TITLE: The Retina As A Testbed For Circuit-level Mechanisms Of General Anesthesia

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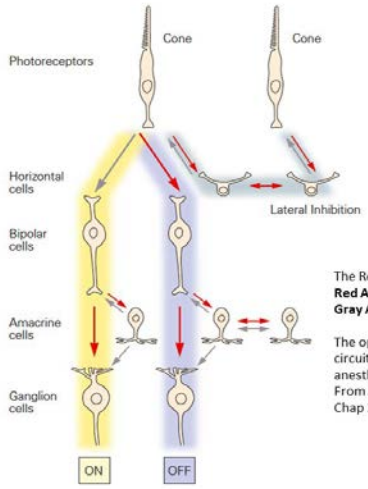
ABSTRACT: General anesthetics have been widely used since the 19th century and are an essential part of medicine. However, open questions still remain about the biological mechanisms of general anesthesia. Macroscopic approaches to the nervous system, such as human brain imaging, are limited to reading out the mean activity of millions of neurons over seconds. Microscale approaches that focus on ion channels are difficult to connect to the systems-level phenomena of anesthesia. As a complementary approach one might want a mesoscale experimental model that reveals the dynamics of neuronal circuits while retaining interpretability at the molecular level. In this spirit, we are developing the mouse retina as a mesoscopic testbed for anesthesiology.

The retina is an accessible part of the CNS in the back of the eye. It contains ~100 different types of neuron that express most of the known neurotransmitters and receptors, including the most common molecular targets of modern anesthesiology theories, such as GABAa and NMDA receptors. These neurons form a family of neural circuits that implement the signal flow from photoreceptors to retinal ganglion cells. Most importantly for the present application, the function of these circuits is exquisitely well understood, such that one may use it for reverse-engineering the effects of drugs. By observing the circuit-level effects of a substance one can draw conclusions about the underlying molecular and cellular mechanisms. By using multiple electrode arrays, we can densely read out from the output layer of the isolated retina at a resolution of single neurons and single action potentials.

In ongoing experiments we superfused the retina with general anesthetics (Ketamine, Alcohols, Isoflurane) while monitoring the dynamics of the light response in retinal ganglion cells (RGCs). Each of these substances was able to silence retinal responses reversibly. This occurred close to each alcohol's (ethanol, butanol, hexanol) Meyer-Overton concentration, suggesting that the effects of general anesthesia are preserved in the retina. However, at intermediate concentrations a surprising effect occurred: About half of the RGCs were excited and the other half inhibited. It turned out that the excited half of the retina are "ON" circuits - which are excited by light - and the inhibited half are "OFF" circuits - inhibited by light. Thus low-dose anesthetics act on the retina like a background light. For comparison we tested drugs that act on the GABAa receptor (muscimol) and the NMDA receptor (D-AP5). Neither of these substances recapitulated the opponent effects on ON and OFF circuits that we observed with general anesthetics, suggesting that a different pathway is involved. An inspection of retinal circuits (Fig 1) suggests that the opponent effects arise prior to the bipolar cell. Thus we are now focusing on cellular mechanisms in photoreceptors and horizontal cells. In summary it appears that using an in vitro neuronal system whose structure and function are well known one can gain mechanistic insights that are complementary to those from microscopic/macroscopic model systems. This opens opportunities for circuit neuroscientists to join forces with researchers in anesthesiology in pursuing the century-old mystery of general anesthesia.

Fig 1



The Retinal Circuitry.
Red Arrows: Sign preserving connections
Gray Arrows: Sign inverting connections

The opposite polarity of the ON vs OFF circuits suggests the opponent effect of anesthetics arise prior to the bipolar cells. From *Principal of Neural Science*, 5th Edition Chap 26