Lipid Protein Interactions in Mitochondria. VIII. Effect of General Anesthetics on the Mobility of Spin Labels in Lipid Vesticles and Mitochondrial Membranes

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Abstract

We have studied the effect of general anesthetics on the mobility of two stearic acid spin labels (5-doxyl stearic acid and 16-doxyl stearic acid) in bovine heart mitochondria and in phospholipid vesicles made from either mitochondrial lipids or commercial soybean phospholipids. The general anesthetics used include nonpolar compounds (alcohols, halothane, pentrane, diethyl ether, chloroform) and the amphipathic compound, ketamine. All anesthetics tested increase the mobility of the spin labels in phospholipid vesicles to a limited extent up to a concentration where the ESR spectra become those of free spin labels. On the other hand, anesthetics have a pronounced effect on mitochondrial membranes at concentrations as low as those known to produce general anesthesia; the effect is lower near the bilayer surface (5-doxyl stearic acid) and very strong in the bilayer core (16-doxyl stearic acid). The effects of anesthetics are mimicked by the detergent, Triton X-100. We suggest that the discrepancy between the action of anesthetics in mobilizing the spin labels in lipid vesicles and in membranes results from labilization of lipid protein interactions.

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Introduction

During the course of our investigations on lipid protein interactions in biomembranes (1–5), we have studied the effect of the series of n-alkanols on membrane lipid fluidity, probed by stearic acid spin labels and by the fluorescent probe ANS. We have found that alcohols enhance the fluidity of membrane lipids; in particular, the spin label studies have given strong indications that alcohols disrupt lipid protein interactions (5), since they appear to abolish the immobilization induced by intrinsic membrane proteins on the lipid bilayer.

A consequence of this effect was the finding that alcohols induce kinetic changes in the mitochondrial ATPase, which have been attributed to changes in membrane lipids (4, 6, 7).

Alcohols are usually included among general anesthetics (8); we have therefore considered it of interest to investigate whether compounds belonging to the class of clinically useful anesthetics induce the same or similar changes as those induced by alcohols. The Meyer rule of anesthesia (9) states that narcosis is the result of attaining a certain molar concentration of any chemically inert substance in the cellular lipids; according to Hill (10), this will increase the entropy of the system, and anesthesia is the result of this entropy increase.

It has been found that anesthetics increase the fluidity of model lipid membranes (8, 11, 12); it is therefore plausible that anesthesia is the result of a change in lipid fluidity of neuronal membranes involved in the transmission of nerve impulses (13).

It is well known, however, that anesthetics act on all membranes (8) although local quantitative differences may be present. For this reason it is customary to investigate physical changes induced by anesthetics on several non-neuronal membranes, e.g., erythrocyte ghosts. In such membranes, expansion by anesthetics has been directly related to a fluidity increase.

We have advanced a working hypothesis (13), based on the results obtained with n-alkanols, that anesthetics change the physical state of membrane lipids leading to an alteration in normal lipid protein interactions, which in turn induces conformational changes in membrane proteins. Such changes in neuronal membranes may be directly related to the mechanism of anesthesia. A similar model has been postulated by Lee (14) for the action of local anesthetics at the level of the sodium channel in nerve membranes. There are data in the literature that anesthetics affect lipid fluidity; such data are obtained mainly on lipid bilayer vesicles or oriented multibilayers (11, 12, 15). Few data are available on natural membranes, and there are indirect indications that natural membranes may have a quantitatively different