Possible Relationships of Alcohol in Membranes to Cancer

Gerhard Freund

Veterans Administration Medical Center and Departments of Medicine and Neuroscience, College of Medicine, University of Florida, Gainesville, Florida 32601

Abstract

Ethanol can be used as a chemical tool to alter membrane fluidity or composition, or both, and to study the effects on induction, growth, spread, or treatment of cancers. Ethanol rapidly equilibrates with total body water and enters all cell membranes. Ethanol molecules are intercalated between the lipids of the bilayer membranes. This expands membranes and increases their fluidity, which in turn affects cell agglutination, phagocytosis, membrane transport, membrane enzyme activities, and many other membrane functions. After 3 to 5 days of continuous ethanol administration, the original membrane fluidity is restored by the incorporation of "stiffening" lipids, such as cholesterol, into the bilayer and by the increase of the chain length and saturation of fatty acids. The desired membrane effects (increased fluidity or altered membrane composition) can be obtained by adjusting time-dose relationships of ethanol administration.

There may be an important role of moderate alcohol consumption in cancer biology that is not presently recognized by epidemiological studies because both cancers and moderate alcohol consumption are very prevalent in the general adult population. Moderate, social alcohol use could potentially either suppress or enhance the induction, growth, spread, or therapy of cancers. Such potential roles of alcohol in cancer biology could easily be tested in animals by incorporating the feeding of alcohol-containing diets into experiments that follow standard cancer protocols.

Introduction

The acute and chronic administration of ethanol induces biological changes at all levels of biological organization, ranging from the molecular to the behavioral. Consequently, interactions between cancer and alcohol may occur at all of these levels. As a sedative drug, alcohol may have a use in cancer therapy. As a drug causing irritation and cell necrosis in the esophagus and liver, alcohol may contribute to the cause of some cancers. Most intriguing, however, are the potential relationships between cancer and ethanol at the membrane level because ethanol rapidly enters, expands, and fluidizes all biological membranes (15, 30, 31). These changes in membrane properties could suppress or enhance the induction, growth, spread, or treatment of cancers. It is proposed here to use ethanol as a research tool with powerful effects on biological membranes to study cancer-membrane interactions.

These membrane changes are ubiquitous, but they are most obviously and easily studied by their effects on the CNS. The manifestations of intoxication and physical dependence are behavioral markers of the generalized membrane changes induced by alcohol. They indicate that ethanol-membrane interactions are not static but are changing with time and dose. For instance, the development of physical dependence and tolerance are accompanied by generalized, reversible membrane changes that progress over a period of days or weeks. The purpose of this paper is to elaborate on these alterations of membranes and to discuss their possible relationships to cancer biology.

Molecular Mechanisms of Ethanol Action

Ethanol may exert its biological effects principally by 5 different mechanisms: physical intercalation of ethanol molecules in membranes; formation of chemically reactive ethanol metabolites like acetaldehyde; change of oxidation-reduction equilibria during ethanol metabolism in the liver; induction of enhanced liver enzyme activity; and facilitation of free radical formation.

In addition to its initial physical effects on membranes, ethanol metabolism may exert chemical effects on the tissues. The liver is the only tissue that metabolizes ethanol to any significant extent. The local formation of ethanol metabolites, such as acetate and acetaldehyde, and secondary shifts in the oxidation-reduction equilibrium (increased NADH:NAD⁺ ratio) are probably unique to the liver. They may explain the great vulnerability of the liver to chronic ethanol consumption. These effects of ethanol are discussed in detail elsewhere in this symposium (21). Reviews of the potential role of free radicals are available (4, 12, 25).

Ethanol rapidly equilibrates with the total body water and enters all membranes and even ischemic tissues. There ethanol must exert its effects by means of its physical presence in the membranes, because there are no specific drug receptors for ethanol and because ethanol is not metabolized except in the liver. Therefore, although the critical organ in mammals is the nervous system, the same ethanol-induced cellular changes occur in all cells throughout the body.

Acute Effects of Ethanol on Membranes

The intercalation of ethanol molecules between the fatty acids of membrane phospholipids increases membrane fluidity and expands the membranes, with resulting narrowing of sodium channels and changes in calcium binding to membranes (1, 3, 5, 28, 30, 31). Other lipid-soluble anesthetics besides ethanol cause increased membrane fluidity and expansion. The same changes are caused by shortening and desaturation of the fatty acid chains, a decreased cholesterol-phospholipid ratio, increased temperature, and decreased hydrostatic pressure (2, 31). Both direct effects of ethanol on membrane proteins and indirect effects mediated by lipid-protein interaction have been postulated (34). It is controversial which of the
several molecular membrane correlates of ethanol intoxication are direct proximate causes of CNS depression and which are unrelated covariates or secondary effects of intoxication. A cascade of events involving transport, enzymes, and neurotransmitters follows the primary physical membrane action. Each may or may not be relevant to the induction of intoxication, physical dependence, and tolerance.

The evidence in favor of postulating that the physical effects of ethanol on membranes are the direct causes of CNS depression may be summarized as follows. (a) The depth of anesthesia is quantitatively related to lipid solubility of anesthetics and their concentrations in membrane lipids (30). (b) Anesthetics, including alcohols, alter membranes. The fact that many agents of widely different chemical structures have anesthetic and certain membrane actions in common suggests that these common physical membrane actions, rather than diverse specific chemical reactions, cause anesthesia (1, 15, 16, 24, 30). (c) Chemically inert gases may cause anesthesia and CNS depression (15, 23, 24). (d) Metabolically inert chemicals such as bromide (8) and tert-butyl alcohol (33, 36) may cause CNS depression and physical dependence. (e) Drugs of widely varying chemical structures, including alcohols, show cross-tolerance and cross-dependence (19). (f) Increased hydrostatic pressure reverses anesthesia by various agents, including ethanol (14, 15, 17, 18, 23). (g) The manifestations of experimentally induced physical dependence on ethanol are suppressible by systemic treatment with local anesthetic drugs acting on membranes, such as lidocaine (9) and d-propranolol (11). (h) Indirect in vitro evidence. Ethanol consumption increases membrane fluidity. After 3 to 5 days, a compensatory stiffening of membranes develops, as measured by a resistance to the effects of in vitro ethanol on spin-label fluidity (1) and on miniature endplate potentials (3). Single-dose ethanol administration in rats depletes brain membrane calcium and increases in vitro calcium binding to synaptosomes. The opposite changes occur after the rats have been drinking aqueous ethanol solutions for 2 weeks (28). Intraventricular calcium increases sleeping time and behavioral intoxication in mice (5).

**Chronic Effects of Ethanol on Membranes**

Ethanol is intercalated between the membrane lipid bilayer molecules and thereby expands membranes and increases fluidity. One would anticipate that prolonged exposure causes compensatory “stiffening” of membranes to restore the original state of fluidity. This stiffening theoretically could involve an increase in membrane concentrations of longer-chain-length fatty acids and a decrease of those that are shorter, and it could also cause a greater saturation of double bonds (2, 31). The membrane cholesterol:phospholipid and sphingomyelin:lecithin ratios should also increase. An increase of membrane cholesterol in mice exposed to ethanol has recently been reported (22). It appears then that membrane fluidity may be preserved at the expense of membrane composition. At the neurophysiological and behavioral levels of biological organization, this molecular adaptation could be the basis of tolerance to the effects of ethanol and of physical dependence after sudden removal of alcohol. As previously discussed, after mice consume ethanol for 3 to 5 days, their membranes become resistant to the fluidizing effects of ethanol in vitro (1). This time course of several days corresponds roughly to the time necessary to induce physical dependence in mice (6, 10). Membrane changes and overt evidence of ethanol withdrawal apparently disappear within 24 hr.

Although the above “chronic” effects are reversible, the administration of ethanol to rodents over a period of 3 to 7 months causes irreversible changes in the CNS (7, 26, 35). It is presently unknown what mechanisms are responsible for transforming reversible changes to permanent ones.

**Administration of Ethanol as a Membrane Modulator**

Depending on the objective of the membrane manipulation, different membrane changes could be obtained by adjusting dose-time relationships. For instance, short, small-dose pulses of ethanol may increase membrane fluidity without compensatory changes in membrane composition. Alternatively, various degrees and duration of membrane stiffening may be desirable for particular experiments involving the induction, growth, spread, or therapy of cancer. The development of acute intoxication and physical dependence on ethanol can be prevented by appropriate dose-time manipulations to include periods of abstinence and gradual rather than sudden cessation of ethanol administration. Finally, ethanol could be combined with other membrane-active agents, including lipid-soluble local anesthetics, administered systemically, such as d-propranolol (11).

The induction of physical dependence is a specific property confined to few chemicals. Most nonaddicting drugs follow classic dose-response relationships. The more drug present, the greater is the biological effect of the drug. In contrast, physically addicting drugs cause some of their effects after drug administration is ceased. The manifestations of withdrawal illness increase with time after the drug is removed from the body until they finally subside. The time course of the manifestations of physical dependence on ethanol in humans is shown in Chart 1. The time course and severity vary among individuals and in recurrent episodes in the same person. It is generally accepted that, to induce physical dependence, these drugs must be present continuously in a minimum amount and for a certain minimum period of time. If the conditions are appropriate, evidence of physical dependence and pathological features after drug withdrawal are demonstrable in virtually all mammalian species tested (10) and even in isolated cell or tissue cultures (13, 32).

**Moderate Alcohol Consumption and Carcinogenesis**

Traditionally, the emphasis has been on the relationship

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Drinking

<table>
<thead>
<tr>
<th>Disorientation (Delirium)</th>
<th>Hallucinations (Visual, Auditory, Tactile)</th>
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<tbody>
<tr>
<td>Tremors, Psychomotor hyperactivity, Seizures</td>
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Chart 1. Time course of the manifestations of physical dependence on ethanol in humans.
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between cancer and heavy alcohol consumption or alcohol abuse. Epidemiological evidence indicates an association between the consumption of large amounts of alcohol and cancers in those areas of the gastrointestinal tract that are exposed either directly to concentrated ethanol solutions (esophagus) or where most of the ethanol is metabolized (liver). However, moderate or slight alcohol consumption possibly has a role in carcinogenesis, cancer spread, or therapy. Moderate alcohol consumption is much more common in our society than alcohol abuse. Even the consumption of large amounts of alcohol and canorate alcohol consumption are so prevalent, possible relationships between the two may be impossible to detect with epidemiological methods. Because of the ubiquitous presence of ethanol in all tissues of the body, cancers originating everywhere could be either promoted or suppressed.

It is impossible at present to obtain epidemiological evidence for such relationships between moderate alcohol consumption and carcinogenesis in humans. To my knowledge, no animal studies in vivo or in vitro have been addressed to these problems. However, various alcohol feeding regimens could easily be incorporated into experimental protocols dealing with carcinogenesis and the growth, spread, and therapy of cancer in rodents. Such alcohol feeding regimens should probably use 2 different types of liquid diets, those developed by Lieber et al. (20, 29) that cause liver damage and those recommended by Freund (6, 10) that do not damage the liver. Various mechanisms may be envisioned whereby changes in membrane fluidity could affect cancer induction, growth, spread, and therapy. For instance, membrane rigidity in the aortic media is known to increase with aging or atherosclerosis, as manifested by an increased sphingomyelin:lecithin ratio (2). Could increased membrane rigidity have something to do with increased carcinogenesis in aging? Decreased plasma membrane fluidity by increased cholesterol content results in increased cell aggregation, decreased phagocytosis by macrophages, decreased carrier-mediated transport, and alterations of membrane enzyme activities and membrane receptor functions (2).

Changes in membrane fluidity induced by ethanol would not be confined to those cells but could affect the membranous envelopes of carcinogenic viruses. For instance, lymphoid cells have been shown to have a much greater cell membrane fluidity than do infecting viruses. This higher fluidity favors the translocation of fluorescent probes from virus to lymphocyte, an early event associated with the infection of the cell (27). Ethanol, in the physiological concentrations found in human blood during alcohol ingestion, has a definite effect on lymphocyte proliferation in vitro (13). Thus, it seems entirely possible that even very short-term alterations of plasma membrane fluidity by alcohol could have profound effects in viral carcinogenesis.

References

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