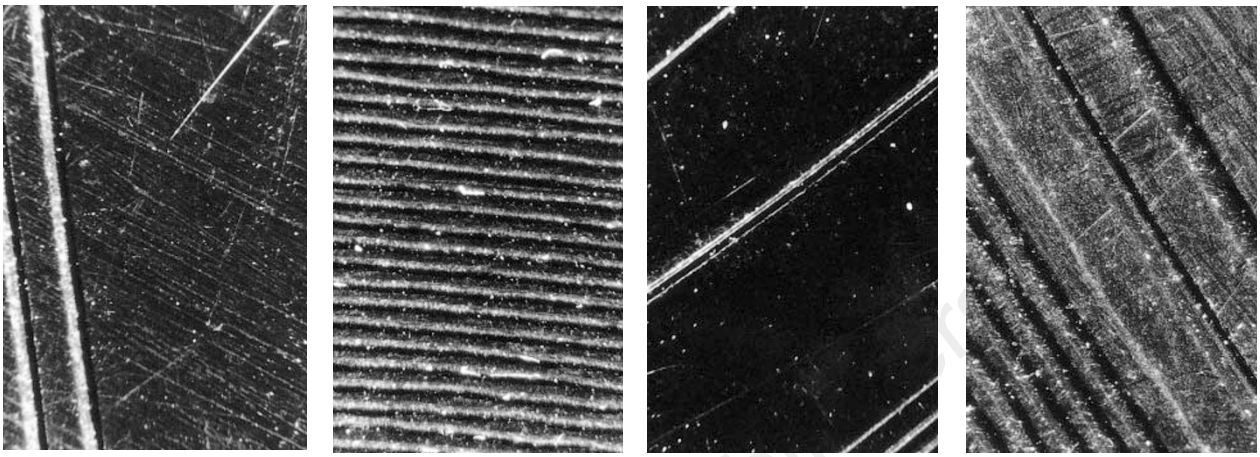
Avoided objects

Nutcracker suite

Grooves in a record that belonged to Hitler



main sites of general-anaesthetic action. At inhibitory synapses (Fig. 1), neurotransmitter — GABA (γ-aminobutyric acid) or glycine — is released from vesicles at presynaptic terminals in response to the voltage-dependent entry of Ca^{2+}, which is triggered by an action potential. The neurotransmitter diffuses to the postsynaptic membrane, and hyperpolarizes the membrane by opening inhibitory, chloride-permeable $GABA_A$ or glycine receptor channels. These channels belong to an anaesthetic-sensitive superfamily of fast, neurotransmitter-gated receptor channels, which also includes the excitatory (Na^+- and K^+-permeable) 5-hydroxytryptamine (5-HT_3) and nicotinic acetylcholine (nACh) receptor channels[6]. Volatile anaesthetics inhibit the nACh receptors, but they markedly potentiate the activities of the other three types of receptor[7,8].

Using molecular genetics, binding sites for volatile anaesthetics that act on nACh receptors have been identified. Site-directed mutagenesis has revealed an inhibitory site within the aqueous pore of the muscle-type nACh receptor[9]. A second inhibitory site has been assigned to the large, neurotransmitter-binding, amino-terminal extracellular domain of another nACh receptor (α7). This was based on the behaviour of a homomeric receptor channel formed from chimaeric subunits — each subunit consisting of an amino-terminal domain from the nACh (α7) receptor, and a transmembrane and carboxy-terminal domain from the 5-HT_3 receptor[10].

Mihic *et al.*[5] have now identified a potentiating site near the extracellular regions of transmembrane domains 2 and 3 on the glycine and $GABA_A$ receptors. They used an extensive set of well-designed chimaeras, consisting of complementary parts of a glycine receptor and a GABA ρ1 receptor. Also known as $GABA_C$, the GABA ρ1 receptor is related to $GABA_A$, but it is inhibited — rather than potentiated — by volatile anaesthetics. From the behaviour of the chimaeric constructs, the authors deduced that a 45-amino-acid region was involved in potentiation by volatile anaesthetics and ethanol. By systematically mutating these amino acids, they identified two residues that were critical for anaesthetic potentiation. Interestingly, one of these also determines the potentiating effect of etomidate[11] (a clinically used intravenous general anaesthetic) on $GABA_A$ receptors. But neither residue seems to be involved in the action of propofol, which is another intravenous agent.

Are these critical residues part of an anaesthetic-binding site, or are they involved only in gating or allosteric linkage? The answer is not known, and the issue is unlikely to be definitively resolved using molecular genetics. In some cases, such as the inhibitory pore site for anaesthetics on the muscle-type nACh receptor, an anaesthetic-binding site is probably involved, because the kinetics of channel inhibition are entirely consistent with the proposed site within the channel pore[9]. But in the case of potentiation — which is less well understood than inhibition — clear-cut interpretation is more problematic. Structural studies should help, although sufficiently high-resolution images of ion channels are still some way off. Yet only when the three-dimensional structures of anaesthetic binding sites on channels are known can we even begin to think in terms of developing 'designer' general anaesthetics. These would bind specifically to selected ion channels, to produce general anaesthesia with fewer of the undesirable side-effects that are associated with currently used agents.

Meanwhile, the use of molecular genetics to identify the amino acids involved in anaesthetic modulation of channel activities will be encouraged by the success of Mihic *et al.* The technique will undoubtedly be extended to other protein targets and classes of anaesthetic. But a broader problem remains. How do we extrapolate from specific sites on ion channels, at the molecular level, to the effects of anaesthetics on the intact animal? For example, how can we be sure that, even though the $GABA_A$ receptor is sensitive to anaesthetics *in vitro*, these effects are essential to — or even contribute to — the state of general anaesthesia itself? Perhaps the most exciting aspect of these new findings is the prospect (suggested by Mihic and colleagues) of using transgenic-animal technology to express anaesthetic-insensitive mutant channels in mice. This would act as a selective test for the relevance and importance of various ion channels to the process of general anaesthesia. □

N. P. Franks and W. R. Lieb are in the Biophysics Section, The Blackett Laboratory, Imperial College of Science, Technology and Medicine, London SW7 2BZ, UK.

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