Lifting the Fog around Anesthesia

Learning why current anesthetics are so potent and sometimes dangerous will lead to a new generation of safer targeted drugs without unwanted side effects

By Beverley A. Orser

A Hollywood thriller due out this year centers on a young man who awakens while undergoing open-heart surgery but is unable to move or cry out. The film's plot will undoubtedly take many more dramatic turns from there, but its early premise is, sadly, not entirely far-fetched. Episodes of intraoperative awareness while under general anesthesia are reported by one or two of every 1,000 patients. In reality, such incidents are usually brief and generally do not involve pain or distress, but they do highlight one of several ways that even the newest generation of anesthetic drugs can sometimes leave much to be desired. Indeed, the medical specialty of anesthesiology has evolved into a sophisticated art form because scientific understanding of how anesthetic drugs actually work, and how to make them better, has lagged behind most other areas of drug development.

Many of the modern anesthetics, in fact, share structural properties and clinical effects with ether, whose application as an anesthetic was first successfully demonstrated in public by Boston dentist William Morton in 1846. Since then, the use of general anesthesia has expanded to 40 million patients each year in North America alone. Yet advances in anesthetic care since Morton's day have come largely from the development of complex drug delivery systems and strategies for managing anesthesia's dangers and side effects.

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Today's general anesthetics are the most potent depressors of nervous system activity used in medicine. They even affect regulation of breathing and heart function. As a result, the drugs have a fairly narrow margin of safety, which is the difference between the therapeutic dose and a dose that is toxic, even lethal. That is one reason why individuals whose lung or cardiovascular function is already unstable—such as trauma victims undergoing emergency operations or patients in the midst of heart surgery—must receive a lighter than normal dose of anesthesia, which could make them susceptible to brief awareness incidents, as in the movie.

Although radical improvements in the care of people under general anesthesia have laid the foundation for complicated procedures such as organ transplants and open-heart surgery, the powerful neurodepressive effects of these drugs make them more likely to cause death during an operation than the surgical procedure itself. And because anesthesia-related mortality has plateaued at a rate of approximately one patient in 13,000 for the past 15 years, it appears that anesthesiologists may have reached the limits of our ability to deliver these toxins safely. Moreover, severe side effects—ranging from loss of airway control to memory and cognitive problems after general anesthesia—may also stem from the broad yet poorly understood influence that current anesthetics exert on the central nervous system.

Science should be able to do much better, and very recent research is beginning to reveal how it can.

Pulling Out the Plugs

ALL OF TODAY'S general anesthetic drugs were developed empirically, which is to say, they were tested for their ability to produce the desirable effects that define the anesthetized state. Anesthesia's main components are sedation, unconsciousness (also sometimes called hypnosis), immobility, absence of pain (analgesia) and absence of memory for the anesthetized period (amnesia). By studying the mechanisms through which anesthetics achieve these end points, many groups, including my own at the University of Toronto, are beginning to tease those effects apart. Such studies are revealing that the activity of these potent drugs involves highly specific interactions with subpopulations of nervous system cells to create each of the separate properties of anesthesia.

Armed with this knowledge, we will be poised to finally move beyond the ether era and develop a new generation of highly specific drugs that can be used in combinations to deliver only the desired results without the dangers. As a bonus, this research is also yielding insights that can improve related therapies, such as sedatives and sleep aids, that

share some of anesthesia's mechanisms.

Anesthetics fall into two main categories based on whether they are delivered by inhalation, such as isoflurane, or intravenously, such as propofol. These drugs may appear to induce a deep sleep, but the state produced by most modern general anesthetics is more of a pharmacological coma. In a step toward clarifying the mechanisms underlying their effects, technologies such as magnetic resonance imaging (MRI) and positron-emission tomography (PET) have helped identify some of the discrete brain regions and neural circuits involved in specific components of the anesthetized state. For instance, anesthetic action on the spinal cord accounts for the immobility produced by the drugs, whereas drug-induced changes to the hippocampus, a brain structure involved in memory formation, have been linked to amnesia. Chronic memory impairment following surgery, one of the undesirable side effects suffered by some patients, may also represent a leftover influence of the drugs on the hippocampus.

Because consciousness is a complex experience whose defining properties are still hotly debated by neuroscientists, it is not as easy to pinpoint a single anatomical source of unconsciousness during anesthesia. One leading theory holds that it is simply the result of "cognitive unbinding"—a severing of communication between the many brain regions that usually cooperate in higher cognitive processing. Even at the local level, if one imagines groups of neurons as forming lines in a vast telephone network, the effect of general anesthesia is analogous to pulling out plugs at the switchboard. Researchers are, however, making encouraging progress in discovering details about the ways that anesthetic drugs physically act on the individual cells of the nervous system to block their transmissions.

During most of the 20th century, anesthetics were widely thought to work by disrupting the lipid components of cell membranes. Most anesthetics are highly fat-soluble compounds with widely differing chemical structures that range from simple inert gases to complex steroids. Their great physical and chemical diversity supported the idea that anesthetics must work in some nonspecific way to depress neuronal functioning. Modern research has shown, however, that anesthetics actually interact with multiple varieties of specific proteins, known as receptors, found on the surface of nerve cells. Families of receptors contain subtly different versions, however, which tend to predominate in different areas of the central nervous system. The presence of particular receptor subtypes on only certain subpopulations of cells will thus determine which cells are influenced by an anesthetic.

Contemporary studies are therefore focusing on identifying which receptor variants are the targets of current anesthetic drugs, understanding how the drugs interact with the receptors to change the cell's function and how those cellular changes produce both the desired and unwanted "symptoms" of anesthesia.

The effect of anesthesia is analogous *to pulling out plugs at the switchboard.*

Overview/*Refining a Blunt Instrument*

- General anesthetics are powerful nervous system suppressors, but how the drugs produce their broad effects throughout the brain and body is poorly understood.
- Investigation of anesthetics' underlying mechanisms is revealing that individual aspects of the anesthetized state are attributable to different sets of nerve cells, which are themselves distinguished by specific surface proteins that interact with the drugs.
- New compounds designed to target just those proteins, and hence only specific cell types, could be combined to selectively produce the desirable effects of anesthetics—as well as sedatives, sleep aids and memory drugs—with fewer risks and side effects.

Signaling Silence

MANY CATEGORIES of receptor proteins are found on the surface of neurons, but those activated by natural neurotransmitter chemicals have garnered the most interest in anesthesia research because they critically regulate communication along the neural "telephone lines." As their name implies, neurotransmitter molecules transmit messages between neurons at points of contact called synapses. They do so by traveling from the so-called presynaptic neuron across a tiny gap to bind to receptors on the postsynaptic neuron's cell membrane. When enough neurotransmitter molecules trigger the appropriate receptors, the postsynaptic cell's membrane generates an electrical potential that travels down its length to the next neuron in its network. Glutamate, serotonin, norepinephrine and acetylcholine are just a few of the neurotransmitters widely studied for their role in promoting such signaling throughout the central nervous system.

In anesthesia research, however, another neurotransmitter called gamma-aminobutyric acid (GABA) has gained the most attention because of its ability

to block neural communication. GABA is an inhibitory neurotransmitter: it helps to maintain overall balance in the nervous system by dampening neurons' ability to respond to excitatory messages from other cells. For that reason, GABA is thought to play a central part in the actions of anesthetic drugs.

Most postsynaptic receptors on cells that interact with GABA belong to a class termed ligand-gated ion channels. When GABA (the ligand) binds to the receptor, the receptor changes its conformation, temporarily opening a channel that admits negatively charged ions into the cell. The increased ion concentration generates a negative potential, preventing the cell from being able to produce an excitatory electrical pulse.

The receptor that is believed to be a primary target for anesthetics is the GABA subtype A, or $GABA_A$, receptor, which is also known to underlie the therapeutic effects of other classes of sedative and hypnotic drugs, most notably benzodiazepines such as Valium. Very low concentrations of benzodiazepines increase $GABA_A$ receptor function, a relation that is easily confirmed because reversal agents that impede benzodiazepine from binding to the $GABA_A$ receptor rapidly blunt the effects of those drugs.

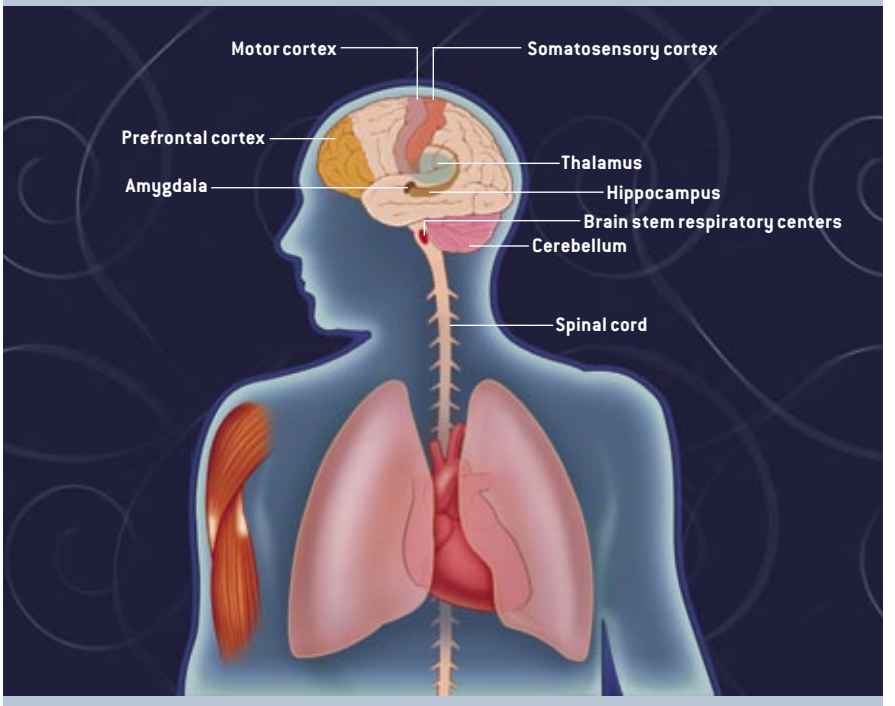
Unfortunately, no such reversal agents exist for general anesthetics that might provide clues to their receptor targets. Nevertheless, studies using slices from many different brain regions and neurons grown in tissue culture have shown that both intravenous and inhaled anesthetics prolong the duration of postsynaptic electric currents generated by $GABA_A$ receptors.

Anesthetics are believed to increase the function of $GABA_A$ receptors by interacting at discrete binding cavities or attaching to specific amino acids in the receptors themselves and prolonging the channel opening, which extends the inhibitory effects of GABA molecules bound to the receptor. At high enough concentrations, anesthetics may even trigger the GABA receptors alone.

The vast majority of neurons contain $GABA_A$ receptors, however, so scientists could not understand how anesthetics

Anesthesia's Broad Impact

Both the desirable and unwanted effects of anesthetic drugs stem from their power to suppress neuronal activity throughout the central nervous system, which encompasses the brain and spinal cord and controls heart rate and breathing. Ongoing research is attempting to pinpoint the neural structures and regions whose changed functioning produces each of the defining properties of the anesthetized state.



Components of the Anesthetized State

Sedation

Reduced arousability, as evidenced by longer response times, slurred speech and decreased movement. Neuronal activity across brain cortical areas drops.

Unconsciousness (also called hypnosis)

Impaired perception of, and response to, stimuli. Cortical depression is deeper than in sedation. Activity in the thalamus, an area important for integrating brain processes, also falls significantly.

Immobility

Lack of movement in response to stimulation such as shaking or heat. Suppression of spinal cord neuronal activity is the main cause of this temporary paralysis, although the cerebellum, a motor control area, may also contribute.

Amnesia

Lack of recall for the anesthetized period. Many brain structures involved in memory formation, including the hippocampus, amygdala, prefrontal cortex and sensory and motor areas, exhibit anesthetic-induced changes.

Others

Muscle relaxation and lack of pain (analgesia) are sometimes included in definitions of the anesthetized state and are largely attributed to depression of spinal cord activity.

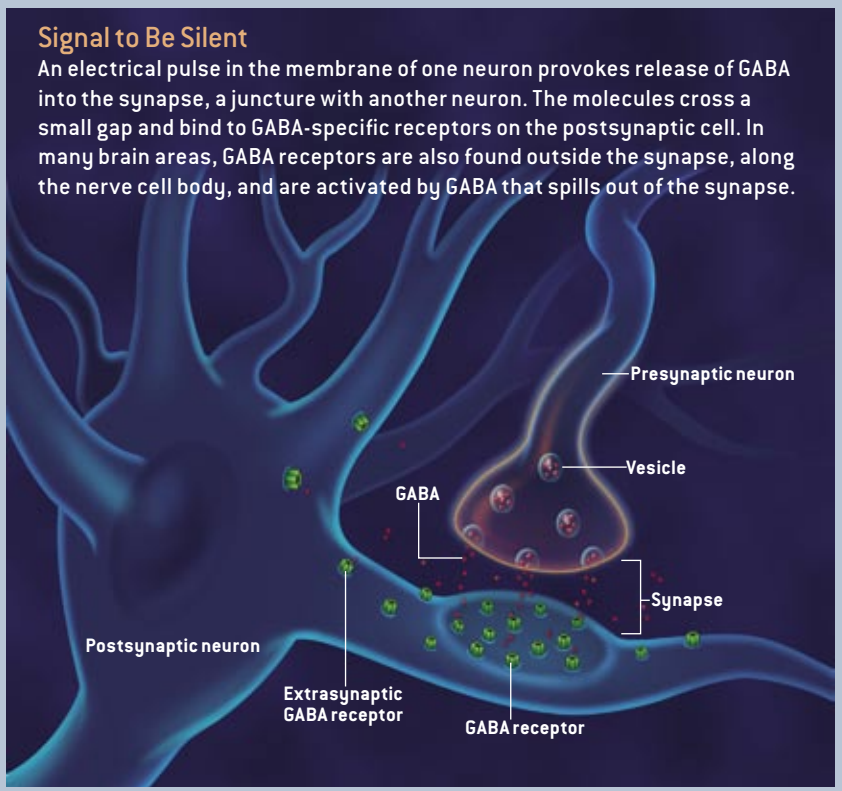
CHRISTINE GRALAPP

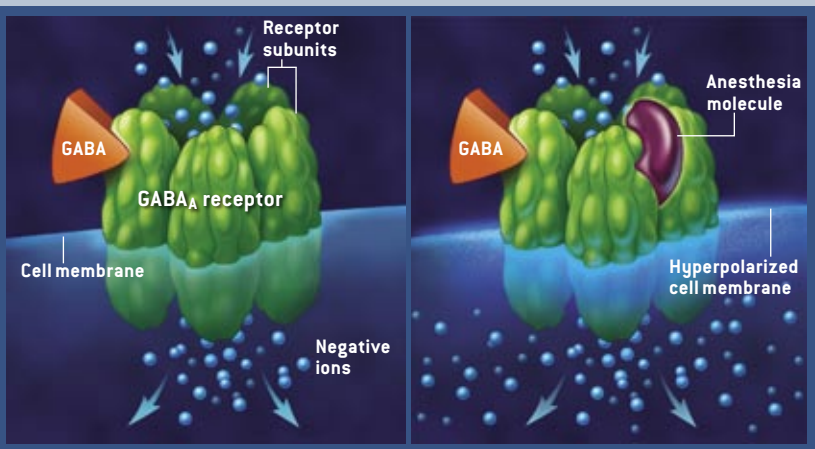
Jamming Transmission

Anesthetic drugs have been found to dampen neuronal communication, in part, by enhancing the effects of the neurotransmitter GABA, a signaling molecule that inhibits nerve cells from firing. Current research is focused on how the drugs interact with cellular GABA receptors to block neural activity.

Signal to Be Silent

An electrical pulse in the membrane of one neuron provokes release of GABA into the synapse, a juncture with another neuron. The molecules cross a small gap and bind to GABA-specific receptors on the postsynaptic cell. In many brain areas, GABA receptors are also found outside the synapse, along the nerve cell body, and are activated by GABA that spills out of the synapse.





Anesthetics and GABA: Changing Charge

A receptor subtype called $GABA_A$ is a channel into the postsynaptic cell composed of five protein subunits. When GABA binds to it, the receptor opens to admit negatively charged ions, which increases polarization of the cell's membrane and prevents the neuron from generating an electrical pulse (*left*). Anesthetics are thought to act by binding to clefts in the $GABA_A$ receptor and prolonging the channel opening, which causes hyperpolarization of the cell membrane (*right*).

could selectively influence different brain regions until research breakthroughs over the past decade revealed that not all $GABA_A$ receptors are structurally or pharmacologically the same. The $GABA_A$ receptor is a protein complex composed of five subunit parts, which can be mixed and matched in various combinations. At least 19 different $GABA_A$ receptor subunits exist in mammals, and most of those have variant subtypes, so the possible number of combinations is high. The subunits most commonly seen in neurons, however, are the ones designated alpha, beta and gamma. In fact, most $GABA_A$ receptors are composed of two alpha subunits, two betas and one gamma, although sometimes a delta or epsilon subunit replaces the gamma, depending on the brain region. But the key discovery was that the receptor's subunit composition dramatically alters its pharmacological properties: just one subunit difference within a $GABA_A$ receptor's structure can determine whether and how it will respond to a particular anesthetic drug.

Because different $GABA_A$ receptor subtypes predominate in different brain regions, researchers are increasingly able to pinpoint how anesthetics produce specific effects in various parts of the central nervous system by examining how the drugs interact in those regions with their particular target receptors.

Narrowing Down Targets

MY COLLEAGUES and I decided to focus on identifying the receptors that influence the memory-impairing properties of anesthetics, so we concentrated our studies on $GABA_A$ receptors in the hippocampus. Anesthetics are known to cause amnesia at doses considerably lower than those required for unconsciousness or immobility, an effect that is clearly evident to anesthesiologists, for example, because patients rarely remember their own animated conversations as they go under or emerge from anesthesia. Yet, for unknown reasons, some patients experience unexpected recall of events during the surgery itself. Thus, by finding the correct target receptors for the amnesia-inducing effects

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of anesthesia, it may become possible to identify patients at risk for intraoperative awareness because they lack those receptors. Alternatively, drug strategies to prevent awareness or at least its recollection could also be developed.

In the course of this work, we discovered to our surprise that even receptors outside the synapse could play a role in anesthetic action. If the synapse serves as a switchboard at the junction between two cells, then receptors at the synapse periphery or scattered along the nerve cell body can be imagined as residing on the telephone line itself. Such extrasynaptic $GABA_A$ receptors are activated by even the very low concentrations of GABA that are naturally present in the extracellular space or that spill over from nearby synapses. As it turns out, high numbers of extrasynaptic receptors are found in certain brain regions, such as the hippocampus and the thalamus (an area involved in consciousness and pain processing), as well as parts of the cortex and the cerebellum.

We serendipitously learned the relevance of extrasynaptic $GABA_A$ receptors as anesthetic targets after struggling unsuccessfully for quite some time to identify postsynaptic receptors that were sensitive to the very low amnesia-inducing concentrations of anesthetics. We had also searched for populations of postsynaptic receptors that were synergistically modulated by midazolam and propofol, two of the most commonly used intravenous neurodepressive drugs, and had not found any of those either. Our work, however, was based on taking electrophysiological recordings of currents generated in hippocampal neurons in tissue culture, and we did notice that amnesia-producing concentrations of anesthetics significantly increased a persistent low-amplitude current generated by extrasynaptic $GABA_A$ receptors. Rather than provoking a response at the "switchboard," the drugs were acting to boost a kind of static or inhibitory buzz in the telephone line itself that interfered with communication.

We found that the injectable anesthetics propofol and etomidate, and even the inhaled anesthetic isoflurane, increased the amplitude of this current by as much as 35-fold at concentrations several times lower than those required to cause immobility. Other investigators, including Stephen G. Brickley, Mark Farrant and their colleagues at University College London, had already described this steady low current even in the absence of anesthetics. But what surprised our group was the extrasynaptic receptors' remarkable sensitivity to tiny amounts of both inhaled and intravenous anesthetics, whereas the low concentrations of anesthetics caused only negligible changes in postsynaptic currents. Previous studies, such as our own, had apparently focused on the right family of receptor proteins but were looking in the wrong location.

Eventually our experiments determined that the extrasynaptic $GABA_A$ receptors were structurally slightly different from the populations of receptors within the synapse in that they predominantly contained an alpha-5 subunit, which the postsynaptic receptors generally lacked. That single change seemed to account for their sensitivity to even tiny amounts of anesthetics. These results were exciting to us because accumulating evidence from neuroscientists working on other research questions had also indicated that $GABA_A$ receptors containing the alpha-5 subunit are involved in normal hippocampal-dependent memory processes, supporting our theory that the extrasynaptic alpha-5 receptors were responsible for the memory effects of an anesthetic. To test our hypothesis further, we turned to experimenting with genetically modified mice that lacked the alpha-5 subunit and wild-type mice that had the normal receptor. As expected, in behavioral tests the wild-type mice were sensitive to amnesia-causing doses of etomidate, whereas the alpha-5-deficient mice failed to manifest the drug's effects on memory.

We also established that the loss of alpha-5 $GABA_A$ receptors had no consequences for any of the other anesthesia end points: sedation, immobility, hypnosis and response to a painful stimulus were the same in both groups of mice. These results demonstrated that the memory-impairing effects of etomidate could be dissociated from the drug's other properties based on the pharmacology of specific receptor subunits. They also provided the first animal model for receptor variations that might occur in humans and could explain some cases of resistance to an anesthetic's ability to induce amnesia. Ongoing studies will determine whether other general anesthetic drugs also preferentially target alpha-5 $GABA_A$ receptors to produce amnesia.

At the same time, laboratories in Europe and the U.S. have been employing similar experimental techniques to ex-

Drug strategies to prevent awareness or *at least its recollection could be developed.*

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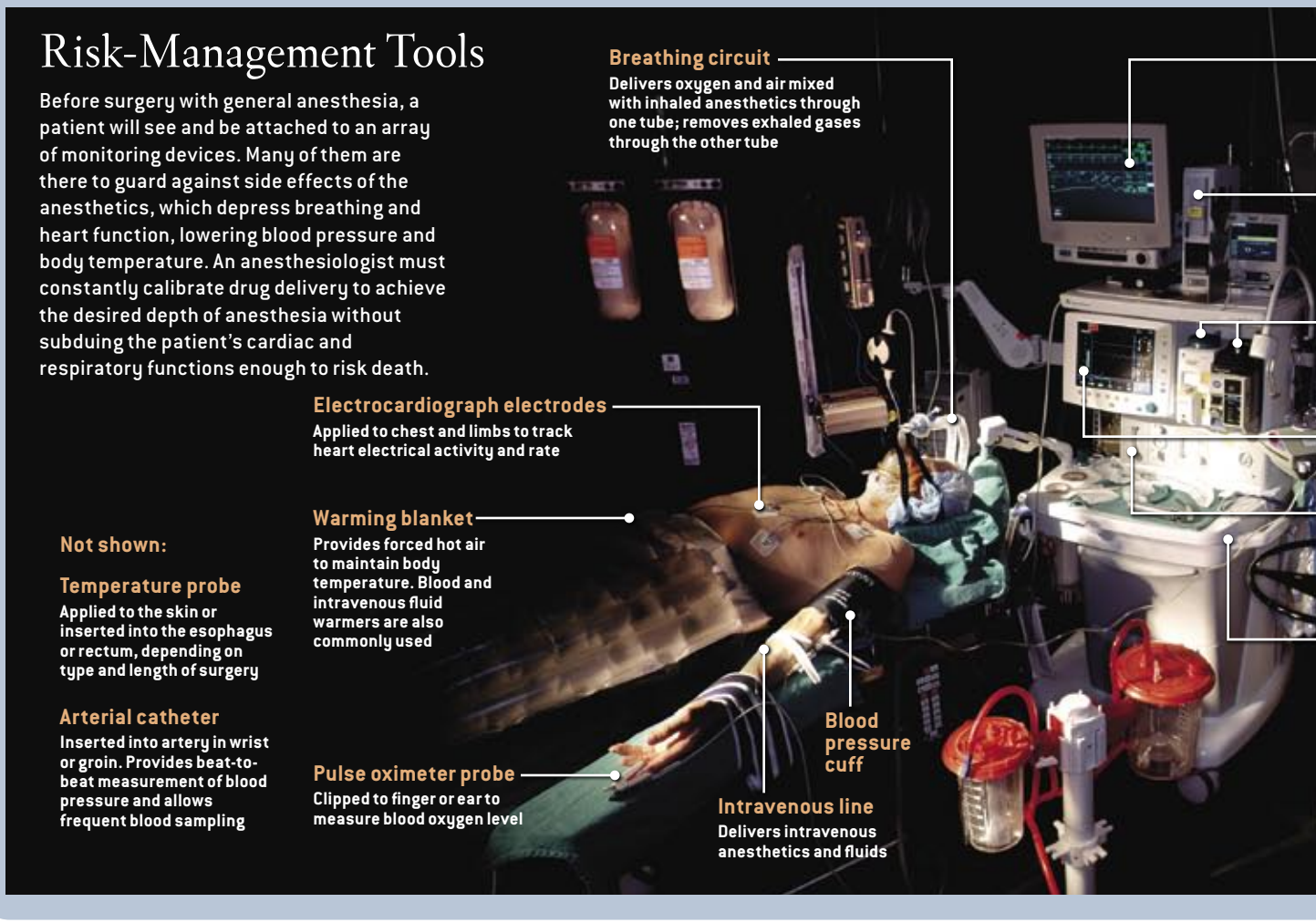
Risk-Management Tools

Before surgery with general anesthesia, a patient will see and be attached to an array of monitoring devices. Many of them are there to guard against side effects of the anesthetics, which depress breathing and heart function, lowering blood pressure and body temperature. An anesthesiologist must constantly calibrate drug delivery to achieve the desired depth of anesthesia without subduing the patient's cardiac and respiratory functions enough to risk death.



plore the hypnotic and immobilizing effects of anesthetics. Gregg E. Homanics of the University of Pittsburgh School of Medicine, for example, developed a mouse lacking the $GABA_A$ receptor delta subunit, which is known to confer high sensitivity to neurosteroids. His group's investigation found that the delta-deficient mice were, predictably, less sensitive to the steroid-based anesthetic alphaxalone in tests of the drug's power to induce unconsciousness. The mutant mice, however, displayed no differences in their responses to propofol, etomidate and other nonsteroidal anesthetics as compared with wild-type controls. Steroid anesthetics are not commonly used today, but these results also demonstrated the principle that different classes of anesthetics target discrete subpopulations of $GABA_A$ receptors.

Such experiments have truly overturned the old notion that because anesthetics are so chemically different from one another they must produce their multiple effects by some general mechanism. Instead empirical development of anesthetic drugs seems to have stumbled on chemicals that produce similar end points, though each by its own unique mechanisms.

Etomidate, for example, is the only anesthetic in clinical use that is selective for $GABA_A$ receptors containing the beta-2 or beta-3, but not the beta-1, subunit. Indeed, the differences between the beta-subunit variants that respond to etomidate and those that do not involve a single amino acid change at a specific point in the subunit's protein structure. The pharmaceutical company Merck developed transgenic mice with a mutation in that amino acid location within the beta-2 subunit and found that etomidate was less effective at producing unconsciousness in the animals; the drug's immobilizing properties remained, however. Uwe Rudolph, while at the University of Zurich, also generated transgenic mice with the same mutation in the beta-3 subunit and found that it greatly diminished the effectiveness of both etomidate and propofol in producing unconsciousness and analgesia in the animals. In contrast, he showed that alphaxalone was equally effective in wild-type mice and those carrying the mutation, which indicates that these receptor subunits are probably not important targets of that drug.

Whether the point mutations in beta-2 and beta-3 receptor subunits also influence the drugs' amnesia-inducing properties has not yet been established. And which central nervous system regions in the transgenic mice are affected

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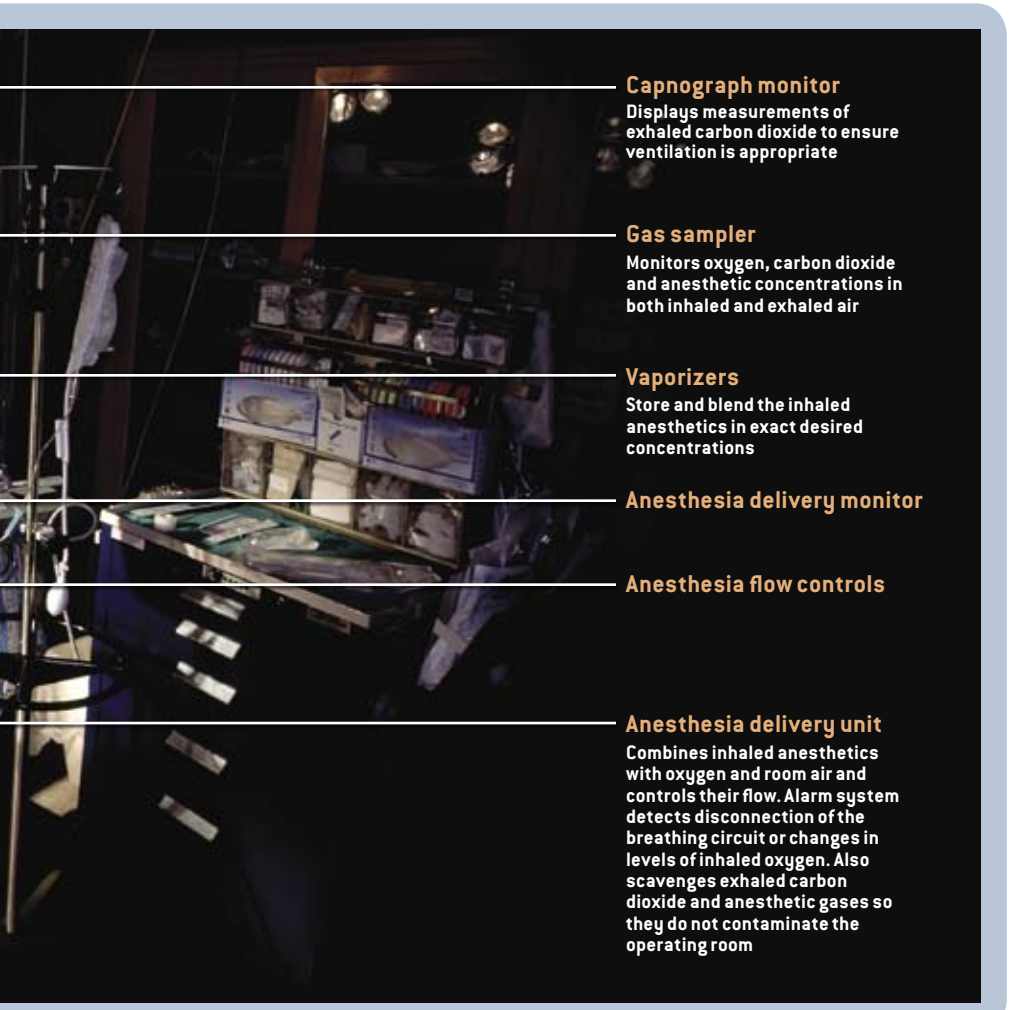

by the mutations is another unknown, although some evidence suggests that extrasynaptic $GABA_A$ receptors in the thalamus may be critical. Taken together, however, such studies are confirming the central role of $GABA_A$ receptors in the actions of anesthetics. The next step is to begin translating this knowledge gained from current general anesthetics into drugs that are anything but general.

Tailored Treatment

AS THE WORK of my research group and others has shown, extrasynaptic alpha-5 $GABA_A$ receptors in the hippocampus are vital to the amnesia-inducing effects of etomidate and possibly to other general anesthetics currently in use. These results suggest that drugs that avoid or target that particular receptor could selectively spare or block memory formation as needed.

In fact, such compounds are already in development for other uses. Sedative-hypnotic drugs that do not act on the alpha-5 subunit, and hence should lack the memory-fuzzing effects of benzodiazepine sedatives and certain sleeping pills, are in the preclinical pipeline. And clinical trials of Gaboxadol, the first drug to selectively *target* extrasynaptic $GABA_A$ receptors to enhance their function, are currently under way. Gaboxadol was initially developed as an anticonvulsant but is now being studied as a sleep-promoting drug. It targets $GABA_A$ receptors containing the delta subunit, primarily found in the thalamus and cerebellum, and therefore may also avoid affecting memory. The memory-blocking potential of similar compounds that do interact with alpha-5 receptors could also prove very useful in the surgical setting, where drugs that cause profound amnesia without depressing respiration, airway reflexes or the cardiovascular system might be highly desirable. In combination with other anesthetics, a potent memory blocker could be used to prevent intraoperative awareness episodes, for example. Alone, such a drug might be helpful in the treatment of patients suffering post-traumatic stress disorder by inhibiting certain distressing memories.

Management of anesthesia's effects on memory is just one example of a new approach to anesthesiology that will become possible with such targeted drugs. In many situations, the broad and profound neurodepression of current anesthetics is unnecessary and undesirable anyway. With a cocktail of compounds, each of which produces only one desirable end point, the future version of anesthesia care could leave a patient conversant but pain-free while having a broken limb repaired or immobile and sedated but aware while having a hip replaced. This polypharmaceutical approach is already widely used for other aspects of surgery-related care, most notably in the treatment of postoperative pain.

Today anesthesia has never been safer, but it is certainly not without risk. A tremendous opportunity now exists for the field to move beyond the ether era and toward a truly modern model of anesthesia care. SA

MORE TO EXPLORE

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