Mechanisms of Actions of Inhaled Anesthetics

Jason A. Campagna, M.D., Ph.D., Keith W. Miller, D.Phil., and Stuart A. Forman, M.D., Ph.D.

Suffering so great as I underwent cannot be expressed in words . . . but the blank whirlwind of emotion, the horror of great darkness, and the sense of desertion by God and man, which swept through my mind, and overwhelmed my heart, I can never forget.”

Such was the experience of surgery before October 1846, when William Morton’s successful public demonstration of ether anesthesia at Massachusetts General Hospital led to its widespread acceptance by surgeons. Today, anesthesiologists employ a wide variety of drugs, some of which they use exclusively to produce general anesthesia.

This review focuses on the inhaled anesthetics (Fig. 1) in current use: nitrous oxide, halothane, enflurane, isoflurane, sevoflurane, and desflurane. Our understanding of how these drugs reversibly alter central nervous system function has changed dramatically in the past two decades.

We will summarize the evidence that inhaled anesthetics ablate movement in response to noxious stimuli (immobilization) by depressing spinal cord functions, whereas their amnesic actions are mediated within the brain. Some compounds, which are referred to as nonimmobilizers, share many chemical features of inhaled anesthetics and possess amnesic activity but do not inhibit movement in animals. These differences suggest that anesthetics induce amnesia and immobility by affecting distinct molecular targets. Our review will also describe new techniques for dynamically assessing regional brain activity, which when combined with electrophysiological and behavioral monitoring, promise to provide important insights into the ways in which anesthetics affect neural networks. Research on anesthetic-sensitive ion-channel proteins that control neuronal excitability has revealed that γ-aminobutyric acid type A (GABA$_A$) receptors are most likely involved in the actions of many general anesthetics. Even so, some inhaled anesthetics may act by inhibiting such excitatory ion channels as neuronal nictinic and glutamate receptors. The subtly different clinical actions of inhaled anesthetics are probably due to distinct, specific actions on a small number of critical molecular targets.

The identification of specific binding sites for inhaled anesthetics on certain proteins is a dramatic departure from the classic view that all general anesthetics act nonspecifically. Differential sensitivities to various anesthetic actions may have a genetic basis. Moreover, new research indicates that the effects of general anesthetics depend on multiple features of their molecular structure and that focusing on such features may further improve the clinical utility of general anesthetics.

Except for the treatment of status epilepticus, general anesthesia is always an adjunct to another procedure. Anesthetic practice has evolved in response to new procedures.
In turn, anesthesia has accelerated the development of these procedures. The number of ambulatory surgical procedures is increasing rapidly in the United States; nearly 75 percent of all surgical procedures are now performed on an outpatient basis. General anesthesia is also increasingly used for noninvasive and minimally invasive diagnostic and therapeutic techniques that require immobilization and deep sedation of the patient, as in pediatric radiology and endoscopy, interventional radiology, electroconvulsive therapy, radiation therapy, various cardiology procedures, transbronchial biopsy, and urologic procedures. In these settings, which emphasize cost effectiveness, rapid discharge, and patient satisfaction, the rapid emergence from anesthesia and minimization of side effects are especially important.

Even though volatile anesthetics can cause cardiopulmonary depression and death at concentrations near those that produce deep anesthesia, improvements in practice have reduced mortality attributable to anesthesia to an estimated 1 per 250,000 healthy patients. More common undesirable and potentially harmful effects that occur during and after general anesthesia are autonomic instability, hypothermia, cardiac dysrhythmias, nausea, vomiting, and delirium; these effects not only cause discomfort for the patient but also delay discharge and increase costs. In some cases the use of general anesthesia outside the operating setting may be the only way to accomplish the procedure. Figure 1. Classes and Generations of Inhaled Anesthetics. Within a few years after their introduction into widespread clinical use, three major classes of inhaled anesthetics were used: hydrocarbons, ethers, and other (non–carbon-based) gases. Nitrous oxide was first recognized as an analgesic in the early 19th century, but its low potency precludes its use as the sole anesthetic agent for most procedures. The hydrocarbons and diethyl ether were either highly toxic (chloroform) or explosive (cyclopropane, ethylene, and ethane). Halogenation of alkanes and ethers reduces their flammability, but fluroxene, the first such compound introduced in 1954, was later withdrawn from use because of residual combustibility. Halothane, the first noncombustible volatile halogenated alkane, entered clinical practice in 1956. Enflurane and isoflurane, both halogenated ethers, were first used clinically in 1972 and 1981, respectively. As compared with diethyl ether and halothane, these are less soluble in blood, allowing faster pulmonary uptake and elimination. The uptake and elimination of sevoflurane and desflurane, introduced in the 1990s, are even faster. Xenon, which was first recognized as an anesthetic in 1951, has highly favorable clinical features including no taste or odor, rapid pulmonary uptake and elimination, no hepatic or renal metabolism, and minimal cardiovascular depression and arrhythmogenicity. The limited supply of xenon and the expense of extracting it from the atmosphere will most likely prohibit its widespread use in the immediate future.
room may pose a greater risk to the patient than the concomitant procedure itself (e.g., magnetic resonance imaging in children). For these reasons, inhaled anesthetics that allow rapid emergence of anesthesia and have few adverse effects are highly desirable.

**WHAT IS GENERAL ANESTHESIA?**

Oliver Wendell Holmes introduced the word “anesthesia” to signify insensibility to surgical pain. However, there is still no consensus on a more objective definition of general anesthesia. At different concentrations inhaled anesthetics induce a variety of reversible, clinically important effects (Table 1 and Fig. 2). Low concentrations can induce amnesia, euphoria, analgesia, hypnosis, excitation, and hyperreflexia. Higher concentrations cause deep sedation, muscle relaxation, and diminished motor and autonomic responses to noxious stimuli, effects that progress to “surgical” anesthesia. Some volatile anesthetics also protect the myocardium against the effects of ischemia, an important component of anesthetic action for many patients.

Rigorous definitions have been introduced for investigations of the underlying mechanisms of anesthetic effects in humans and animals and for the clinical assessment of the depth of anesthesia (Table 1). These effects must be reversible and produced without the need for supplemental muscle relaxants, benzodiazepines, narcotics, or autonomic modulators. Loss of appropriate response to specific spoken commands is used to identify hypnosis (the impairment of perceptive awareness) in anesthetized subjects. In addition, patients may have perceptive awareness without recall because memory is more sensitive to anesthetics than awareness. Laboratory animals are assessed for loss of righting reflexes — the inability to return to an upright position in response to nonpainful stimuli — whereas other tests, such as behavioral conditioning, assess the effects of anesthesia on learning and memory.

Scales that assess the potency of inhaled anesthetics are based on alveolar (in practice, usually end-expiratory) anesthetic concentrations that are associated with defined behavioral end points (Table 1). The most widely used scale is the minimal alveolar concentration of anesthetic that suppresses purposeful movement in response to a standard noxious stimulus (MAC or MAC-immobility), although today the acronym MAC is more frequently used to refer to the median alveolar concentration, indicating the median value for a population under controlled conditions. Analogous potency scales define the MAC that prevents voluntary responses to spoken commands (MAC-aware) and the MAC required to blunt autonomic responses to painful stimuli (MAC-BAR).

**NONSPECIFIC PHARMACOLOGY AND LIPID THEORIES OF ANESTHETIC ACTION**

For more than a century, two concepts — the unitary hypothesis and the Meyer–Overton rule — dominated thinking about the mechanisms underlying anesthetics. By the 1870s, a wide range of structurally unrelated agents were known to possess anesthetic activity, leading Claude Bernard to postulate that all of them acted through a common mechanism. Approximately 30 years later, Meyer and Overton observed a strong correlation between the potency of anesthetics and their solubility in olive oil (Fig. 2). These two ideas led to the theory that volatile anesthetics act nonspecifically on hydrophobic lipid components of cells.

Most researchers have abandoned this theory, despite its elegance, because anesthetics cause only slight perturbations in lipids, which can be reproduced by small changes in temperature that do not alter behavior in animals. There are, moreover, a number of apparent exceptions to the Meyer–Overton correlation. These can be explained by variations in the size, rigidity, and polarity of the anesthetic and by the location of anesthetics within lipid bilayers, which differs from that of related compounds without anesthetic activity.

Interest in the possible role of lipids and lipid–protein interactions in anesthesia continues, but models for the rigorous testing of current hypotheses are lacking. Furthermore, it now appears unlikely that the different structural classes of inhaled anesthetics (Fig. 1) act through a single common mechanism.

**BEHAVIORAL PHARMACOLOGY OF INHALED ANESTHETIC ACTIONS**

Behavioral studies have revealed a number of critical exceptions to the Meyer–Overton rule and the unitary hypothesis. For instance, homologous series of anesthetics such as the n-alcohols and n-alkanes...
exhibit a steady increase in potency as successive methylene (CH₂) groups are added, up to a cutoff point at which adding another methylene eliminates anesthetic activity (the “long-chain alcohol cutoff”). Furthermore, the so-called nonimmobilizers, which are volatile halogenated alkanes with structural similarities to volatile anesthetics, are predicted by the Meyer–Overton rule to be potent anesthetics. However, they lack immobilizing activity and in some cases induce convulsions. In contrast to the historical focus on hydrophobicity alone, analyses of molecular structure and activity indicate that hydrophobicity, electrostatics, and size all contribute to the immobilizing potency of inhaled anesthetics.

Comparing pharmacology among several anesthetic actions reveals distinct relations between structure and activity. For example, the ratios of MAC-awake to MAC-immobility for nitrous oxide and diethyl ether are significantly higher than those for some halogenated volatile anesthetics. And, because volatile nonimmobilizers produce amnesia in animals, it is likely that immobilization and amnesia are mediated by separate mechanisms. Amnesia and hypnosis in humans can also be distinguished clinically, electrophysiologically, and pharmacologically.

### Glossary of Terms

<table>
<thead>
<tr>
<th>Term</th>
<th>Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>loss of righting reflexes (LORR)</td>
<td>The failure of an animal to regain upright posture when placed on its back.</td>
</tr>
<tr>
<td>median alveolar concentration for blunting autonomic responses (MAC-BAR)</td>
<td>The end-tidal concentration of inhaled anesthetic that prevents appropriate voluntary responses to spoken commands (e.g., to open one’s mouth or to raise a hand) in 50 percent of a test population.</td>
</tr>
<tr>
<td>median alveolar concentration awake (MAC-awake)</td>
<td>The end-tidal concentration of inhaled anesthetic that prevents appropriate voluntary responses to spoken commands in 50 percent of a test population.</td>
</tr>
<tr>
<td>potency</td>
<td>A measure of relative drug activity that is inversely related to the concentration required to produce a standard effect. A volatile anesthetic that produces a behavioral effect at half the concentration of another anesthetic is said to be twice as potent.</td>
</tr>
<tr>
<td>amnesia</td>
<td>The partial or complete loss of memory. Usually anterograde (affecting recall of experiences after the onset of anesthesia), amnesia may also be retrograde (affecting recall of experiences that precede the onset of anesthesia).</td>
</tr>
<tr>
<td>explicit memory</td>
<td>Information that is consciously perceived and retained and that can subsequently be reported accurately.</td>
</tr>
<tr>
<td>implicit memory</td>
<td>Information that is unconsciously perceived and retained and that cannot subsequently be reported. However, this experience affects a subject’s subsequent performance.</td>
</tr>
<tr>
<td>hypnosis</td>
<td>There are various functional definitions of this term. We use it to connote drug-induced impairment of cognitive functions required for responding appropriately to environmental stimuli, including attention and perception. For a patient in the awake state, administration of inhaled anesthetics can produce a wide range of hypnotic depths, from mild inattention to unresponsiveness to noxious stimuli.</td>
</tr>
<tr>
<td>sedation</td>
<td>There are various functional definitions of this term, which is sometimes used as a synonym for “hypnosis.” We use the term to connote drug-induced hypnosis with anxiolysis, diminished motor activity, and decreased arousal.</td>
</tr>
</tbody>
</table>

### Table 1. Glossary of Terms

<table>
<thead>
<tr>
<th>Term</th>
<th>Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>MAC</td>
<td>Median alveolar concentration, an end point that measures perceptive awareness rather than memory.</td>
</tr>
<tr>
<td>MAC-awake</td>
<td>The end-tidal concentration of inhaled anesthetic that prevents appropriate voluntary responses to spoken commands in 50 percent of a test population.</td>
</tr>
<tr>
<td>MAC-BAR</td>
<td>The end-tidal concentration of inhaled anesthetic that blocks changes in blood pressure and heart rate in response to surgical incision in 50 percent of a test population.</td>
</tr>
<tr>
<td>MAC-immobility</td>
<td>The end-tidal concentration of inhaled anesthetic that produces a behavioral effect at half the concentration of another anesthetic is said to be twice as potent.</td>
</tr>
<tr>
<td>MAC-stim</td>
<td>The end-tidal concentration of inhaled anesthetic required to block purposeful movement in an individual subject.</td>
</tr>
<tr>
<td>MAC-LOC</td>
<td>The end-tidal concentration of inhaled anesthetic that produces a behavioral effect at half the concentration of another anesthetic is said to be twice as potent.</td>
</tr>
<tr>
<td>LOC</td>
<td>The end-tidal concentration of inhaled anesthetic required to block purposeful movement in an individual subject.</td>
</tr>
<tr>
<td>LOC-stim</td>
<td>The end-tidal concentration of inhaled anesthetic required to block purposeful movement in an individual subject.</td>
</tr>
</tbody>
</table>

### The Spinal Cord

It is remarkable that anesthetic-induced ablation of movement in response to pain is mediated primarily by the spinal cord. Experiments in animals have shown that anesthetic actions in the brain are not required to inhibit motor responses to pain. In anesthetized rats, cervical transection of the spinal cord does not alter the MAC of a given anesthetic for limb stimulation. Similarly, in goats, selective administration of isoflurane to the body but not the brain (the medulla and above) also has little effect on the concentration that inhibits withdrawal from hind-leg pain. By contrast, hypnosis and amnesia are supraspinal effects.

General anesthetics decrease the transmission of noxious information ascending from the spinal cord to the brain, thereby decreasing supraspinal arousal. In goats, selective delivery of general anesthetics to the torso slows cortical electroencephalographic signals. Only when volatile anesthetics are delivered to the brain in concentrations that are nearly three times the control MAC do they produce immobility in goats. Therefore, it is likely that ascending signals from the spinal cord affect the hypnotic actions of anesthetics in the brain, whereas descending signals modify the immobilizing actions of anesthetics in the spinal cord.
Above the spinal cord, inhaled agents globally depress blood flow and glucose metabolism and selectively depress several supraspinal regions. For example, mildly hypnotic concentrations of isoflurane reduce task-induced brain activation in several distinct cortical regions, whereas activity in the visual cortex, motor cortex, and subcortical regions remains unchanged. Tomographic assessment of regional uptake of glucose in deeply anesthetized humans also indicates that the thalamus and midbrain reticular formation are more depressed than other regions. Evoked potentials traveling from the periphery to the sensory cortex show increased latency and decreased amplitude in patients under deep anesthesia with a volatile anesthetic. This signal degradation is discontinuous, occurring at specific relay sites in the thalamus and the deep cortex.

Although there is no definitive evidence that specific regions of the brain are targets of inhaled anesthetics, attention has focused on structures with roles in anesthetic-sensitive functions. The reticular-activating system, thalamus, pons, amygdala, and hippocampus are involved in cognition, memory, learning, sleep, and attentiveness. Interestingly, sleep states and general-anesthesia states share electroencephalographic and behavioral features. In both there is suppression of sensory input, inhibition of motor output, and analgesia. Although sleep and general-anesthesia states are clearly distinct, subcortical neuronal networks involved in the generation of sleep may also mediate some anesthetic effects. Recent studies imply that the tuberomammillary nucleus, a GABA-modulated region of the hypothalamus that is linked to sleep states, in the sedative actions of some intravenously administered general anesthetics and perhaps inhaled agents.

Most inhaled anesthetics produce generalized slowing, increased amplitude, and “frontal dominance” of electroencephalographic activity, yet surgical anesthesia has no electroencephalographic signature. As a result, some measurements derived from electroencephalographic data correlate well with hypnotic and immobilizing end points for individual agents, but no one measure can predict the depth of anesthesia induced by all inhaled agents or combinations of these agents. Nevertheless, uncoupling of coherent anteroposterior and interhemispherical electrical activity is consistently associated with anesthetic-induced unconsciousness (in this case, cessation of counting by patients during induction).

General anesthetics have long been known to interact with small cavities within most globular proteins, but with considerable selectivity. In a series of seminal experiments, Franks and Lieb established that a wide variety of anesthetics inhibit the lipid-free enzyme firefly luciferase in accord with the Meyer–Overton rule. The inhibition of luciferase even exhibits a long-chain alcohol cutoff, which is related to the size of the anesthetic-binding pocket. These observations were important because they demonstrated that protein sites may also contribute to the effects of general anesthetics. Although anesthetics alter the functions of a variety of cytoplasmic signaling proteins, including protein kinase C, the proteins considered most likely molecular targets of anesthetics are ion channels.

Ion channels are proteins that regulate the flow of ions across the cytoplasmic membrane. A variety of ion channels that modulate the electrical activity of cells are linked to the behavioral or physiological actions of anesthetics. Some of these channels are sensitive to various inhaled anesthetics. Ion channels that are sensitive to volatile anesthetics at clinically effective concentrations include both the superfamily of “cysteine-loop” neurotransmitter receptors, which includes nicotinic acetylcholine, serotonin type 3, GABA, and glycine receptors, and the glutamate receptors that are activated by N-methyl-d-aspartate (NMDA) or α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA). Within synapses, ion channels can influence the presynaptic release of neurotransmitters and alter postsynaptic excitability in response to the release of neurotransmitters. Voltage-gated ion channels for sodium, potassium, and calcium are also sensitive to some inhaled anesthetics, albeit usually at concentrations higher than those used clinically. A working hypothesis is that inhaled anesthetics enhance inhibitory postsynaptic channel activity (GABA and glycine receptors) and inhibit excitatory synaptic channel activity (nicotinic acetylcholine, serotonin, and glutamate receptors).
Accepted actions at GABA<sub>A</sub> receptors have received the most attention.

**GABA<sub>A</sub> receptors**

The GABA<sub>A</sub> receptors are the most abundant inhibitory neurotransmitter receptors in the brain. Each receptor is a heteromeric transmembrane protein complex that opens a chloride-permeable pore in response to GABA binding (Fig. 3). There are at least 18 distinct GABA<sub>A</sub>-receptor subunit genes in the human genome, and although most receptor complexes are thought to contain combinations of α, β, and γ subunits, a variety of combinations of subunits can form functional channels, and the neuroanatomical distribution of the various types of subunits is not homogeneous(256,388),(741,503). At clinically effective concentrations, general anesthetics increase the apparent sensitivity of receptors to GABA and prolong the receptor-mediated inhibitory current after a pulse of GABA release (Fig. 3). This augments GABA<sub>A</sub>-receptor–mediated inhibition of postsynaptic neuronal excitability. The potency with which volatile anesthetics enhance the function of GABA<sub>A</sub> receptors in vitro parallels MAC-immobility. Many other classes of general anesthetics also enhance GABA<sub>A</sub> responses, but nonimmobilizers do not. Paralleling the enhanced responses of GABA<sub>A</sub> receptors in vitro, positron-emission tomography in humans demonstrates concentration-dependent anesthetic modulation of GABA<sub>A</sub> receptors in the brain. These observations support a central role for GABA<sub>A</sub> receptors in anesthesia and, until recently, seemed to suggest a common mechanism for all inhaled general anesthetics.

**Other ion channels**

The modulation of GABA<sub>A</sub> receptors, however, is neither necessary nor sufficient to account for ev
Table 2. Roles of Some Anesthetic-Sensitive Ion Channels in Cellular Excitability, Behavior, Physiological Processes, and Pharmacology.

<table>
<thead>
<tr>
<th>Ion Channel</th>
<th>Cellular Roles</th>
<th>Behavioral, Physiological, and Pharmacologic Roles</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Ligand-gated</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>γ-Aminobutyric acid type A receptors</td>
<td>Increased chloride permeability; membrane hyperpolarization; inhibition of excitability</td>
<td>Enhanced activity associated with anxiolyis, sedation, amnesia, myorelaxation, anticonvulsant action</td>
</tr>
<tr>
<td>Glycine receptors</td>
<td>Increased chloride permeability; membrane hyperpolarization; inhibition of excitability</td>
<td>Spinal reflexes and startle responses; major inhibitory receptor in spinal cord</td>
</tr>
<tr>
<td>Neuronal nicotinic acetylcholine receptors</td>
<td>High permeability to monovalent cations and calcium; release of neurotransmitters</td>
<td>Association with memory, nociception, mutations linked with seizure disorders; autonomic functions</td>
</tr>
<tr>
<td>Muscle nicotinic acetylcholine receptors</td>
<td>Neuromuscular transmission</td>
<td>Skeletal-muscle contraction</td>
</tr>
<tr>
<td>Serotonin type 3 receptors</td>
<td>Increase excitability by inhibiting resting potassium-leak currents</td>
<td>Arousal; possible role in emesis</td>
</tr>
<tr>
<td>Glutamate receptors*</td>
<td>Fast excitatory neurotransmission Cation conductance for calcium and magnesium</td>
<td>Perception; learning and memory; nociception Perception and memory</td>
</tr>
<tr>
<td>N-methyl-d-aspartate</td>
<td>Cation conductance for calcium and magnesium</td>
<td></td>
</tr>
<tr>
<td>α-Amino-3-hydroxy-5-methyl-4-isoxazole propionic acid and kainate</td>
<td>Cation conductance for calcium and magnesium</td>
<td></td>
</tr>
<tr>
<td><strong>Other types</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Potassium channels</td>
<td>Modulation of cell resting potential and excitability; role in chemical, mechanical, and pH sensitivity</td>
<td>Nonspecific role; most likely widespread</td>
</tr>
<tr>
<td>Non–voltage-gated background channels</td>
<td>Recovery from action potentials</td>
<td>Nerve conduction; cardiac action potentials; mutations associated with cardiac arrhythmias Glucose sensor in β-cells Possible role in ischemic preconditioning</td>
</tr>
<tr>
<td>Voltage-activated</td>
<td>Inward rectifying channels; pH-sensitive</td>
<td></td>
</tr>
<tr>
<td>Non–voltage-dependent neurotransmitter or ATP-activated</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sodium channels</td>
<td>Generation and propagation of action potentials</td>
<td>Nerve conduction; cardiac action potentials (arrhythmias)</td>
</tr>
<tr>
<td>Calcium channels</td>
<td>Generation of pacemaker potentials in neurons (T-type) Presynaptic localization; neurotransmitter release</td>
<td>Cardiac inotropy and chronotropy; vascular tone Nonspecific role; most likely widespread</td>
</tr>
<tr>
<td>Voltage-gated cardiac (T-, N-, L-, and P-type)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Voltage-gated neuronal</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Calcium-induced calcium release</td>
<td>Intracellular channels Release of intracellular calcium stores after stimulation of surface receptors; production of calcium oscillations</td>
<td>Excitation–contraction coupling</td>
</tr>
<tr>
<td>Ryanodine receptor</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Inositol trisphosphate receptors</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* This group excludes metabotropic receptors.
The gaseous anesthetics xenon and nitrous oxide only minimally enhance GABA-mediated currents in vitro, and even high concentrations of cyclopropane and butane fail to alter the function of GABA_A receptors. These inhaled anesthetics clearly do not act directly through GABA-mediated mechanisms. Instead, clinical concentrations of these gases inhibit NMDA-sensitive glutamate channels and neuronal nicotinic acetylcholine receptors, suggesting that excitatory ligand-gated ion channels mediate an alternative pathway to anesthesia.

In addition to GABA_A receptors, other ion channels probably have roles in anesthetic-induced immobility. In spinal motor neurons, volatile anesthetics augment the activity of inhibitory glycine receptors and inhibit postsynaptic AMPA and NMDA receptors. The inhibition of glutamate receptors is apparently direct and is not due to augmented inhibitory GABA currents. Distinct ion channels may mediate different behavioral and physiological effects of inhaled anesthetics. Neuronal nicotinic acetylcholine receptors are inhibited by inhaled anesthetics at low concentrations that cause amnesia but not immobility, as well as by the volatile nonimmobilizers. Anesthetic inhibition of these receptors most likely impairs memory and learning but not immobility. In the heart, anesthetic inhibition of potassium and calcium channels is thought to underline negative chronotropic and inotropic actions as well as the pro-arrhythmogenic effects of anesthetics. The relative cardiac stability of patients under xenon anesthesia as compared with that of patients receiving halogenated agents correlates with xenon’s

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**Table 3. Functional Effect of Different Classes of Inhaled Anesthetics on Ion Channels.**

<table>
<thead>
<tr>
<th>Ion Channel</th>
<th>Halogenated Alkanes and Ethers</th>
<th>Nonhalogenated Alkanes</th>
<th>Xenon and Nitrous Oxide</th>
</tr>
</thead>
<tbody>
<tr>
<td>γ-Aminobutyric acid type A⁶⁹,₇₀</td>
<td>Enhancement</td>
<td>No effect</td>
<td>No effect</td>
</tr>
<tr>
<td>Glycine receptors⁶⁰,⁶¹</td>
<td>Enhancement</td>
<td>No effect</td>
<td>No effect</td>
</tr>
<tr>
<td>Neuronal nicotinic acetylcholine receptors⁵¹-⁶⁵</td>
<td>Strong inhibition</td>
<td>Strong inhibition</td>
<td>Inhibition</td>
</tr>
<tr>
<td>Muscle nicotinic acetylcholine receptors⁶⁶</td>
<td>Inhibition</td>
<td>Inhibition</td>
<td>ND</td>
</tr>
<tr>
<td>Serotonin receptors⁶⁴,⁶⁷</td>
<td>Weak inhibition</td>
<td>ND</td>
<td>No effect</td>
</tr>
<tr>
<td>Glutamate receptors</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>N-methyl-D-aspartate⁶⁴,⁶⁸,⁶⁹</td>
<td>Inhibition</td>
<td>Inhibition</td>
<td>Inhibition</td>
</tr>
<tr>
<td>α-Amino-3-hydroxy-5-methyl-4-isoxazole propionic acid and kainate⁶⁴,⁶⁵,⁶⁷</td>
<td>Inhibition</td>
<td>Inhibition</td>
<td>Inhibition</td>
</tr>
<tr>
<td>Background potassium channels⁷¹,⁷²</td>
<td>Enhancement or no effect†</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Voltage-activated potassium channels⁷²,⁷³</td>
<td>Inhibition or no effect</td>
<td>ND</td>
<td>No effect</td>
</tr>
<tr>
<td>ATP-activated potassium channels⁷⁴</td>
<td>Enhancement or no effect†</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Voltage-activated sodium channels⁷⁵,⁷⁶</td>
<td>Weak inhibition</td>
<td>Weak inhibition</td>
<td>ND</td>
</tr>
<tr>
<td>Voltage-activated calcium channels⁷³,⁷⁷,⁷⁸</td>
<td>Weak inhibition</td>
<td>ND</td>
<td>No effect</td>
</tr>
<tr>
<td>Ryanodine-activated calcium channels⁷⁹,⁸⁰</td>
<td>Enhancement or inhibition</td>
<td>ND</td>
<td>ND</td>
</tr>
</tbody>
</table>

* Effects are indicated qualitatively and represent the preponderance of data at clinical concentrations of anesthetic agents. In some cases, sensitivities of channels to anesthetics are strongly dependent on the composition of the subunits. ND denotes no data.
† The effects are agent-specific.
weaker inhibition of both L-type calcium currents and voltage-gated potassium currents in cardiac myocytes. The efficacy with which anesthetic agents produce ischemic preconditioning in the myocardiun also correlates with their actions on ATP-sensitive potassium channels.

**ANESTHETIC SITES ON ION-CHANNEL PROTEINS**

Because ion channels function within lipid membranes, it is difficult to discern whether their modulation by anesthetics is caused indirectly by changes in membrane structure or directly by binding to protein sites. The most thoroughly verified protein site is on the peripheral nicotinic acetylcholine receptor, a structural homologue of both neuronal nicotinic acetylcholine and GABA<sub>A</sub> receptors (Fig. 3). Reversible binding of a radio-labeled general anesthetic was demonstrated with highly purified peripheral nicotinic acetylcholine receptors. The kinetics of the interruption of the opening of single nicotinic acetylcholine–receptor channels by anesthetics are consistent with the existence of a direct channel-blocking mechanism but not with an indirect (e.g., lipid-mediated) mechanism. Competition between two anesthetics for an inhibitory site on open nicotinic acetylcholine–receptor channels was also demonstrated. Experiments involving mutagenesis, electrophysiology, and photolabeling have mapped the inhibitory site to the nicotinic acetylcholine–receptor pore. In the homologous GABA<sub>A</sub> receptor, sites that appear critical for the modulation of volatile anesthetics have also been identified. These are located in multiple transmembrane domains that may form a single binding pocket (Fig. 3).

**SYNAPtic MECHANISMS**

The neuronal functions underlying network activity are axonal conduction and synaptic transmission. Clinical concentrations of inhaled anesthetics affect the latter much more than the former. Inhaled anesthetics both depress excitatory synapses and augment inhibitory synapses. Studies aimed at quantifying the presynaptic and postsynaptic effects of anesthetics have demonstrated actions on both the release of neurotransmitters and the function of neurotransmitter receptors, with the latter having a dominant role. In addition, some volatile anesthetics interact at GABA<sub>A</sub>-receptor sites formed between several transmembrane elements.
Inhibitory synapse

- Presynaptic neuron
- Action potential
- GABA release
- Postsynaptic membrane
- Anesthetic site on GABA_A receptor
- Agonist binding sites

Excitatory synapse

- Presynaptic neuron
- Release of acetylcholine
- Postsynaptic membrane
- Anesthetic site on nicotinic acetylcholine receptor
- Agonist binding sites

Inhibitory Postsynaptic GABAergic Currents

- No anesthetic
- With anesthetic
- Membrane Current vs. Log GABA

Excitatory Postsynaptic Cholinergic Currents

- No anesthetic
- With anesthetic
- Membrane Current vs. Log Acetylcholine

In vivo and in vitro neural networks

Elucidating the effects of inhaled anesthetics on neural networks in vivo has proved technically difficult, in part because reliable electrophysiological recording in animals often requires sedating them with other anesthetic agents. Many in vitro studies have recorded electrical activity in brain or spinal cord slices, which maintain local synaptic interactions. Small cortical slices with intact local networks demonstrate synchronized electrical rhythms that can be slowed either by enhancing GABA-mediated transmission or by inhibiting glutamate-mediated processes.
ated transmission with general anesthetics. The effects of anesthetics on the frequencies of local cortical networks probably contribute directly to electroencephalographic slowing. On the other hand, long-range (e.g., thalamocortical) circuits coordinate rhythmic activity among distant brain regions. The effects of anesthetics on the rhythmic frequency of these networks may depend on the rates of decay of GABA<sub>A</sub> receptor–mediated inhibitory potentials, which are prolonged by inhaled anesthetics (Table 3). The hippocampus and spinal cord also contain circuits that are most likely involved in the amnesic and immobilizing actions of general anesthetics.

Highly simplified neural circuits have proved more amenable to analysis. The effects of inhaled anesthetics on respiration have been investigated by recording caudal ventral respiratory neurons of the medulla in decerebrate dogs. Clinically effective concentrations of inhaled anesthetics alter both glutamate-mediated and GABA-mediated signals to pacing neurons; sevoflurane reduces output motor-neuron activity more than does halothane, paralleling the clinical observation that sevoflurane depresses respiration more deeply than does halothane.

**STUDIES IN GENETICALLY MODIFIED MAMMALS**

**Knockout Studies**

Animals in which specific genes have been knocked out have been used to evaluate the role of two ligand-gated ion channels in anesthesia, the AMPA-sensitive glutamate receptor and the GABA<sub>A</sub> receptor. The GluR2 subunit of AMPA receptors was chosen for study, because it is the most common subunit and it determines anesthetic sensitivity in vitro. In GluR2-knockout mice, the MAC of volatile agents was unaltered, sensitivity to loss of righting reflexes was moderately increased, and nociception was increased. The genes for the <i>β</i>3 and <i>δ</i> subunits of GABA<sub>A</sub> receptors were targeted, because of their patterns of expression in the brain and the anesthetic sensitivity that they confer on in vitro receptors containing different types of subunits. In GABA<sub>A</sub> <i>β</i>3–knockout mice, the MAC of enflurane was slightly diminished, but there was no change in sensitivity to the loss of righting reflexes. In GABA<sub>A</sub> <i>δ</i>–knockout animals, there was no change in the MAC of volatile anesthetics or in the sensitivity to the loss of righting reflexes. Several themes emerged from studies of such knockout mice. First, anesthetic sensitivity measured on the basis of the loss of righting reflexes and MAC-immobility were affected in different ways by genetic alteration, adding to the evidence that different anesthetic-induced forms of behavior are mediated by distinct mechanisms. Second, the mechanisms that mediate even a single end point, such as immobility, are complex and agent-dependent, as shown by the following: knockout of the GABA<sub>A</sub> <i>β</i>3 gene decreased the MAC of enflurane but had far less effect on the MAC of halothane and no effect on the enflurane-induced depression of evoked spinal motor potentials. The data on GABA<sub>A</sub> receptor–knockout mice also suggest that specific types of receptor subunits, specifically <i>β</i>3, may have dominant roles in some anesthetic actions. Other mice have been created in which GABA<sub>A</sub> receptor <i>α</i>1, <i>β</i>2, and <i>δ</i> subunits have been inactivated, but the sensitivity of these animals to inhaled anesthetics has not yet been reported.

**“Knock-In” Studies**

Knocking out the expression of an ion-channel–subunit gene may induce changes in the composition of the subunits, the network circuitry, or both. The introduction of specific mutations into...
native genes (“knock-in” animals) avoids these pitfalls and enables an assessment of the physiologic and pharmacologic roles of specific proteins and even small regions within proteins. The power of this approach is evident from recent revelations about the roles of specific GABA<sub>A</sub>-receptor subunits on the actions of benzodiazepines, which induce some behavioral effects similar to those of general anesthetics. Knock-in models are also being employed to examine the role of GABA<sub>A</sub> receptors in mediating general anesthesia. A mutation in the GABA<sub>A</sub> receptor β<sub>3</sub> subunit that attenuates in vitro modulation by the intravenous anesthetic etomidate has been introduced into mice. These animals have dramatically reduced sensitivity to etomidate with respect to the end points of nociceptive withdrawal and the loss of righting reflexes, but their sensitivities to volatile anesthetics are only moderately decreased (with respect to MAC) or unaffected (with respect to the loss of righting reflexes). This result is further evidence of a role for the β<sub>3</sub> subunit in determining MAC and is consistent with in vitro studies showing that mutations in α subunits have a greater effect on the sensitivity of GABA<sub>A</sub> receptors to volatile anesthetics than do mutations in β subunits. Studies using knock-in animals with these α-subunit mutations are expected to provide additional insights into the roles of GABA<sub>A</sub> receptors in the actions of inhaled anesthetics.

**Summary**

Simplifying assumptions such as the unitary hypothesis led early research on anesthesia to focus on nonspecific biophysical mechanisms. Reevaluation of the actions of anesthetics on a variety of neurobiologic-systems levels has revealed important new insights into mechanisms that contradict these nonspecific hypotheses. Thus, although all inhaled general anesthetics produce amnesia and suppress motor responses to noxious stimuli, their actions on other behavioral and physiological responses vary. The suppression of nociceptive motor responses by anesthetics is mediated primarily by the spinal cord, whereas hypnosis and amnesia are mediated within the brain. These actions may also be associated with separate molecular targets. Important actions of inhaled anesthetics are associated with altered activity of neuronal ion channels, particularly the fast synaptic neurotransmitter receptors such as nicotinic acetylcholine, GABA<sub>A</sub>, and glutamate receptors. There is also growing evidence that anesthetics affect neuronal ion channels by binding directly to protein sites. Different ion channels display strikingly unique sensitivities to various inhaled anesthetics, suggesting that different channels are involved in distinct behavioral effects of anesthetics and that several mechanistic pathways may converge to produce similar anesthetic states. Neuroanatomical differences in the distribution of various ion channels and their specific subunits are likely to influence specific behavioral effects of inhaled anesthetics. This emerging view of the specific neurobiologic actions of inhaled anesthetics suggests that these widely used drugs should be amenable to improvements by rational design.

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