New and Notable

Anesthesia, Analgesia, and Euphoria

Betty J. Gaffney
Department of Biological Science, Florida State University, Tallahassee, Florida

Seeking a unified theory of achieving the state induced by anesthetics is deep in the biophysicist's psyche. A search of the *Biophysical Journal* contents for titles containing ''anesthetic'' yields 84 hits. Both sleep and anesthesia uncouple functional connections between activities in hemispheres of the brain, as demonstrated in functional magnetic resonance imaging (1). At the molecular level, however, it appears that different anesthetics have varied targets, and instead of a unified theory of action, there is a complicated puzzle. Specific interactions between neurotransmitter-gated channels and anesthetics receive much attention. Many volatile (and also some intravenous) anesthetics modulate $GABA_A$ receptors. However, the group of inhaled anesthetics—xenon, nitrous oxide, and cyclopropane—mainly influence the function of postsynaptic glutamate NMDA receptors instead (reviewed in Sanders et al. (2)). At intermediate doses, these inhalant anesthetics lead to analgesia and euphoria and, at higher doses, to deep, surgical anesthesia.

An article by Colloc'h et al., in this issue, places one more piece of the puzzle by reporting the first observation of nitrous oxide (N_2O) bound in x-ray structures of proteins relevant to possible cellular targets (3). This study compares nitrous oxide and xenon binding in a putative model of the NMDA receptor and in a globular protein. One site in each protein is available to either gas, whereas a second molecule of nitrous oxide also binds to each protein. Since the relative anesthetic potencies of these gases have been determined previously, structural results and function can be compared. AnnexinV has properties of ligand (calcium) gating that make it a reasonable model of the extracellular part of NMDA receptor function, whereas urate oxidase is taken to represent intracellular proteins with large hydrophobic cavities. The annexinV structure was solved in the presence of calcium. In this state, a tryptophan side chain is displaced away from the core of the structure, leaving the cavity to which both gases bind.

The interaction of xenon with protein cavities can be examined experimentally by methods of x-ray diffraction, illustrated in Colloc'h et al. (3) and reference 27 in that article and by NMR (4). The nature of xenon-cavity interactions can be approached with two types of NMR experiment. NMR chemical shifts (^1H-^{15}N correlation spectroscopy) of side chains lining protein cavities titrate with xenon overpressure, and the ^{129}Xe chemical shifts titrate at constant xenon pressure with variation in protein concentration (4). In either case, xenon binds to known protein cavities with weak affinities of $\sim$10–200 M^{-1} ((4) and references therein). The volumes of some, but not all, cavities expand when xenon is bound (3), and xenon is able to displace other small ligands from cavities (4). Colloc'h et al. find that xenon binding expands the annexinV cavity left vacant by Trp displacement, and likewise the other cavities studied. They suggest that binding of anesthetics of the xenon/nitrous oxide class, by expanding cavity volume, may reduce some feature of protein flexibility that is necessary for function. This general concept is a good basis for design of future experiments, but most puzzling is how it can lead to specificity for a particular neurotransmitter target.

One place to look for the next piece in the puzzle is the membrane portion of neurotransmitter receptors. Recent studies of channels that transport gas molecules may point the way. The Rh factors of human red blood cells are thought to facilitate entry of carbon dioxide gas and these membrane proteins are structurally related to the ammonia family of transporters (the Amts) (5). The x-ray structure of one Amt in fact reveals 15 sites per monomer for xenon binding (6). A site occupied by two of the xenon atoms is a large, hydrophobic cavity on the cytoplasmic side of Amt-1. It is suggested that this cavity in the Amt may be required for conformational changes related to function (6), and in that sense, arguments could be advanced that cavity expansion by xenon occupation might inhibit Amt-1 function, as is argued for annexinV (3). Even more intriguing is that three strong xenon sites are found in the Amt-1 transport channel, and there are others between monomers in the trimer. From other examples, xenon binding is not limited to hydrophobic sites. It seems possible then that the effect of xenon and other anesthetics on neurotransmitters might involve the channel itself. This returns the discussion to the original study implicating NMDA receptors as the target of xenon anesthesia. Xenon inhibits by 60% the maximum inward current through NMDA receptors in cultured neurons (reference 9 in Colloc'h et al. (3)) but does not compete with NMDA. Conformational flexibility in the ligand-binding core of NMDA receptors is understood at the structural level (7). Revealing xenon/nitrous oxide sites in both the ligand-binding portion and the channel itself could bring us closer to a general theory of inhaled anesthetic action and to a general state of euphoria.

REFERENCES

1. Peltier, S., C. Kerssens, S. B. Hamann, P. S. Sebel, M. Byas-Smith, and X. Hu. 2005. Functional connectivity changes with concentration of sevoflurane anesthesia. *Brain Imaging*. 16:285–288.

Submitted September 8, 2006, and accepted for publication September 15, 2006.

Address reprint requests to Betty J. Gaffney, Tel.: 850-644-8547; E-mail: gaffney@bio.fsu.edu.

© 2007 by the Biophysical Society

0006-3495/07/01/1/02 $2.00

doi: 10.1529/biophysj.106.096503

2. Sanders, R. D., D. Ma, and M. Maze. 2005. Xenon: elemental anesthesia in clinical practice. *Br. Med. Bull.* 71:115–135.

3. Colloc'h, N., J. S. Santos, P. Retailleau, M. Lemaire, T. Prangé, and J. H. Abraini. 2007. Protein crystallography under xenon and nitrous oxide pressure: Comparison with in vivo pharmacology studies and implications for the mechanism of inhaled anesthetic action. *Biophys. J.* 92:217–224.

4. Lowery, T. J., M. Doucleff, E. J. Ruiz, S. M. Rubin, A. Pines, and D. E. Wemmer. 2005. Distinguishing multiple chemotaxis Y protein conformations with laser-polarized ^{129}Xe NMR. *Protein Sci.* 14:848–855.

5. Khademi, S., J. O'Connell 3rd, J. Remis, Y. Robles-Colmenares, L. J. W. Miercke, and R. M. Stroud. 2004. Mechanism of ammonia transport by Amt/MEP/Rh: structure of AmtB at 1.35 Å. *Science.* 305:1587–1594.

6. Andrade, S. L. A., A. Dickmanns, R. Ficner, and O. Einsle. 2005. Crystal structure of the archaeal ammonium transporter Amt-1 from *Archaeoglobus fulgidus*. *Proc. Natl. Acad. Sci. U.S.* 102:14994–14999.

7. Furukawa, H., and E. Gouaux. 2003. Mechanisms of activation, inhibition and specificity: crystal structures of the NMDA receptor NR1 ligand-binding core. *EMBO J.* 22:2873–2885.