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Molecular Mechanism of Action of Local Anesthetics: A Review

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Abstract: Strichartz and Richie have suggested that the mechanism of sodium conductance block of local anesthetics involves their interaction with a specific binding site within the sodium channel. However, there is evidence that local anesthetics can interact electrostatically with membrane proteins as well as membrane lipids. Whether or not all actions of local anesthetics are mediated by common site remains unclear. Thus, it can not be ruled out that local anesthetics concurrently interact with neuronal membrane lipids since sodium channels were found to be tightly associated with membrane lipids through covalent or noncovalent bonds. In summary, it is strongly postulated that local anesthetics, in addition to their direct interaction with sodium channels, concurrently interact with membrane lipids, fluidize the membrane, and thus induce conformational changes of sodium channels, which are known to be tightly associated with membrane lipids.

Local anesthetics are drugs that block nerve conduction when applied locally to nerve tissue in appropriate concentrations. Despite the continuous clinical use of local anesthesia since the phenomenon was first described by Kôller and Freud[1] more than a century ago, molecular mechanism of action of local anesthetics on nerve has been studied only within the past 25 years.

It is now widely accepted that local anesthetics act predominantly by an interaction with nerve membranes. Local anesthetics block nerve conduction by decreasing or preventing the large transient increase in the permeability of the cell membrane to sodium ions that produced by slight depolarization of the membrane.[2] Theories for molecular mechanism of this block of sodium conductance include the specific receptor theory,[3,4] the protein perturbation theory,[5] the membrane expansion theory,[6,7] and the annular transition model.[8] Unfortunately, none of them could satisfactorily explain the local anesthetic inhibition or prevention of sodium conductance.

Opinions have been divided as to whether local anesthetics interfered with nerve membrane sodium channel function by directly binding to the proteins, or whether the main modes of action occurred indirectly through a change in the physicochemical properties of the lipid membranes into which the local anesthetics readily dissolved.[9] Since biological membranes are of highly complex composition, it has not been feasible to monitor changes in the local lipid environment and to determine its effect on membrane protein function at the same time.

In this review, we first discuss the biochemistry of the sodium channel. Four key questions address the electrophysiology and chemistry of local anesthesia: 1) Which microscopic events regulate the ion permeability changes that underlie the nerve impulse? 2) What is the structure of the sodium channel? 3) Where and how do local anesthetics bind to the sodium channel? 4) What are the fundamental microphysiological actions of local anesthetics?

Structure of the Sodium Channel

Is there any direct evidence to place the local anesthetic receptor site in the intracellular end of the transmembrane pore of the sodium channel? Probably the experiment that comes closest to direct evidence on this point is examination of the effects of extracellular permeant cations on inhibition of sodium channels in squid giant axon.[10] Inward movement of permeant cations through the activated channel appears to oppose the block of open channels by quarternary local anesthetic derivatives. This effect is blocked by tetrodotoxin since it prevents ion movement through the channel. Thus, this effect of permeant cations is most easily interpreted in terms of competition with the charged form of local anesthetics for an anionic binding site in the sodium channel. Additional structural information seems necessary, however, before a defined conclusion on the location of local anesthetic receptor sites can be made.

The amino acid sequence of the sodium channel found in eel electroplax has been deduced from its gene sequence.[11] The channel has large hydrophobic regions, probably in α-helical conformations that span the membrane, interspersed with hydrophilic regions that presumably either "line" the Na^+-conducting "pore" of the channel or the aqueous, polar interfaces of the membrane.

Sodium channels possess, in addition to one major glycoprotein with molecular weight of roughly 200,000, differing numbers of other subunits, depending on the species and tissue of origin. Channels isolated from the eel's electroplax organ include only a single large glycoprotein[12] those from rabbit muscle have an additional smaller subunit[13, 14] and those from rat brain have two additional smaller subunits, each having a mass of about 40,000d.[15] The electroplax sodium channel protein is densely glycosylated and contains an unusually large fraction of acidic groups, totally about 100 negative charges per channel.[16] It is assumed that the sodium channel, in common with other large integral membrane proteins, is oriented with its glycosylated groups on the outside surface of the cellular membrane.[17]

Manipulations of Drug Structure and Conditions

A variety of protein and nonprotein toxins modify the physiology of sodium channel.[18] At present six different binding sites for toxins have been postulated. These include extracellular surface sites for tetrodotoxin/saxitoxin and for two different classes of peptide toxins(α and β, usually isolated from scorpion venom), intramembranous sites for two classes of lipophilic organic molecules (brevotoxin/ciguatoxin), and the site(s) of local anesthetic action. Each of these sites appears to be linked to at least one other site, accounting for conformationally coupled interactions among drug classes that often are dependent on the membrane potential.

There is no direct biochemical evidence identifying the location of any local anesthetics binding site on sodium channels. Instead, physiologic and biochemical data provide indirect clues about the site. The relevant experiments take three forms; studies on local anesthetics with different structures and under different conditions, examinations of local anesthetic inhibition of chemically modified sodium channels, and studies of interactions between local anesthetics and other drugs. By integrating these findings, we can deduce many properties of the routes to and properties of a local anesthetic site on the sodium channel.

The fluorescent probe technique was used to examine the binding of spin-labeled local anesthetics to lipid model systems, to the membranes of human red blood cells, and to rabbit sarcoplasmic reticulum.[19] Local anesthetics, once intercalated into the bilayer, may diffuse laterally and interact with membrane components, lipids as well as proteins.[19] The binding properties of spin-labeled anesthetic to human erythrocyte membranes and to vesicles made from human erythrocyte lipids were studied.[20] Wang *et al*. conclude that membrane-bound tertiary amine

anesthetics in cationic form do interact selectively with phospholipids and proteins.[20] In addition to sodium channels, local anesthetics can also bind to other membrane-bound proteins.[21]

Based on the chemical structures known for local anesthetics and implied for sodium channels, there are several loci on the channel where local anesthetics are likely to bind. These possibilities are compounded further by the ionizable character of most local anesthetics because channels might be inhibited by either the protonated or the neutral form of local anesthetics. The problem has been approached by two strategies: altering local anesthetics structure to produce permanently neutral or permanently charged molecules (permanently charged molecules having limited permeation of membranes), and controlling the pH to set the level of ionization of tertiary amine local anesthetics. Manipulation of external pH affects both tonic and phasic inhibition. Tertiary amine local anesthetics act to inhibit sodium channels of single isolated axons much faster in alkaline solution than when the same drug is applied at neutral pH. In contrast, the rate of onset of inhibition by permanently charged (slow) or permanently neutral (fast) local anesthetics is independent of external pH. Thus, the major effect of pH is on local anesthetics ionization, not on the channel protein. Most investigators believe that these results show that local anesthetics molecules must pass into and/or through the nerve membrane to reach their site of action, and that the neutral species penetrates much faster than the protonated one.

By controlling pH inside or outside single squid giant axons during the application of tertiary amine local anesthetics, Narahashi *et al.* showed that the protonated (charged) form in the axoplasm was the most potent species.[22] This finding is consistent with the relatively weak sodium channel blockade produced by a permanently neutral lidocaine homologue, 5-HHX, which is 10-20 times less potent than lidocaine at pH 7.3.[23] However, quantitative potency ratios for charged and neutral species depend on the particular local anesthetics. For example, at alkaline external pH(8−9), procaine is five to seven times more potent tonically as it is at neutral pH,[24] but four to five times more potent than its neutral homologue, benzocaine.[25-28] Curiously, Richie *et al.* found that impulse conduction in nerves pretreated with dibucaine, a highly lipophilic local anesthetic, could be either blocked or relieved by setting the pH of anesthetic-free bathing solutions at neutral (pH 7) or alkaline (pH 9) values, respectively.[28, 29] As will be shown below, pH has effects on the distribution of local anesthetics between aqueous phases and the membrane as well as on the charge of membrane-associated local anesthetics. More hydrophilic anesthetics will be drawn out of the membrane by acid pH and their potency thereby reduced. In contrast to the findings of Narahashi *et al.*,[22] with dibucaine the extracellular pH regulates block by drug molecules already within the nerve. This is an example where more hydrophobic local anesthetics, which tend to stay within the nerve membrane, may be potentiated by aqueous acidification.

For most local anesthetics, lower cytoplasmic pH should favor protonated over neutral tertiary amine local anesthetics, and should thus increase the apparent local anesthetic potency. This effect was observed for tonic inhibition when the axoplasmic compartment of the squid giant axon was acidified during extracellular local anesthetics application.[22] In contrast, phasic block by local anesthetics applied externally to frog muscle was not potentiated when cytoplasmic pH was lowered.[30] Protons by themselves can block sodium channels[31] as well as alter their gating, and the effects of altered pH on local anesthetic action may be complicated by the separate actions of the local anesthetics on the channel.[24, 26, 30] The effect of internal pH on the different interactions between local anesthetics and the sodium channel is unclear.

Cationic local anesthetics derivatives are potent sodium channel inhibitors, much like protonated tertiary amine local anesthetics. These small quaternary ammonium local anesthetics, permanently charged due to an additional alkyl substituent on their terminal amine nitro-

gen, are relatively lipid insoluble and membrane impermeant. When applied outside the cell membrane, small quaternary local anesthetics do not block sodium currents. However, when applied on the cytoplasmic side of a membrane, quaternary local anesthetics strongly block sodium currents.[32, 33] These results have suggested an "internal binding site" for charged local anesthetics accessible via a hydrophilic pathway from the cytoplasm.

Chemically modified procaine and tetracaine block neuronal action potentials upon external application, even when covalently bound to biotin at the local anesthetic's aromatic region. But when these local anesthetics derivatives are bound by the large biotin-binding protein, avidin (molecular weight 60,000d), they do not block impulses when applied externally even though the unmodified tertiary-amine portion of the conjugated drug can, in theory, still penetrate 10-15Å into the membrane.[34] In light of the relative potency of externally versus internally applied quaternary local anesthetics, it appears that local anesthetics permeation and distribution in the membrane, and not the chemical reactivity of the terminal amine nitrogen, are the factors controlling local anesthetic action.

Phasic block with internal quaternary local anesthetics is profound, even at low depolarization frequencies, and appears to require an activated conformation of the sodium channel.[33, 35, 36] In contrast, the neutral local anesthetics benzocaine produces an insignificant phasic inhibition of sodium currents[26, 37, 38] and the neutral lidocaine homologue, 5-HHX, produces a weak phasic block.[23] Ionizable, tertiary amine local anesthetics (e. g., lidocaine) also produce phasic inhibition that is maximized by neutral or mildly acidic external solutions (pH 6.5-7) but is minimized by alkaline external solutions (pH 9).[26, 30, 39] External pH has only small effects on the kinetics of phasic inhibition by nonionizable local anesthetics, showing that the primary role of protons is on the drug itself and not on the channel.

As mentioned previously, phasic inhibition may result from differential local anesthetics binding affinities among various channel conformations (the "modulated receptor") or from differential access to and from a binding site of constant local anesthetics affinity (the "guarded receptor"). Either local anesthetics access or affinity might be modulated by the channel's conformation and by the ionization of the local anesthetics. Examination of phasic block by tertiary amine local anesthetics during channel activation as well as recovery from phasic block between pulses has revealed some surprising results. The on-rate for binding of tertiary amine local anesthetics to the activated channel that occurs during brief depolarizations, increased with alkaline pH, consistent with a much faster binding by the neutral species than by the protonated form.[23] The pH-dependence of the on-rate for binding was quite similar to that for the fraction of the nonionized species of local anesthetics. The rate of local anesthetics dissociation from the activated channel was independent of external pH, as if extracellular protons could not change the ionization of drug bound to activated channels. In contrast, the dissociation of tertiary amine local anesthetics from closed sodium channels in repolarized membranes depended strongly on external pH, being 10 to 50 times slower in mildly acidic (pH 6.2) than in alkaline (pH 9-10) solutions. The dependence on external pH of this off-rate was described by a local anesthetics ionization with pKa 0.3-0.4 pH units higher than that measured in aqueous solution.

A physical model for phasic inhibition by tertiary amine local anesthetics emerges from these studies. Local anesthetics dissolved in the membrane or in the cytoplasm bind to a site on the activated channel; membrane-dissolved drug approaches the binding site through a "hydrophobic" pathway whereas local anesthetics in the cytoplasm has access to the site via a "hydrophilic" pathway.[32] Either protonated and neutral species of local anesthetics may dissociate equally rapidly from the activated channel or external protons cannot reach the activated local anesthetics binding site. Conversely, when the channel closes, extracellular protons can reach the bound local anesthe-

tics: dissociation of protonated local anesthetics from the closed channel is slow but finite and a charged local anesthetics can dissociate without first losing its proton. Whether this dissociation follows the same "hydrophobic" pathway as that of the neutral species remains to be shown. At low external pH, tertiary amine local anesthetics behave more like their charged homologues, whereas at high pH they behave like the uncharged compounds.

Charge is not the only factor influencing the kinetics of phasic local anesthetics binding. Recent results reveal marked phasic inhibition by a neutral lidocaine homologue in which an hydroxyl group replaces the tertiary amine.[23] The rate constant for dissociation of this drug from closed channels, roughly 1 s, lies between the estimated values for the protonated and neutral forms of lidocaine (0.1 s and 7 s, respectively), demonstrating that even uncharged local anesthetics can dwell for relatively long times on the binding site. The slow dissociation rate may result from larger size, lower lipophilicity (limiting hydrophobic escape), or hydrogen bonding of this local anesthetic to the phasically activated channel.

Pathways to the Local Anesthetics Binding Site

Indirect evidence implicates the channel's ion-conducting pore as the hydrophilic pathway. For example, sodium flow through the channel alters local anesthetics action. Phasic block by quaternary drugs is enhanced when impermeant cations are substituted for external sodium.[35, 40] Similarly, application of tetrodotoxin (which occludes the channel on its external surface and prevents entry of sodium ions) to an axon simultaneously exposed to internal quaternary local anesthetics reduces the gating current measured with infrequent stimulation to its most phasically reduced level.[41] In both of these studies, reducing the sodium flux potentiates local anesthetic actions, implying that influx of external sodium ions inhibits local anesthetics binding to its receptor. Direct, competitive antagonism would require either that the local anesthetics receptor lie within an aqueous region of the sodium channel (where it could be reached easily by sodium ions), or that there be allosteric antagonism between sodium and local anesthetics two distinct by interacting sites.

The hydrophobic pathway is difficult to define. "Hydrophobicity" is usually defined by comparing the relative concentration of a substance in a hydrophobic solvent with its concentration in an immiscible aqueous phase at steady-state. Hydrophobicity may be quantified by the partition coefficients for each of separate forms of that substance.

Where are local anesthetics absorbed in membranes ? Membranes composed of phospholipids have at least three separate regions: 1) a charged or zwitterionic (i. e., containing both positive and negative charges) interface with the aqueous solvent: 2) a region of high-dipole intensity (near the ester bonds that join fatty acyl groups to the glycerol or ceramide moieties); and 3) an apolar core containing only the acyl hydrocarbon tails. Local anesthetics interact with the lipid molecules differently in each of these different zones, none of which behaves identically to octanol. From studies of local anesthetics and lipid behavior in model membranes during local anesthetics binding, it appears that local anesthetics are bound primarily near the membrane interface.[42, 43] Protonated local anesthetics extend further towards the polar head groups of phospholipids while the unprotonated species dwells a bit deeper in the membrane.[44] The drug molecules shuttle relatively rapidly between deeper and more superficial sites; much of the binding energy arises from hydrophobic interactions,[45] some from stabilization of the local anesthetic's dipole (ester or amide bond) in the membrane's dipole field, and some from interactions of tertiary amines with polar regions of the lipids.[42]

The procaine is predominantly distributed on the surface area, while tetracaine has a greater accessibility to the hydrocarbon interior of the synaptosomal plasma membrane vesicles isolated from bovine cerebral cortex.[2]

Ionization of local anesthetics will also be altered by membrane adsorption. Double layers form boundaries of electrical potential extending 10-20 Å from the membrane surface. Negative charges on the membrane surface will concentrate protons from the bulk solution in the "double layer" adjacent to the membrane.[45] This localized acidity may raise the apparent pKa for pH can only be measured in the bulk solution. The opposite effect will result from immersing the tertiary local anesthetic's amine group in a medium with a dielectric constant lower than that of water (i. e., a medium in which the charged local anesthetics is less soluble); the protonated species will be destabilized relative to the neutral species, leading to a drop in the apparent pKa.[46] Both local anesthetics adsorption and changes in ioniztion will be altered by the uptake of large amounts of local anesthetics. The complex overall effects may depend on the particular local anesthetics and on the membrane in question.

Experiments measuring rates of sodium channel inhibition by local anesthetics applied at different pH cannot discriminate the relative importance of the hydrophilic versus hydrophobic pathways for phasic inhibition. The faster onset of tonic block by tertiary amine local anesthetics at more alkaline pH could result from higher concentrations of either neutral or protonated drug species in both the membrane and cytoplasm.[26, 27, 47] Neutral drug molecules diffuse so rapidly through membranes that their concentration differs insignificantly from one side to the other, being only a small fraction of the concentration gradient across the internal and external "unstirred layers". The unstirred layer is an immoveable slab of solution adjacent to the membrane surface.[45, 48] Estimates of the thickness of unstirred layers range up to 10^2 μm, whereas the membrane is only 10^{-2} μm thick. For protonated local anesthetics, the membrane is the major diffusion barrier, but local anesthetics distribution in the unstirred layers will depend on buffer strength and the relative mobility of the buffering species.

The relationship between hydrophobicity and rate of local anesthetic inhibition is complex. At equipotent doses, small local anesthetics with modest hydrophobicity ($P^o < 10^2$) have slower rates of inhibition than larger local anesthetics with intermediate hydrophobicity ($10^2 < P^o < 10^3$). But local anesthetic molecules that are very hydrophobic ($P^o > 10^4$) do not inhibit more rapidly; rather their rate of tonic block and their dissociation from closed channels, both assumed to depend on "hydrophobic" partitioning, are slower than those of intermediately hydrophobic local anesthetics.

There are three explanations for this. The first, proposed by Courtney, is that molecular size as well as hydrophobicity is a factor in local anesthetic kinetics and that values of octanol : buffer partition coefficients must be modified by a molecular weight correction to account for the slow actions of hydrophobic, relatively large local anesthetics.[49]

The second explanation is that membranes are heterogeneous compartments. Even if octanol-based hydrophobicity accurately models the membrane's hydrocarbon core, it cannot acount for local anesthetics adsorption at the dipolar region or near the phospholipid head groups. Such adsorption will not only concentrate amphiphilic drugs at certain intramembranous zones, but will also orient these molecules, restricting their motion and diffusion within and across the membrane.[50]

The third explanation questions whether one may use equilibrium measurements to make kinetic predictions. The partition coefficient expresses the relative distribution of a drug between two phases at equilibrium, a value that is the ratio of rate constants for adsorption and desorption but is otherwise unrelated to the absolute rates. One would conclude from the second and third explanations that until we know how fast local anesthetics move into and out of the various phases of a membrane we cannot correctly attribute physiologic effects to a particular pathway. But those dynamic parameters are not available, and present efforts at modelling local anesthetic mechanisms must rely on simple physicochemical proper-

ties, even though any correlation may be merely fortuitous.

Properties of Local Anesthetics Binding Sites

The local anesthetics binding on the sodium channel remains undefined. It is possible that local anesthetics binding to any one of several sites may inhibit sodium currents. In this section, we will summarize the relationship between the physicochemical properties of local anesthetics and their pharmocologic actions, as these determine the general characteristics of a putative binding site. Courtney has studied sodium channels in a variety of tissues using structurally diverse agents that vary in their aromatic residues, hydrocarbon, or amine regions.[49] He found correlations between local anesthetics hydrophobicity tonic potency and the local anesthetics dissociation rate from closed channels, but features of the local anesthetics other than hydrophobicity also varied, including pKa, size, and the region of the local anesthetics molecule that contained the altered hydrophobicity.[51]

Physicochemical qualities may also be addressed by examining a homologous series of drugs or by comparing the kinetics of quaternary compounds in which differences in hydrophobicity exist independent of effects on ionization. The more hydrophobic local anesthetics in a homologous series are more potent sodium channel inhibitors. Both tonic and phasic inhibition increase with hydrophobicity, although these two modes of action are characterized by different kinetic parameters. Tonic inhibition reflects the equilibrium local anesthetics occupancy of binding sites. Phasic inhibition, an intrinsically transient process, depends on the interplay between increased binding of local anesthetics during depolarizing pulses and dissociation from the site between pulses. Structural changes that alter hydrophobicity inevitably produce changes in pKa, but when these are factored into an estimation of the degree of ionization, the important interrelationship of hydrophobicity and potency emerges. For a series of lidocaine homologues, both tonic impulse inhibition and the steady-state phasic inhibition have potencies proportional to the calculated octanol : buffer distribution coefficient.[52] The potencies of the local anesthetics in these series were thus proportional to their relative tendencies to distribute into a hydrophobic medium. Similar findings were reported by Hille[26] for a more diverse collection of local anesthetics inhibiting sodium currents tonically over a range of pH values.

One interpretation of the dependence of local anesthetic potency on hydrophobicity views the membrane concentration of a local anesthetic as proportional to its hydrophobicity. If this "membrane concentrating" hypothesis were true, then the effective concentration of free local anesthetics in the membrane would be equal for all local anesthetics at equipotent doses (identical to the Meyer-Overton hypothesis for general anesthesia) and the rates of onset of inhibition would be comparable despite large differences in local anesthetics in solution. Another interpretation is that local anesthetics in the bulk membrane do not directly equilibrate with the binding site, but that a hydrophobic component of the total energy for binding the local anesthetics to its site accounts for the strong correlation of blocking potency with hydrophobicity. Accordingly, the dissociation rates of local anesthetics from the site should be proportionately slower for the more potent compounds.

Chernoff examined these possibilities using a series of lidocaine homologues with potencies and hydrophobicities ranging over two orders of magnitude. Despite the large range of potencies for phasic and tonic inhibition, the rates of binding to phasically activated channels differed minimally. This was supported with the hypothesis that the membrane was concentrating "free" local anesthetics near the phasic local anesthetics binding site. Dissociation rates from the phasically activated channel also differed by a small amount, threefold, which is too small to be consistent with the hypothesis that hydrophobic

binding to the active site accounts for observed potency difference.

Courtney has measured dissociation rates from closed channels of a more widely varying sample of local anesthetics to test the role of local anesthetics size.[51] He found that smaller local anesthetics dissociate more rapidly than larger ones and that moderate hydrophobicity aids departure of local anesthetics from the binding site, although extreme hydrophobicity (for example, etidocaine and bupivacaine) lengthens the duration of local anesthetics occupancy more than would be predicted from less hydrophobic drugs. Courtney attributed much of this rate dependence to molecular size and proposed that local anesthetics molecules must depart from closed channels through a long, narrow passage (roughly 3.6 Å in radius) where the local anesthetics molecules literally scraped the walls.

Cooling increases local anesthetics inhibition of neuronal impulses. Lidocaine potency increases about tenfold when the temperature drops from room temperature to near 10℃, and phasic impulse blockade is also potentiated. The solution pKa for lidocaine also rises slightly upon cooling, approximately halving the fraction in the neutral form in the bathing solution (from 9-4% at pH 7.2), while marginally increasing the protonated form in solution.[53] However, bulk uptake of protonated local anesthetics by bilayer membranes rises with cooling; the partition coefficient for protonated tetracaine increases from 3.1×10^3 at 25℃ to 6.3×10^3 at 4℃, whereas that of the neutral species changes by less than 20%. The overall effect is to increase the protonated local anesthetics concentration within the membrane but not in the solution. This underscores the importance of the protonated species for channel blockade. However, potencies of uncharged local anesthetics are also enhanced by cool ing,[54,55] indicating that protonation alone does not account for this increased potency. At low temperatures (4C) benzocaine develops a pronounced phasic action virtually absent at room temperature.[54] Stronger local anesthetics binding or slower diffusion of "free" drug away from the channel may explain the potentiation of phasic block by cooling.

Local Anesthetics-Membrane Lipids Interactions

It has long been predicted that biomembranes should expand in the presence of local anesthetics, on the basis that local anesthetics penetrate and expand films spread at the air/water interface.[6] Membrane expansion theory proposes that local anesthetics increase the fluidity of membrane lipids and expand the membrane.[8] Although local anesthetics have indeed shown to cause an expansion of erythrocyte membranes,[8] it is not clear how an expansion of the nerve membrane would lead to a block of sodium conductance.

The basic postulate of the annular transition model is that the sodium channel is surrounded by an annulus of lipid, which is in the crystallin, or gel state.[8]

Purified sodium channels can be dissolved in micelles of nonionic detergent stabilized with phospholipids. The unusually hydrophobic sodium channel macromolecule has an anomalously high detergent-binding capacity, due in part to more than a dozen long-chain fatty acids associated with each channel molecule. Bound to the protein by covalent or noncovalent bonds, these acyl chains may anchor and orient the channel in the membrane, stabilizing the channel's three-dimensional structure. Long-chain fatty acids also may participate in binding of lipophilic drugs such as local anesthetics.

The microviscosity of the hydrocarbon region of synaptosomal plasma membrane vesicles isolated from bovine cerebral cortex was decreased by lidocaine and procaine.[57] The differential scanning thermograms of dimyristoylphosphatidylcholine multilamellar liposomes showed that local anesthetics significantly lowered the phase transition temperature, broadened the thermogram peaks, and reduced the size of the cooperative unit.[56]

Exploiting intracellular excimer formation of 1, 3-di

(1-pyrenyl)propane, we showed that lidocaine increases the lateral diffusion of the probe in the lipid bilayer of synaptosomal plasma membrane vesicles (SPMV) and phosphatidylcholine model membranes (PC).[57, 58] Utilizing fluorescence polarization of 1,6-diphenyl-1,3,5-hexatriene, prilocaine and procaine have been proven to increase the rotational diffusion of the probe in LM fibroblast plasma membranes, SPMV, and PC bilayer membranes.[59, 60] In LM fibroblast plasma membranes and SPMV, prilocaine exhibited a greater fluidizing effect on the outer monolayer as compared to the inner monolayer.[57, 59]

Conclusion

Local anesthetics have been known to produce their specific effects through their direct interactions with sodium channels. However, it cannot be ruled out that local anesthetics concurrently interact with neuronal membrane lipids since sodium channels were found to be tightly associated with membrane lipids through covalent or noncovalent bonds. Thus, it may be premature to take sides in the controversy about whether membrane lipids or membrane proteins are the site of local anesthetic action. In summary, it is strongly postulated that local anesthetics, in addition to their direct interaction with sodium channels, concurrently interact with membrane lipids, fluidize the membrane, and thus induce conformational changes of sodium channels, which are known to be tightly associated with membrane lipids.

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초록 : 국소마취제의 분자적 약리작용기전 : 총설

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비록 국소마취제가 신경세포막 당단백질중의 하나인 sodium channel과의 직접적인 상호작용에 의하여 그 특유의 국소마취제를 나타낸다고 할지라도 신경세포막의 골격을 형성하고 있는 지질과 국소마취제와의 상호작용을 분자적 국소마취제 작용기전을 설명함에 있어 배제할 수는 없다. 왜냐하면 sodium channel은 지질과의 공유 또는 비공유결합을 통해 단단하게 결합되어 있기 때문이다. 따라서 국소마취제가 직접 sodium channel과 결합, 상호작용하여 Na^+의 유입을 차단할 뿐만 아니라 직접 지질의 측방 및 회전확산 운동을 증가시킴으로써 간접적으로 sodium channel의 conformational change가 유발되어 Na^+의 유입이 차단되는 등의 복합적인 기전에 의하여 그 특유의 국소마취작용이 나타나는 것이다.