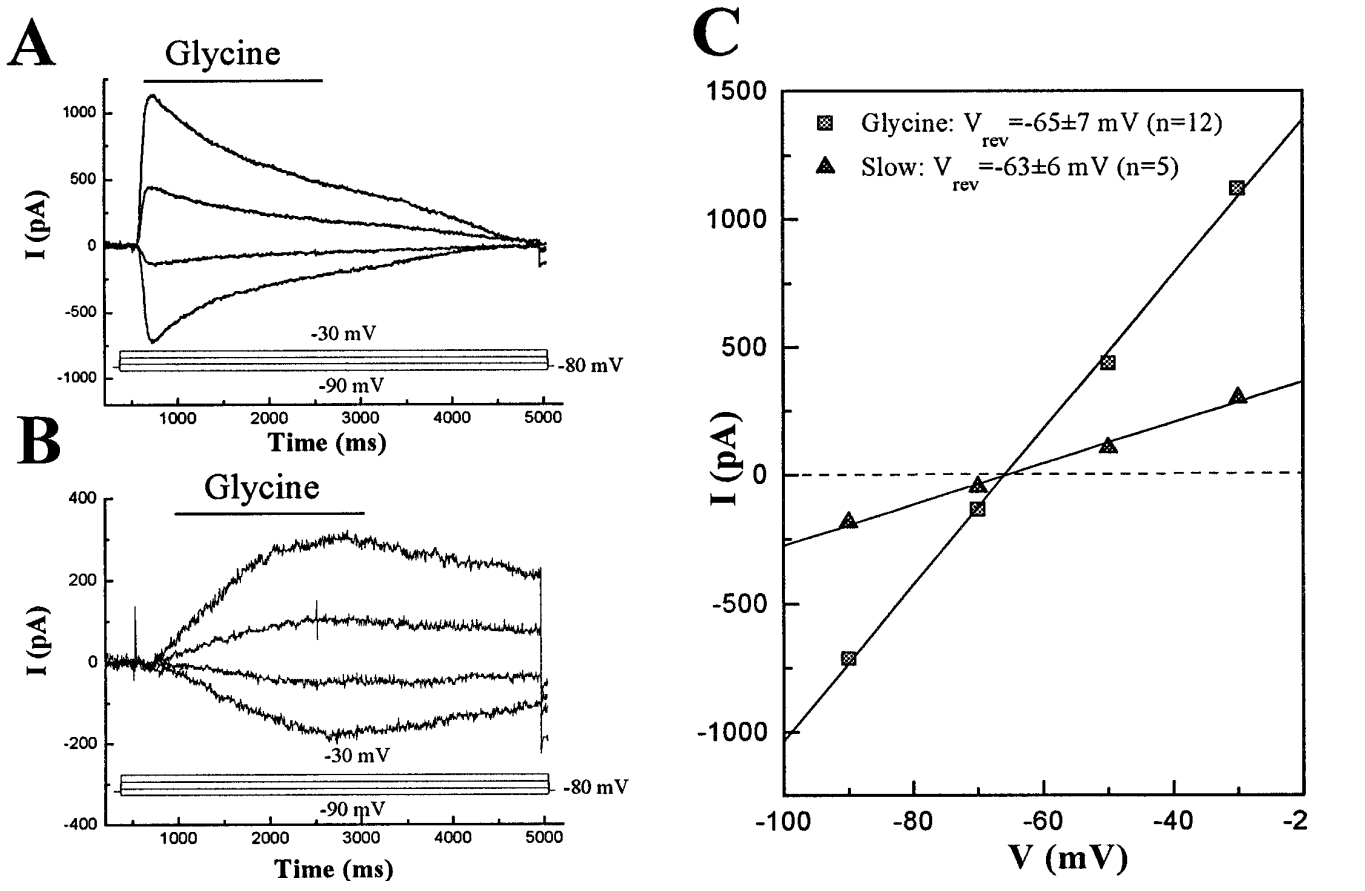

Figure 6. The fast glycine-induced current was associated with a larger conductance than the slow current, but both had similar reversal potentials. *A*, The neuron was held at −90 mV, −70 mV, −50 mV, and −30 mV, and at each voltage 100 μM glycine was applied. *B*, A similar protocol was used but in the presence of 1 μM strychnine. *C*, Using data such as that illustrated in *A* and *B*, mean peak currents for the fast and slow components were plotted.

strychnine, the fast current was totally suppressed but the slow current was only partially suppressed. This provided a scaled representation of the slow response. Each component could be fitted by the product of two exponentials, representing the concomitant activation and desensitization processes. These components were used to reconstruct the original glycine current (Fig. 8*C*). The rise time of the fast glycine current was 40 msec, similar to but slightly slower than the current produced by a potassium puff. The difference is attributable to the effect of desensitization; however, this perfusion delay was much faster than any other time constant, indicating that perfusion delay did not significantly alter these values. Activation of the slow component had a time constant of almost 3 sec. The desensitization time constant for the fast component was 1 sec in 250 μM glycine, which agrees fairly well with studies on rat retinal ganglion cells (4.4 sec for 100 μM glycine) (Tauck et al., 1988) and rat hypothalamic neurons (1.5 sec for 100 μM glycine) (Akaike and Kaneda, 1989). The desensitization time constant of the slow component was derived from the fit to be 8 sec, although this was not determined experimentally. This rate is similar to a slow desensitization rate found in hypothalamic neurons (5.5 sec) (Akaike and Kaneda, 1989). The total glycine-induced current could be reasonably reconstructed by scaling and summation of these two components (Fig. 8*C*). The two components could also be used to fit the glycine responses at various intermediate levels of strychnine (not shown). This supports the proposal that the total glycine-induced current results from these two discrete components.

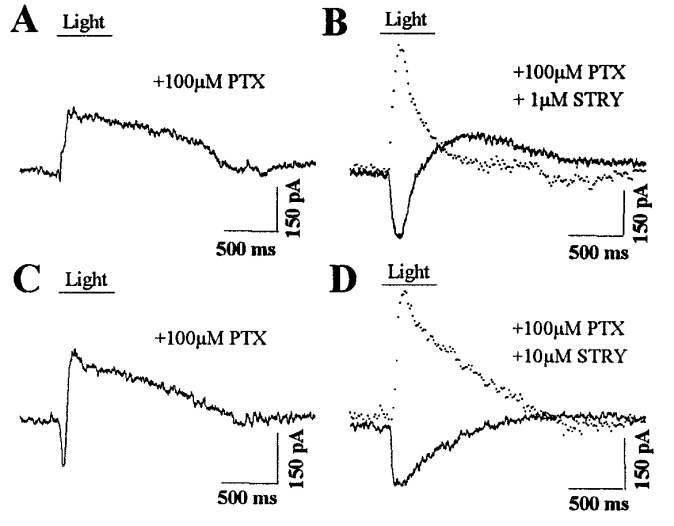

Figure 7. Light-evoked glycinergic IPSPs in retinal ganglion cells consisted of fast and slow components. Neurons in the ganglion cell layer of the retinal slice were clamped at −10 mV, and 100 μM picrotoxin (*PTX*) was applied continuously. *A*, Light stimulation elicited an outward current. *B*, After application of 1 μM strychnine the outward current was reduced, leaving a fast inward current followed by a slow outward current. The *dotted curve* shows the difference current, which estimates the fast glycinergic current blocked by 1 μM strychnine. *C*, *D*, When the same protocol was used in the presence of 10 μM strychnine, much of the outward current was blocked. The *dotted line* estimates the outward, glycinergic current that was blocked.

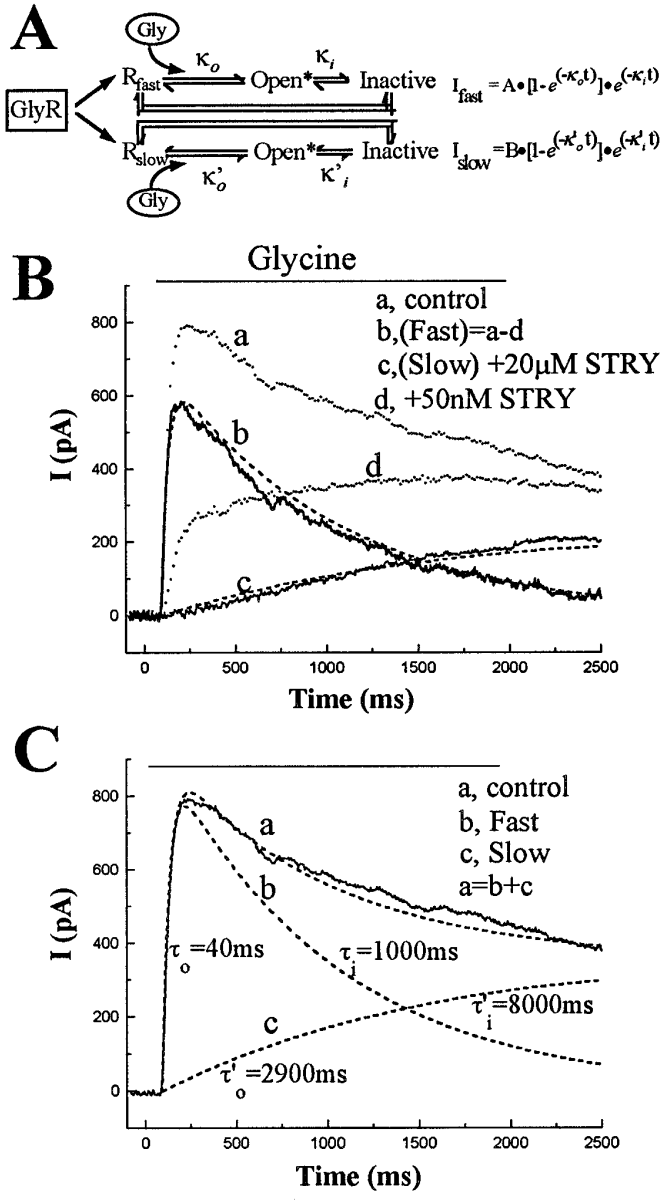

Figure 8. Modeling of the glycine currents. *A*, The slow and fast currents produced by glycine puffs were fitted to the above equations where I is glycine-induced current, t is time, κ_o is the rate constant for current onset, κ_i is the rate constant for the decline of the current, and A and B are scaling factors. *B*, The fast component was determined by a partial block with 50 nM strychnine (*d*). This was subtracted from the total current (*a*) to obtain a scaled fast glycine-induced current (*b*). The scaled slow component was determined by applying glycine in the presence of 20 μM strychnine, which blocked all of the fast and about half of the slow component. *C*, The time course of the fast (*b*) and the slow (*c*) calculated components are shown, as is the fit to the original glycine current (*a*).

Alternative mechanisms of action

Strychnine and DCKA could change the kinetics of a single type of glycine receptor, giving the appearance of two receptors. For example, the slow glycine current in the presence of strychnine could result from a slow dissociation of strychnine leading to a new equilibrium between glycine and strychnine; however, the rise time of the glycine response in the presence of 50–100 nM strychnine was not slowed, despite the ~50% suppression of the transient response (Fig. 1). Also, low concentrations of strychnine had no effect on the late portion of the glycine current (Figs. 1, 3). The time course of the total glycinergic current could be fitted with the same two components in the presence of various concentrations of strychnine (Fig. 8). This would not be true if the kinetics of one component was altered by these drugs. Similarly, DCKA could have produced an apparent suppression of the slow component by increasing the rate of desensitization of the fast component, but our experiments demonstrated that DCKA did not reduce the time constant of the fast component of the glycine response. These experiments suggest that strychnine and DCKA are identifying two distinct responses, probably resulting from two subtypes of the glycine receptor.

Another possibility is that there are two conformations of the same receptor, one in which strychnine binds with high affinity and the other preferentially binds DKCA. This cannot be excluded by the data, although the similar glycine affinity of the two receptors would argue against this possibility if it is analogous to the high- and low-affinity states of the acetylcholine receptor (Boyd, 1987).

Agonists and antagonists of glycine receptors

Comparing our data to that on glycine receptors with known subunit compositions manifests both similarities and differences. We did not find a difference in the agonist sensitivity of the two receptors. Glycine reportedly has similar affinity for the neonatal and adult glycine receptor in spinal cord (Becker et al., 1988) but much lower affinity for α_1 receptor mutants associated with hereditary hyperekplexia (Langosch et al, 1994; Rajendra et al., 1994). L-serine, L-alanine, β-alanine, and taurine were all found to produce transient and sustained current components. This contrasts with *in vitro* expression systems in which taurine stimulated glycine receptors containing the α_1 subunit, but was ineffective when α_2 or α_3 subunits were expressed (Betz, 1991; Malosio, 1991a). Thus, the DCKA-sensitive retinal glycine receptor is similar to the neonatal spinal glycine receptor in its low strychnine sensitivity but dissimilar in that only the former is taurine-sensitive.

A theme running through much of the literature on glycine receptor subtypes is a differential sensitivity to strychnine. For example, the glycine receptor in the neonatal rat spinal cord is less strychnine-sensitive than that in the adult (Becker et al., 1988; Betz, 1991). In neonatal rat sympathetic preganglionic neurons, Wu et al. (1995) found two glycine currents: a strychnine-sensitive hyperpolarizing current and a strychnine-insensitive depolarizing current. The former had faster kinetics, somewhat analogous to our findings. In neonatal rat hippocampal neurons, Ito and Cherubini (1991) found a nondesensitizing glycine receptor with a low strychnine affinity (apparent K_D = 0.35 μM). This is similar to the slower retinal glycine response (IC_{50} = 1 μM). The strychnine sensitivity of the fast retinal glycine response (IC_{50} = 37 nM for 250 μM glycine) is similar to that of that in recombinant α_1 subunits (IC_{50} = 30 nM for 200 μM glycine) (Schmieden and Betz, 1995). Lewis et al. (1991) found desensitizing and nondesensitizing glycine currents in isolated rat medullary neurons. The glycine EC_{50} was 26 μM for the nondesensitizing and 69 μM for the desensitizing current. They found that the strychnine IC_{50} was 15 nM and 500 nM (using 100 μM glycine) for the desensitizing and non-desensitizing currents, respectively. This is similar to our results, but unlike our observations, they noted that 1 μM strychnine completely blocked all glycine currents. Overall, there is a consensus that slow acting, less desensitizing glycine currents are less strychnine-sensitive. It remains to be seen whether all of these slow glycine receptors are selectively inhibited by DCKA.

Glycine receptor subtypes in retinal physiology

In the salamander retina, glycine is contained in approximately one third of amacrine cells (Yang and Yazulla, 1988), which provide feedforward inhibition to ganglion cells. Glycinergic inhibition to ganglion cells has two components. One glycinergic input to ganglion cells provides fast, transient IPSPs at the onset and offset of the light response (Belgum et al., 1984). Another glycinergic input is tonic (Miller et al., 1981). The retinal slice data (Fig. 7) indicate that both responses are localized to synapses at single ganglion cells and are differentially sensitive to strychnine. Presumably, the relative activity of these two receptors can modulate the temporal aspects of ganglion cell light responses, acting as high-pass (DCKA-sensitive receptor) or low-pass (strychnine-sensitive receptor) filters of synaptic excitation.

Comparing GABA and glycine receptors

The division of glycinergic receptors into subtypes with different kinetics has a precedent in the GABA receptor. The $GABA_AR$ initiates a large, fast-onset, rapidly desensitizing chloride current, whereas the $GABA_CR$ is relatively nondesensitizing, with slow activating and deactivating kinetics (Cutting et al., 1991; Qian and Dowling, 1993; Pan and Lipton, 1995). Amin and Weiss (1994) found that $GABA_\rho$ subunits expressed in *Xenopus* oocytes produced a very slow activation (half-time of 9 sec at the GABA EC_{50}) and deactivation (half-time of 12.5 sec). Although it is often proposed that the ionotropic receptors are responsible for fast synaptic signals and the metabotropic receptors relay slower signals, GABA and glycine receptors reveal that there is a broad kinetic spectrum within the ionotropic receptor system.

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