**Competition Between Lidocaine and One of Its Metabolites, Glycylxylidide, for Cardiac Sodium Channels**

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The modulated receptor hypothesis states that sodium channels have a specific receptor for antiarrhythmic drugs. Therefore, two agents that block sodium channels by binding to this receptor are expected to compete for occupancy. Glycylxylidide (GX) is a deethylated metabolite of lidocaine that accumulates in patients on lidocaine therapy. In single, voltage-clamped cardiocytes, GX, like lidocaine, blocked cardiac sodium channels in a use-dependent manner. However, its kinetics of recovery from block were markedly different from lidocaine: at potentials between -80 and -100 mV, GX-blocked channels recovered faster and more completely than lidocaine-blocked channels but recovered more slowly at more negative potentials (-120 to -140 mV). If lidocaine and GX compete for a common receptor, then there are conditions in which addition of a “faster” drug to a “slower” drug will produce less block than the slower drug alone. At potentials between -120 and -140 mV, addition of GX (slower drug) to lidocaine always increased the level of block, but addition of lidocaine to GX decreased the block in four of nine experiments and did not increase it in three of nine experiments. Conversely, at potentials between -80 and -100 mV, addition of lidocaine (slower drug) to GX always increased block, whereas addition of GX to lidocaine reduced the level of block in five of 16 experiments and did not increase it in seven of 16 experiments. Thus, upon addition of more blocker, the sodium current increased in 36% of cases or did not decline in 76% of cases. These results can be explained by the modulated receptor hypothesis with two drugs competing for the same receptor. This is the first demonstration of an antiarrhythmic drug metabolite that can competitively displace its parent compound in vitro. Whether competitive displacement of a parent compound by a metabolite also occurs in humans will require clinical studies. *(Circulation 1988;78:692–700)*

There is uniform acceptance of the antiarrhythmic efficacy of lidocaine in acute myocardial infarction. However, in many cases, arrhythmias are initially suppressed by lidocaine, but with continued administration, some patients no longer respond or develop ventricular fibrillation. Indeed, Chopra et al. have documented that in 10% of patients whose arrhythmias had responded quickly, resistance to lidocaine developed on the 2nd day of therapy. It is often assumed that this occurs because of progression of the underlying disease, but in some cases, there is no evidence that the patient’s clinical condition has changed, which suggests another cause may underlie the resistance to therapy.

We have recently observed two patients (see “Patients and Methods”) who demonstrated this phenomenon while being treated for recurrent ventricular tachycardia in the absence of acute ischemia. These patients initially responded to lidocaine but later became refractory. The plasma lidocaine concentrations were similar at the time of the “response” and later when the patients were refractory. In both patients, the deethylated lidocaine metabolite glycylxylidide (GX) was undetectable during their initial response to lidocaine, but the failure of lidocaine therapy was accompanied by the appearance of GX in the blood.

These observations raise the possibility that accumulation of GX may be responsible for the loss of responsiveness to lidocaine. It is conceivable that structural analogues of lidocaine can bind to
common receptor site associated with sodium channel block and that the less active analogue would interfere with the action of the parent compound. Plasma concentrations of GX can equal those of lidocaine during continued administration of lidocaine, and myocardial levels of both drugs can exceed plasma levels by a factor of 2–3.6

The modulated receptor hypothesis states that local anesthetic and antiarrhythmic drugs block sodium channels by binding to a common receptor.7-9 Consequently, two agents that bind to this receptor are expected to compete for occupancy. In a mixture of two such drugs, a fraction of the sites will become occupied by each drug. Under certain conditions, some sites that were occupied by the first drug will instead become occupied by the second drug. If one drug dissociates from the receptor faster than the other, then an increase in sodium current could result even in the presence of a higher blocker concentration. These principles have been shown true for mixtures of lidocaine, bupivacaine, and quinidine10 and for propafenone, lidocaine, and quinidine11 in heart, and for benzocaine and lidocaine in nerve and heart.12,13

To test whether GX competes with lidocaine and reduces the total block of sodium channels, we addressed the following experimental questions by directly measuring current through sodium channels under voltage-clamp conditions: 1) does GX block cardiac sodium channels, 2) is the block use dependent, 3) what is the voltage dependence of recovery of GX-blocked channels, 4) can competition between GX and lidocaine give less total block than lidocaine alone under certain conditions?

Our results indicate that GX does block sodium channels in a use-dependent manner. Recovery of currents from block depends strongly on membrane potential, and under appropriate conditions of timing and membrane potential, GX can reduce the block of sodium channels by lidocaine.

Patients and Methods

These experiments were performed in 119 cells from 49 different cell isolations. Isolated ventricular myocytes were obtained from guinea pigs (weighing 200–300 g) by a collagenase dissociation technique.14 Briefly, hearts were perfused through the aorta and coronary arteries with a nominally calcium-free solution for approximately 1 minute followed by a 10–20-minute perfusion with solution containing 0.15% (123–153 units/mg, Worthington type II) collagenase and 50 μM CaCl₂. After the perfusion with enzyme, the heart was minced and gently agitated in a shaking water bath at 37° C. Cells were washed three times in enzyme-free solution supplemented with 1 mg/ml bovine serum albumin (Sigma Chemical, St Louis, Missouri) and 0.05, 0.1, and 0.5 mM CaCl₂, respectively.

Solutions

The solution for cell isolation and storage was Joklik-modified minimum essential medium (GIBCO Laboratories, Chagrin Falls, Ohio). Solutions were equilibrated with 95% O₂-5% CO₂. The pH was adjusted to 7.40 at 37° C with NaOH. Recordings were made in a bath solution containing (mM) 20 NaCl, 110 CsCl, 2 MgCl₂, 1 CaCl₂, 3 CoCl₂, 10 d-glucose; 10 N-2-hydroxyethylpiperazine-N'-2-ethanesulfonic acid (HEPES); 10 tetraethylammonium chloride (TEA); pH was adjusted with CsOH to 7.35 at 22° C. The pipette solution contained (mM) 110 CsF, 10 NaF, 30 CsCl, 2 MgCl₂, 10 HEPES, 2 ethyleneglycol-bis-(β-aminoethyl ether)-N,N,N',N'-tetraacetic acid; pH was adjusted to 7.35 with CsOH at 22° C.

Drugs

GX and lidocaine were obtained from Astra Pharmaceutical, Worcester, Massachusetts. Glycylxylidide is also referred to as glycinexyldide. Concentrations of lidocaine tested ranged 9.3–74 μM (2.5–20 μg/ml), and GX concentrations tested ranged 9.4–155 μM (2–33 μg/ml), and we have observed sodium channel blocking effects by both drugs at all concentrations tested. Under the conditions described, 37 μM lidocaine and 47 μM GX induced approximately 50% block of sodium current; therefore, we have used these concentrations in all experiments presented except when stated otherwise.

Electrical Recording

The dissociated cells were transferred to a 0.5-ml chamber mounted on the stage of an inverted Nikon microscope. After the cells had settled to the bottom of the chamber, they were superfused at 1 ml/min. The bath temperature was maintained at 16–18° C (±0.2° C for a particular experiment) by a Peltier-effect device. The cells used in these experiments were rod shaped with clear striations and were quiescent in 1 mM [Ca⁺]o.

All recordings were made in the whole-cell voltage-clamp configuration of the “patch-clamp” technique with an Axopatch voltage clamp (Axon Instruments, Burlingame, California). Electrodes had resistances between 0.3 and 1.0 MΩ, and series resistance was compensated by an analog circuit. Capacitative and linear leakage currents were subtracted by an analog circuit and in some cases by digital subtraction on a computer. Before forming a giga-seal, current flowing between the grounded bath and the pipette was calibrated to zero by applying a small (<1 mV) offset voltage to balance the junction potential caused by the difference in the pipette and bath solutions. Command potentials have not been corrected for junction potentials.

After formation of a GΩ seal, the patch of membrane under the electrode was ruptured by a brief pulse of suction. Under these conditions, currents flowing through the entire cell surface were recorded.
Potassium currents were eliminated by replacing potassium with cesium and by TEA. Calcium-dependent currents were eliminated by internal fluoride and external cobalt and magnesium. Sodium current was the only measurable transmembrane current under the conditions used.

Voltage-clamp command pulses were generated by a twelve-bit digital-to-analog converter. Membrane currents were filtered at 5 kHz by a four-pole Bessel filter and digitized at 20 kHz by a twelve-bit analog-to-digital converter controlled by an IBM PC-AT (Danbury, Connecticut).

**Data Analysis**

In most cases, we have avoided pooling data from separate experiments and have endeavored to make important comparisons between control and drug in a single cell (paired comparisons). It was not possible, however, to obtain a complete set of data (control + drug + recovery) in each cell studied. Therefore, in some cases, we pooled data from a number of cells to allow comparisons. In these cases, a grouped t test was used to compare two means. A probability level of 5% (p<0.05) was considered statistically significant. Curve fitting was accomplished by a nonlinear least-squares parameter estimation procedure based on the Marquardt-Levenberg method.\(^16\)

**Adequacy of Voltage Clamp of Sodium Current in Ventricular Myocytes**

The use of individual heart cells facilitates the achievement of rapid and uniform control of membrane potential, but this alone is not sufficient for the quantitative voltage clamp of sodium channel currents during conditions of normal extracellular sodium concentrations at 37°C. In these cells, it is necessary to reduce both the transmembrane sodium concentration gradient and the temperature to measure sodium current. These conditions must be borne in mind when interpreting our results (see "Discussion"). During these conditions, the current-voltage relations for sodium current were graded functions of voltage with a peak near −20 mV. These findings are indicative of adequate voltage control as shown by Brown et al.\(^17,18\) when two suction electrodes are used. Furthermore, when the maximum conductance was reduced by inactivating sodium channels, the current-voltage relations were simply scaled in magnitude. This finding is evidence that there were no large voltage errors associated with the series resistance between the pipette interior and the cell interior.

**Patients**

Patient 1 is a 55-year-old white woman (60 kg) with a history of postpartum cardiomyopathy and documented frequent episodes of unsustained ventricular tachycardia that causes episodes of dizziness. She had been treated successfully with oral tocainide until she developed a rash after which she had to discontinue therapy. Two days later, she experienced the sudden onset of tachycardia and became weak and diaphoretic. An esophageal electrogogram documented the presence of atrioventricular dissociation with a ventricular rate of 175 beats/min. Five minutes after administration of a loading infusion of lidocaine (8 mg/min for 30 minutes), she converted to sinus rhythm, and a blood sample was collected. The concentration of lidocaine in the sample was 3.6 μg/ml, whereas the concentration of the metabolite was below the detection limit of the assay (20 ng/ml). Lidocaine infusion was maintained at 1.5 mg/min. Approximately 21 hours later, the patient’s clinical condition was unchanged, but she again developed sustained ventricular tachycardia at a rate of 165 beats/min. The lidocaine concentration in plasma at the time of arrhythmia recurrence was 4.4 μg/ml and that of GX was 0.9 μg/ml.

Patient 2 is a 67-year-old white man who suffered an anteropapical myocardial infarction 3 years before his admission for evaluation of presyncope. During telemetric monitoring, he was found to have frequent runs of unsustained ventricular tachycardia and a mean frequency of 400 ventricular premature depolarizations/hr. An infusion of lidocaine totally suppressed his arrhythmia for the next 8 hours. His lidocaine level 4 hours after beginning the infusion was 2.9 μg/ml, and the GX concentration was 0.1 μg/ml. His arrhythmia slowly returned within 8–24 hours with an average of 300 ventricular premature depolarizations/hr and occasional runs of unsustained ventricular tachycardia. His plasma lidocaine concentration at that time was 3.1 μg/ml, and the GX concentration was 0.8 μg/ml.

**Results**

**Time Course of Block by Lidocaine and Glycylxylidide**

Block of sodium channels by antiarrhythmic agents often consists of at least two components: tonic block and use-dependent block.\(^19\) Figure 1 shows the time course of block development after lidocaine (Panel A) or GX (Panel B) was added to the bath perfusate. The membrane potential between pulses was held at −120 mV (to remove inactivation), and 10-msec pulses to −10 mV were applied every 115 msec for 14 pulses. These trains of 14 pulses were applied every 20 seconds during the wash in of drug. The peak inward current during the first (circles) and 14th (squares) pulses are shown. The decrease of current during the first pulse is an indication of the amount of tonic block, and the reduction of sodium current during the 14th pulse indicates use-dependent block of sodium channels. Figure 1A shows the time course of block caused by 37 μM lidocaine. Lidocaine was added to the bath perfusion at zero time in the graph. As lidocaine washed into the bath, the magnitude of sodium current decreased during the trains, but sodium current during the first pulse of each train was not
much affected. The inset shows current recorded during trains before (a) and during (b) lidocaine exposure. Thus, this concentration of lidocaine caused little tonic block but caused marked use-dependent block, and steady-state block was achieved within 5 minutes of exposure to lidocaine.

Figure 1B shows the onset of block as 47 μM GX washes into the bath. GX was added to the bath perfusion at the zero time and was discontinued at 8 minutes (arrow). The inset shows individual current records in a train before (c) and during (d) exposure to GX. The steady-state block of sodium channels by 47 μM GX was achieved in about 5 minutes and was similar to that of 37 μM lidocaine under these conditions. Figure 1B also shows that the block of channels by GX is readily reversible upon washout of GX. The reversibility upon washout of drug was similar in lidocaine (data not shown). All subsequent measurements described in this study were made during a steady-state drug effect.

Development of Use-Dependent Block

The development of use-dependent block during a train indicates that the interval between pulses was too short to allow unblocking of sodium channels before the next pulse. Figure 1C shows that similar degrees of use-dependent block can be induced by trains of pulses in either 37 μM lidocaine or 47 μM GX. The voltage-clamp train protocol was the same as in Figure 1. The use-dependent block approached steady state in 14 pulses; however, to ensure attainment of a steady-state level of block, we have used 50 pulses to induce block in experiments described in the next section. Figure 1C shows that 37 μM lidocaine or 47 μM GX caused approximately the same amount of use-dependent block with little tonic block.

Recovery From Lidocaine and Glycylxylidide Block

After termination of a pulse train, blocked channels recover with a rate that depends on membrane potential.20,21 Figure 2 shows the magnitude of the peak sodium current as a function of time after a train. Each curve represents recovery at a different membrane potential after a train of fifty 5-msec pulses to +5 mV. Panel A shows recovery from normal train-induced inactivation in control. The recovery process in control experiments was more than 90% completed in 250 msec (93% at −90 mV and 95% at −140 mV).

The rate of recovery in 37 μM lidocaine (Figure 2B) or 47 μM GX (Figure 2C) depended on membrane potential. In general, hyperpolarization increased the rate of recovery. However, channels blocked by GX appeared to recover more slowly at −140 mV than channels blocked by lidocaine. Conversely, at −90 or −100 mV, recovery in GX was usually faster and more complete than in lidocaine. Table 1 shows averaged variables for experiments in which recovery was measured at −90 mV or at −140 mV. We have used the fractional sodium current at 1 second as an index of the degree of recovery of channels in lidocaine or GX. Sodium

**Figure 1.** Plots of use-dependent block in lidocaine and glycylxylidide (GX). Panel A: Development of use-dependent block during wash in of 37 μM lidocaine. Cell membrane potential was held at −120 mV between pulses and 8-Hz pulse trains to −10 mV were delivered every 20 seconds during wash in of lidocaine. Symbols indicate the magnitude of peak inward sodium current during the first (circles) and 14th pulse (squares) of a train normalized to the first pulse of a control train. Insets, sodium current transients measured during a train before (a) adding lidocaine and during (b) steady-state drug effect. In the lidocaine inset (b), sodium current during the first, second, and then alternate pulses (1, 2, 4, 6...14) are shown for clarity. Calibration bars in inset are 5 msec and 2,000 pA. Panel B: Development of use-dependent block during wash in and recovery after washout of 47 μM GX. GX was added to the bath perfusate at zero time. Pulse trains were identical to those in Figure 1A. Insets, sodium current recorded before (c) and during (d) steady-state GX block. GX inset shows only the first, second, and then alternate pulses for clarity. Panel C: Use-dependent block in lidocaine or GX. A train of 14 pulses was applied from a holding potential of −120 mV. Test-pulse duration was 5 msec, and the interpulse interval was 115 msec. Under these conditions, 47 μM GX produced the same amount of block as 37 μM lidocaine, approximately 50% block of sodium channels. Block developed e-fold in five pulses for either drug; therefore, the block was 95% developed in 15 pulses.
Table 1. Recovery of Sodium Channels From Block by Lidocaine (37 μM) or Glycylxylidide (47 μM)*

<table>
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<tr>
<th>V_recover</th>
<th>n</th>
<th>I/I_max† (1 sec)</th>
<th>I/I_max (steady state)</th>
</tr>
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<tr>
<td>Lidocaine</td>
<td>4</td>
<td>0.18 ± 0.07</td>
<td>0.27 ± 0.09</td>
</tr>
<tr>
<td>Glycylxylidide</td>
<td>4</td>
<td>0.44 ± 0.06</td>
<td>0.52 ± 0.05</td>
</tr>
<tr>
<td>-90 mV</td>
<td></td>
<td></td>
<td>p&lt;0.05</td>
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<tr>
<td>Lidocaine</td>
<td>5</td>
<td>0.91 ± 0.02</td>
<td>1.0</td>
</tr>
<tr>
<td>Glycylxylidide</td>
<td>4</td>
<td>0.85 ± 0.02</td>
<td>1.0</td>
</tr>
<tr>
<td>-140 mV</td>
<td></td>
<td></td>
<td>p = 0.047</td>
</tr>
</tbody>
</table>

Values are mean ± SEM.

*Recovery in either drug was significantly (p<0.05) slower than in control.
†Relative to maximum steady-state recovery at -140 mV.
 p, probability of observing this difference in means by chance.

Current measured at 1 second was normalized by the sodium current measured after clamping the membrane potential to -140 mV for 8 seconds. After 1 second of recovery, the reduction of sodium current was due entirely to drug-blocked channels that had not recovered in 1 second (channels that were not initially blocked had recovered from inactivation). Therefore, the reduction of the relative sodium current at 1 second reflects the block at that time. Faster unblocking rates will increase the sodium current measured at 1 second. In addition, 1 second was considered a clinically relevant recovery interval. At a membrane potential of -140 mV, there was generally less sodium current in GX (85%) than in lidocaine (91%) after 1 second of recovery from a train (Table 1). This suggests that at -140 mV, GX-blocked channels recover more slowly than lidocaine-blocked channels. On the other hand, at -90 mV, current recovered much faster and more completely after a train in GX than in lidocaine (Table 1). Indeed, the steady-state fraction of current recovered in lidocaine was approximately one half (0.27 vs. 0.52) that observed in GX. Furthermore, after 1 second, sodium current reached 85% of steady state (0.44 vs. 0.52) in GX but only 67% (0.18 vs. 0.27) in lidocaine. Thus, the recovery rate is faster in GX than in lidocaine at potentials near -100 to -80 mV, and recovery in GX becomes slower than in lidocaine at more negative potentials.

Voltage Dependence of Sodium Channel Availability in Glycylxylidide or Lidocaine

The results presented have shown that GX and lidocaine block sodium channels in a use-dependent manner to a similar extent under the conditions we have used. Use-dependent block could result because the drug-associated channels accumulate in an inactivated pool. Thus, as shown for lidocaine block of cardiac sodium channels by Bean et al.20 a negative voltage shift is expected in the sodium channel availability curve if the drug promotes inactivation. Figure 3A shows the voltage dependence of sodium channel availability measured dur-

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**Figure 2.** Plots of recovery of sodium current. Panel A: Recovery of sodium current from a train. Fifty 5-msec pulses from -100 to +5 mV were delivered at 10 Hz. Immediately after the train, the membrane potential was changed to one of the recovery potentials indicated next to each curve (-140 to -90 mV), and recovery was allowed for variable times as indicated on the x axis. Process was repeated for each time point shown and for each test potential. Test pulse was to -10 mV for 10 msec. Symbols indicate the peak inward sodium current normalized by the maximum current at -10 mV obtained after clamping to -140 mV for 8 seconds. Panel B: Recovery of sodium current after a train recorded in 37 μM lidocaine. Symbols represent normalized sodium current at various times of recovery at the potentials indicated by each curve. Details are identical to those described in Figure 2A. Panel C: Recovery of sodium current from a train recorded in 47 μM glycylxylidide (GX). Details are identical to those described in Figures 2A and 2B.
multiple depolarizations. However, in all cases, lidocaine shifted the sodium channel availability curve. These results are representative of 20 experiments (nine with GX and 11 with lidocaine) and show that lidocaine blocks more channels as the membrane is depolarized, whereas GX blocked a fixed fraction of channels at each membrane potential tested.

**Interactions Between Lidocaine and Glycylxylidide**

If there is a common receptor for GX and lidocaine, then the two drugs should compete for occupancy. Clarkson and Hondeghem have shown that whenever a compound with faster unblocking kinetics occupies a receptor site instead of a slower compound, under appropriate conditions, one can expect a reduction of block even though more blocker is added. Conversely, addition of an agent with slower recovery kinetics to one with faster kinetics should always increase the total block. Thus, a mixture of lidocaine and GX could give a total block that is less than the block in GX alone if the cell is hyperpolarized (−120 to −140 mV), but the total block should be greater than in GX when the cell is depolarized (−80 to −100 mV). However, if total block in both drugs is compared with block by lidocaine alone, then block will be enhanced at hyperpolarized potentials but may be relieved at depolarized potentials.

Figure 4 confirms these predictions. In the left upper panel, 94 μM GX alone is compared with block in 94 μM GX with 37 μM lidocaine at a holding potential of −90 mV. In this case, as predicted, there was more block when lidocaine was added to GX. However, when the measurement was repeated from a negative holding potential (−140 mV) in the same cell, there was less block in the combination of drugs than in GX alone. The converse experiment (adding GX to lidocaine) is shown in the lower two panels of Figure 4 (Panels C and D). On the left, block in 37 μM lidocaine is compared with the block in 37 μM lidocaine and 100 μM GX when the holding potential was −100 mV. Under these conditions, lidocaine block was relieved by GX; there was more current after addition of GX than in lidocaine alone. Finally, addition of GX to lidocaine always increased the block (see Figure 4D) at a more negative holding potential (−120 mV).

In five of 16 experiments, we observed a decrease in block when GX was added to lidocaine at holding potentials between −90 and −100 mV. In seven additional experiments, we observed no increase in block upon addition of GX. Thus, in 12 of 16 experiments, addition of a normally blocking concentration of GX did not increase the amount of block observed. In nine experiments at hyperpolarized potentials (−120 to −140 mV) in which we added lidocaine to GX, we observed a decrease in block in four experiments and no additional block in three experiments upon addition of more blocker.
Discussion

Our results demonstrate that block of cardiac sodium channels by GX was use dependent. Recovery from GX block was dependent on membrane potential. At hyperpolarized potentials (−120 to −140 mV), GX-blocked channels recovered more slowly than channels blocked by lidocaine. At potentials between −100 and −80 mV, unblocking was faster and more complete in GX than in lidocaine. These kinetic differences between lidocaine and GX were used to determine whether they compete for a common receptor to block sodium channels.

At hyperpolarized membrane potentials (−120 to −140 mV), addition of GX to lidocaine always increased the amount of block (Figure 4D), but addition of lidocaine to GX, under appropriate conditions, resulted in a reduction of block compared with GX alone (Figure 4B). At potentials (−90 to −100 mV) in which lidocaine had slower unblocking kinetics than GX, addition of lidocaine to GX resulted in more block (Figure 4A), but addition of GX to lidocaine could decrease the total block compared with lidocaine alone (Figure 4C).

Experimental Conditions and Clinical Relevance

These experiments were necessarily carried out with a reduced transmembrane sodium gradient and at a reduced temperature; therefore, it is relevant to ask to what extent these conditions affected interpretations of these results. Essentially, all available data on voltage-clamped sodium channels have been obtained at reduced temperature.7,12,17,18,20,21 Our data are no exception (see Brown et al.17,18 for details and Grant et al.22 for a review), and it is not possible to make these measurements at 37°C with presently available technologies. These conditions appeared to have three major effects on sodium current: gating kinetics were slowed; there was a shift toward more negative potentials of voltage-dependent kinetic variables; and sodium conductance was reduced. As a consequence, the temporal evolution of block and recovery from block was slowed; the membrane potential dependence of sodium channel availability was shifted from its normal position (−90 to −50 mV) to a more negative potential range (−110 to −70 mV); and the sodium current amplitude was reduced. From presently available data, it appears that the effects were merely quantitative; that is, there were no qualitative changes in the behavior of the channels. Quantitatively, the sodium channels behaved as if they had experienced a membrane potential that was about 20 mV more depolarized than normal. Thus, −90 mV in our experiments would be equivalent to −70 mV in vivo; otherwise, the channels appear to behave normally. In most cases in which experiments were carefully controlled, the results from \( V_{\text{max}} \) (maximum upstroke velocity of the action potential; an indirect estimate of sodium current) at 37°C and 145 mM [Na\(_\text{a}\)] agree with data obtained by...
Measurements in nerve have suggested that reduction of [Na] enhances the potency of local anesthetics; however, our estimates of lidocaine potency agree with those obtained in higher [Na]. Similarly, changes in [Na] did not alter the block of cardiac sodium channels by lidocaine in the experiments of Bean et al. Furthermore, we have observed that similar levels of block can be induced by the same concentrations of GX when tested in small neonatal ventricular myocytes with higher (65–110 mM) concentrations of sodium (P.B. Bennett, R.L. Woosley, and L.M. Hondeg, unpublished observations).

The standard concentrations of lidocaine (37 μM or 10 μg/ml) and GX (47 μM or 10 μg/ml) used in the present study are slightly higher than the clinical range (2–6 μg/ml). We selected these concentrations on a theoretical basis because competitive interactions are usually most demonstrative near half-maximal blocking concentrations (Figure 1). It is not known at which membrane potentials and heart rates clinical concentrations of lidocaine and GX produce 50% block in humans. However, because lidocaine has a low affinity for rested sodium channels (mM range), but a high affinity (approximately 1 μM) for inactivated sodium channels, the amount of block by a given therapeautic concentration of lidocaine will depend upon membrane potential and heart rate. We can expect that the block will be minimal in healthy tissue and may approach 100% in depolarized tissue. Intermediate levels of block will occur in cells with membrane potentials between these extremes. Thus, maximum competitive interactions as described in the present experiments are most likely to occur in tissues that are partially depressed and that are most likely to be arrhythmogenic.

Thus, we believe that reduction of sodium and temperature and increased drug concentrations caused only quantitative changes in our observations, but the qualitative interpretations remain valid. However, extrapolations to arrhythmias in humans must be made with caution; whether GX can competitively displace lidocaine in patients will require verification.

**Kinetics of Sodium Channel Recovery Explain Drug Interactions**

Although these results are consistent with the simple interpretation that GX is “slow” and lidocaine is relatively “fast” at negative potentials and that GX is fast and lidocaine is relatively slow near the resting potential, we have observed that the recovery rate in GX is not a simple linear function of membrane potential. We have not attempted to define these kinetic properties in detail; however, we wish to stress that the interactions between GX and lidocaine appear to be complicated functions of membrane potential, time, and concentration. The slowing of recovery in GX with membrane hyperpolarization may be explained in part by the low lipid solubility of GX (log Poctanol/water = 0.4). Such a drug is expected to access sodium channels through a hydrophilic pathway, and it may become trapped during hyperpolarization, thus slowing recovery from block. In contrast, lidocaine (log Poctanol/water = 2.39) can leave the channel through either a hydrophilic or hydrophobic pathway and does not become trapped with membrane hyperpolarization. Because GX appeared slower than lidocaine at hyperpolarized potentials, but frequently was faster than lidocaine at depolarized potentials, there must exist a crossover point (approximately −110 mV) where the rate of unblocking for both drugs is similar. Near this crossover point, substitution of one drug for the other will make little difference, and hence, adding one drug to another will always increase the block observed. Only at potentials more positive than the crossover point, at which GX is significantly faster than lidocaine, can addition of GX to lidocaine result in reduction of block. The latter result can only occur, however, when GX displaces a significant fraction of the channels initially occupied by lidocaine, and this result will, thus, also depend on cycle length, relative concentrations of the two compounds, and membrane potential. Because the crossover point does not always occur exactly at −110 mV, this may provide a reason why displacement was not observed in all experiments.

In the general case of two drugs that compete for a single receptor and have unblocking rates that depend on membrane potential, if the rate of recovery for drug A is equal to or slower than the recovery rate for drug B then addition of drug A to drug B must increase the amount of block. Conversely, if rate of recovery for drug A is greater than that for drug B and if drug A displaced a significant fraction of the sites occupied initially by drug B, then there may be, depending on the cycle length, a decrease in block in the mixture of drugs A and B compared with the block in drug B alone. This again will depend on the relative differences in the recovery rates, the cycle length, and the fraction of sites that became occupied by the faster drug to the exclusion of the slower drug (and hence the relative concentrations). It is, therefore, not surprising that a net displacement of block was not seen at all concentrations, rates, and potentials. The important result is that in 12 of 16 experiments, addition of GX to lidocaine either increased the sodium current or did not reduce it, whereas a reduction of sodium current in all cases would occur if no competitive displacement existed. Similar observations have been made in studies of benzocaine and lidocaine block of sodium channels in node of Ranvier, and similar synergistic and antagonistic interactions of antiarrhythmic drugs have been described in detail in terms of the modulated receptor hypothesis. However, this is the first time that both types of interaction have been demon-
demonstrated for a single pair of drugs by modulation of membrane potential. These findings support the hypothesis that GX and lidocaine compete for a common receptor site.

To our knowledge, this is the first demonstration that a metabolite of a sodium channel blocker can competitively displace its parent compound. Thus, under appropriate conditions, accumulation of the metabolite could actually prevent the blocking effects of the parent drug. Although our study was prompted by clinical observations, it is premature to suggest that these mechanisms can completely explain the clinical finding that accumulation of GX is accompanied by a loss of effectiveness of lidocaine. We have demonstrated that these effects can and do occur under certain conditions. However, these principles must now be tested more fully in a clinical setting. More importantly, we believe that competitive displacement of the parent compound by its metabolite is a general principle that must be considered when examining antiarrhythmic agents.

Acknowledgments

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