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ALCOHOL AND CARCINOGENESIS

Helmut K. Seitz and Ulrich A. Simanowski

Alcohol Research Laboratory, Department of Medicine, University of Heidelberg, Heidelberg, Federal Republic of Germany

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INTRODUCTION

Ethanol must be considered one of the most important toxins consumed regularly and in large quantities by humans. Since alcohol intake has steadily increased during the last two decades, alcoholism has become one of the major health problems worldwide. Heavy alcohol consumption exerts deleterious effects on almost every organ of the human body. The most important ethanol-related diseases are cirrhosis of the liver, pancreatitis, cardiomyopathy, hematologic, neurologic, and psychiatric disorders, tuberculosis, and tumor development. This chapter, however, focuses on the effect of

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ethanol on carcinogenesis. Epidemiologic and experimental evidence is presented supporting the cocarcinogenic action of alcohol. In addition, various pathogenetic mechanisms by which ethanol may enhance carcinogenesis are discussed.

EPIDEMIOLOGY OF ALCOHOL AND CANCER

Clinically, a link between alcoholism and certain types of cancer has been observed for many years. In France at the beginning of this century, Lamu (55) had already showed a highly significant association between esophageal cancer and the consumption of absinth, a concentrated herbal liquor. Meanwhile a great number of epidemiologic studies showed an association between excessive drinking of alcoholic beverages and cancer of the oropharynx (20, 30, 48, 99, 131, 138), the larynx (31, 39, 76, 104, 131, 140, 142), and the esophagus (103, 104, 127–129, 141). In a series of studies (138) heavy drinkers were found to have roughly a 10-fold increased risk of developing cancer of the mouth, mainly because of consuming hard liquors.

Subjects who drink heavily often smoke heavily also. This fact was first taken into account by Flamant et al (22), who assessed both factors and reported that there was a "strong" association of alcohol intake with cancer of those sites that come most directly in contact with alcohol (tongue, hypopharynx, larynx, esophagus). The risk of developing oral cancer for heavy drinkers who smoke was 6–15 times higher than for nondrinkers and nonsmokers (99). It appears that alcohol plays a more important role than smoking with respect to cancer of the esophagus, whereas smoking seems to be more strongly associated with cancer of the mouth and pharynx (22). Of these cancers 76% could be eliminated in males if exposure to alcohol and tobacco were avoided (99). Both factors, alcohol and smoking, also have a synergistic effect on the carcinogenesis in the upper alimentary tract. In a study from Brittany, Tuyns (127–129) demonstrated clearly that alcohol intake of more than 80 g per day increases the risk of esophageal cancer by a factor of 18; smoking more than 20 cigarettes per day increases the risk of developing esophageal cancer by a factor of 5. Alcohol drinking and smoking together enhance the risk for this cancer by a factor of 44!

Recently additional sites of cancer associated with alcohol were detected in the liver (2, 35, 37, 48, 122, 145), the rectum (14, 18, 51, 77, 92, 94, 136), the pancreas (16, 40), the cardia of the stomach (74), the lungs (39, 92), and the breast (41, 56, 57, 98, 101, 135).

Hepatocellular carcinoma (HCC) in alcoholics is commonly thought to be associated with cirrhosis of the liver. Indeed, the incidence of cirrhosis in patients with HCC has been reported to vary between 16 and 80% (63, 64), with most reports indicating a 55–80% association. It has been suggested that viral hepatitis is also more common in alcoholics than in corresponding

nonalcoholic populations and thereby could contribute to the increased incidence of HCC. This is discussed below in detail. Further evidence for the cocarcinogenic role of alcohol in human hepatocarcinogenesis may derive from epidemiologic studies on aflatoxin exposure. It was calculated that the daily consumption of 24 g of ethanol or more increases the risk of developing HCC induced by 4 μg of dietary aflatoxin B_1 by a factor of 35 (8).

In addition, Gottfried et al (29) reported a case of histologically proven HCC in an alcoholic. The tumor vanished spontaneously following abstinence from alcohol. Furthermore, an unusual paired human accident demonstrated the cocarcinogenic role of alcohol in two chemical workers working with vinyl chloride. One worker consumed large quantities of ethanol in addition to his exposure to vinyl chloride and developed both angiosarcoma and HCC, while his colleague, a nondrinker, developed only angiosarcoma of the liver (122).

Most recently epidemiologic studies demonstrated an association between alcohol ingestion (mainly beer) and the occurrence of rectal but not colonic cancer. An Irish brewery study demonstrated that the risk of developing rectal cancer is almost double in beer drinkers (14), but a similar study in Denmark failed to show such an association (46). The only prospective study, from Hawaii, included more than 8000 subjects observed over more than 12 years; it found a significant correlation between beer consumption (starting at a daily dose of 500 ml of beer) and the occurrence of rectal tumors (92).

With respect to pancreatic cancer, controversial results have been published. While most recently Mack et al (66) could not demonstrate any effect of ethanol on the occurrence of this tumor, Heuch et al (40) from Norway showed in a prospective study with more than 16,000 subjects a strong positive association between frequent abuse of ethanol and the development of pancreatic cancer. The relative risk in the alcohol abusers was 5.4. The results of this prospective study are in accordance with the data obtained from earlier retrospective investigations.

Several epidemiologic studies have also shown a correlation between moderate drinking of alcoholic beverages and breast cancer. Some of these studies have been questioned because they were performed retrospectively (56, 57, 98). However, three recently published prospective studies give further evidence that moderate alcohol consumption (starting with a daily dose of 15 g of ethanol) is associated with an approximately 50 to 100% increase of risk for breast cancer (41, 101, 135).

EFFECT OF ETHANOL IN EXPERIMENTAL CARCINOGENESIS

Ethanol per se is not a carcinogen (49). However, when administered in combination with a chemical carcinogen, ethanol enhances carcinogenesis in

some organs. Depending on the experimental conditions, ethanol may exert a cocarcinogenic effect, especially when given before or together with the carcinogen, or it may act as a tumor promoter when administered after tumor initiation.

Local Effects

When polycyclic hydrocarbons such as benzo(a)pyrene (BP) or dimethylbenzanthracene (DMBA) were applied locally to the esophagus of mice or to the pouch and skin of mice and hamsters, their carcinogenic power was found to be significantly enhanced if ethanol was used as the solvent (17, 38, 43, 119). In these experiments ethanol most likely acted as a tumor promoter by irritating the mucosa.

Hepatic Carcinogenesis

Table 1 summarizes the effects of ethanol on chemically induced experimental hepatic cancer. The majority of these experiments were performed with nitrosamines as tumor inducers; only a few other procarcinogens have been studied. Hepatic carcinogenesis with nitrosamines was only enhanced when ethanol was given during promotion or when a methyl-deficient diet was administered simultaneously (15, 93, 121). However, when alcohol was applied prior to or together with the carcinogen, carcinogenesis was not affected or even inhibited. The possible mechanisms for this observation are discussed below.

Radike et al (97) found a fourfold increase in vinyl-chloride-induced hepatic cancer as well as a change in the cell type of the tumors after chronic ethanol consumption in the rat. This was associated with severe mitochondrial damage due to the combined effect of vinyl chloride and ethanol (81). The finding seems of particular importance because of the case report (122) mentioned above.

When aflatoxin B_1 was used for tumor induction, no effect of ethanol on hepatic carcinogenesis was noted. However, a significant increase in the occurrence of hepatic peliosis was observed in the rat (79).

Extrahepatic Carcinogenesis

Research on experimental extrahepatic carcinogenesis has focused on three target organs, the upper alimentary and respiratory tract, the rectum, and the pancreas (Table 2). Again, the results obtained depend mainly on the experimental design. This is especially true with respect to the carcinogen used for tumor induction and to the method of alcohol application (110). Among all these factors, the administration of ethanol in a liquid diet, first introduced by Lieber and DeCarli (62, 107, 111), is certainly of great importance. Only this feeding technique guarantees an adequate ethanol intake and takes care of

Table 1 Effect of ethanol on chemically induced hepatic carcinogenesis

Authors	Species	Carcinogen[a]	Ethanol administration[b]	Ethanol effect
Griciute et al (32, 33)	Mouse/Rat	DMNA, i.g./NNN, i.g.	40% i.g. with carcinogen	Inhibition[c]
Habs & Schmähl (34)	Rat	DMNA, orally	25% in d.w. after carcinogen	Inhibition
Teschke et al (123)	Rat	DMNA, i.p.	6% l.d. prior to carcinogen	No effect/inhibition[d]
Gibel (27)	Rat	DENA, i.g.	30% i.g. with carcinogen	No effect[e]
Porta et al (93)	Rat	DENA, i.p.	25–32% in l.d. and methyl deficiency after carcinogen	Stimulation
Takada et al (121)	Rat	DENA, i.p., 70% hepatectomy	20% in l.d. after carcinogen	Stimulation
Driver & McLean (15)	Rat	DENA, i.p.	5% in d.w. after carcinogen	Stimulation
Mendenhall & Chedid (79)	Rat	Aflatoxin B_1, i.g.	6% l.d.continuously	No effect[f]
Misslbeck et al (83)	Rat	Aflatoxin B_1, i.g.	6% l.d. after carcinogen	No effect
Radike et al (97)	Rat	Vinyl chloride, in air	5% in d.w. prior to and with carcinogen	Stimulation
Weisburger et al (132)	Rat	N-OH-2AAF, orally	10% in d.w. with carcinogen	No effect

[a] DMNA = dimethylnitrosamine; DENA = diethylnitrosamine; NNN = *N*-nitrosonornicotine; N-OH-2AAF = *N*-hydroxy-2-acetaminofluorene; i.g. = intragastrically; i.p. = intraperitoneally.
[b] d.w. = drinking water; l.d. = liquid diet.
[c] Occurrence of olfactoric neuroepithelioma after alcohol.
[d] Similar tumor yield, but prolonged latency period after alcohol.
[e] Enhanced esophageal cancer after alcohol.
[f] Stimulation of hepatic peliosis after alcohol.

nutritional factors. If ethanol is given in the drinking water, alcohol intake may be extremely low. In addition, nutritional deficiencies may occur, which by itself can influence carcinogenesis.

Using the liquid diet feeding technique or intragastric application of ethanol, researchers have found an enhancement of nitrosamine-induced carcinogenesis in the esophagus and in the upper respiratory tract (Table 2). The inhibition of nitrosamine-induced hepatocarcinogenesis and the concomitant stimulation of extrahepatic tumor development by alcohol may be due to a complex interaction between ethanol and nitrosamine metabolism, which is discussed below. It is noteworthy that alcohol consumption in combination with dimethylnitrosamine (DMN) treatment led to the development of olfactoric neuroepithelioma, tumors otherwise never reported (32, 33).

Liver function also seems important in extrahepatic carcinogenesis. Mice that received either ethanol or carbon tetrachloride, and therefore developed liver damage, more frequently exhibited BP-induced tumors than controls (96). With respect to rectal cancer, again some controversial results have been published (Table 2). Most recent data demonstrate that chronic ethanol ingestion stimulates rectal carcinogenesis initiated by the primary carcinogen acetoxymethylmethylnitrosamine (AMMN), which was applied locally to the rectal mucosa and does not need further metabolism (26). These results suggest that alcohol may influence rectal carcinogenesis by a local effect on the rectal mucosa.

The effect of alcohol on pancreatic carcinogenesis seems to be species dependent. While ethanol feeding did not enhance chemically induced pancreatic cancer appearance in the hamster, it did stimulate tumor occurrence in the rat when given during promotion (Table 2). However, a major point of criticism in all these studies is that ethanol was given in the drinking water.

Alcohol consumption can also affect the development of spontaneous tumors of the mammary gland in female C3H/st mice by decreasing the latency period and by increasing the tumor volume of spontaneously occurring mammary adenocarcinoma (105).

POSSIBLE MECHANISMS BY WHICH ETHANOL ENHANCES CARCINOGENESIS

Ethanol may increase the susceptibility of various tissues to chemical carcinogens by a variety of mechanisms. Among these are activating chemical procarcinogens, altering the metabolism and/or distribution of carcinogens, interfering with the repair of carcinogen-mediated DNA alkylation and the immune response, stimulating cellular regeneration, and exacerbating dietary deficiencies (Figure 1).

Table 2 Effect of ethanol on chemically induced extrahepatic carcinogenesis

Authors	Species	Carcinogen[a]	Ethanol administration[b]	Target	Ethanol effect
Gibel (27)	Rat	DENA, i.g.	30% i.g. with carcinogen	Esophagus	Stimulation
Gabrial et al (23)	Rat	MBNA, i.g.	4% in d.w. continuously, Zn deficiency	Esophagus	Stimulation
Schmähl (102)	Rat	MPNA, s.c.	25% in d.w. continuously	Esophagus	No Effect
Konishi et al (50)	Rat	NNP, in diet	50% i.ph. and 10% in d.w., continuously	Esophagus	No Effect
McCoy et al (73)	Hamster	NPYR/NNN, i.p.	5% l.d. prior to and with carcinogen	Nasal cavity, trachea	Stimulation/no effect
Castonguay et al (11)	Rat	NNN, p.o./NNN, s.c.	6% l.d. prior to and with carcinogen	Nasal cavity	Stimulation/no effect
Griciute et al (32)	Mouse	DMNA, i.g.	40% i.g. with carcinogen	Forebrain	Stimulation
Griciute et al (33)	Rat	NNN, i.g.	40% i.g. with carcinogen	Forebrain	Stimulation
Capel et al (10)	Mouse	BP, i.p.	i.p. prior to carcinogen, concentration unknown	Muscle	Stimulation
Hamilton et al (36)	Rat	AM, s.c.	l.d. with various concentrations prior to and with carcinogen	Distal colon Proximal colon	Stimulation No Effect/ Inhibition
Seitz et al (107)	Rat	DMH, s.c.	6% l.d. prior to and with carcinogen	Rectum	Stimulation
Garzon et al (26)	Rat	AMMN, i.r.	6% l.d. prior to and with carcinogen	Rectum	Stimulation
Howarth & Pihl (44)	Rat	DMH, s.c.	5% in d.w.	Colon/rectum	No effect
McGarrity et al (75)	Rat	DMH, s.c.	6% l.d. with carcinogen	Rectum	No effect
Tweedie et al (130)	Hamster	BOP, s.c.	25% in d.w. prior to and after carcinogen	Pancreas	Inhibition
Pour et al (95)	Hamster	BOP, s.c.	5% in d.w. prior to or after carcinogen	Pancreas	No effect
Woutersen et al (137)	Hamster	BOP, s.c.	15% in d.w. after carcinogen	Pancreas	No effect
Woutersen et al (137)	Rat	Azaserin, i.p.	15% in d.w. after carcinogen	Pancreas	Stimulation

[a]DENA = diethylnitrosamine; MBNA = methylbenzylnitrosamine; MPNA = methylphenylnitrosamine; NNP = *N*-nitrosopiperidine; NPYR = *N*-nitrosopyrrolidine; NNN = *N*-nitrosonornicotine; DMNA = dimethylnitrosamine; BP = benzo(a)pyrene; AM = azoxymethane; DMH = dimethylhydrazine; AMMN = acetoxymethylmethylnitrosamine; BOP = nitroso(2-oxopropyl)amine; i.g. = intragastrically; s.c. = subcutaneously; i.p. = intraperitoneally; p.o. = orally; i.r. = intrarectally.[b] i.ph. = intrapharyngeal; d.w. = drinking water; l.d. = liquid diet.

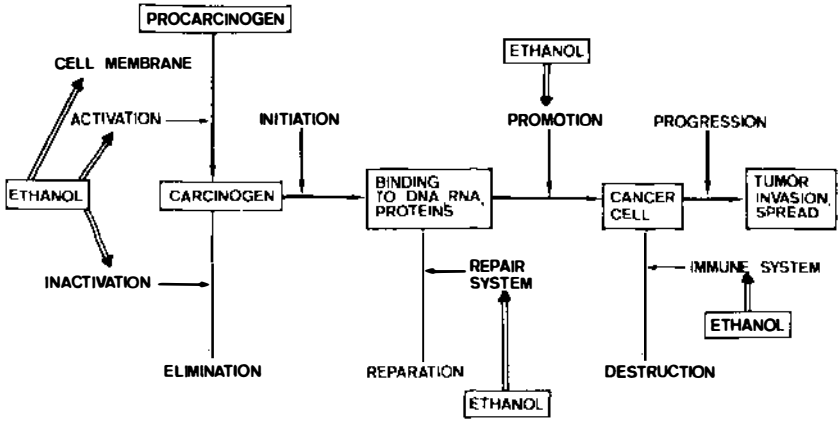

Figure 1 Simplified scheme of two-step carcinogenesis and possible sites of action of ethanol.

Contaminants of Alcoholic Beverages

In addition to the various mechanisms mentioned, some congeners (nonalcoholic components) in alcoholic beverages may play an etiologic role in the development of cancer. Indeed, esophageal cancer has been produced in animals by administering relatively large amounts of nitrosamines, congeners in some alcoholic beverages (63, 64, 110, 117). Studies of brewery workers in Denmark (46) and Ireland (14) also support the possibility that carcinogens present in some alcoholic drinks contribute to the development of cancer. Both groups regularly consumed large amounts of beer but only the Irish brewery workers exhibited an increased risk of rectal cancer. This was attributed to the fact that Danish beers have a significant lower DMN content than Irish beers.

Locally prepared alcoholic beverages peculiar in some geographic regions, likely to contain increased contaminants have in several instances been associated with increased cancer incidence. In France, the risk of esophageal neoplasia is particularly pronounced in alcoholics who drink apple brandy but is less apparent in those consuming beer and wine (63, 127–129). Similarly, in Eastern and Southern Africa, beverages derived from maize have been implicated as risk factors for esophageal carcinoma (63).

Subsequently, a variety of carcinogens such as polycyclic hydrocarbons (phenanthrene, fluoranthrene, benzanthrene, benzopyrene, chrysene) (70, 71), aflatoxins, nitrosamines (117), and asbestos fibers (derived from filters) have been detected in beer, wine, sherry, and vermouth (5, 63, 64).

Microsomal Enzyme Induction by Ethanol and Its Role in Carcinogenesis

As shown in Figure 1, ethanol seems capable of affecting carcinogenesis at different stages during initiation and promotion. Many environmental carcinogens exist in their procarcinogenic form and require metabolic activation by microsomal cytochrome-P-450-dependent enzymes (110). The activated procarcinogens exhibit a high capacity to bind to macromolecules such as DNA, RNA, or proteins and thus lead to initiation of the carcinogenic process. Induction of microsomal enzyme activities increases the mutagenic effect of many compounds in the Ames Salmonella-mutagenesis assay (1). Since the extent of metabolic activation of various secondary carcinogens can be correlated with microsomal enzyme activities (13, 21), factors such as environmental pollutants, drugs, and diet (which can influence the activity of this enzyme system) are also expected to affect tumor formation in animals exposed to carcinogens. In light of this fact, it seems important that ethanol is a well-known microsomal enzyme inducer in the liver and in other tissues (61, 111). Both ethanol and procarcinogens can be metabolized via cytochrome-P-450-dependent microsomal pathways (111).

It has been demonstrated that chronic ethanol consumption results in the appearance of a distinct form of cytochrome P-450 in the liver (52). This specific cytochrome P-450 has a preferential affinity toward aniline and 7-ethoxycoumarin and is the major P-450 responsible for the low K_m-DMN-demethylase activity (143). In addition, the capacity of hepatic, intestinal, and esophageal microsomes to activate a variety of chemical procarcinogens to mutagens, including polycyclic hydrocarbons (108, 109, 118), 2-aminofluorene (108, 109, 118), amino acid pyrolysates (65, 109), nitrosamines (25, 72, 86, 116), and aflatoxin B_1 (125, 126), is enhanced following chronic ethanol ingestion. The effect of alcohol on the metabolism of some of these procarcinogens was reviewed recently (146).

Alcohol and tobacco play a synergistic role with respect to carcinogenesis in the upper alimentary and respiratory tracts. Therefore it is of interest to note that when tobacco pyrolysate from commercially available cigarettes was tested for mutagenicity following activation by lung microsomes in the Salmonella-mutagenesis assay, an enhanced activation to mutagenic derivatives was observed with lung microsomes from rats chronically fed ethanol (109). Ethanol was also shown to enhance the induction of nasopharyngeal tumors in hamsters treated with nitrosopyrrolidine (NPY), a procarcinogen present in cigarette smoke (73), and also to increase the activation of NPY to a mutagen by microsomes isolated from hepatic, lung, and esophageal tissue (19).

Although the microsomal cytochrome-P-450-dependent biotransformation system is essential for the activation of most chemical procarcinogens, induc-

tion of this enzyme system does not necessarily entail an increased cancer risk. This is probably related to the fact that microsomal metabolism of some compounds, such as BP, gives rise to multiple products and that the components of the microsomal enzyme system and associated enzymes, such as epoxide hydratase and glutathione transferase are also involved in the detoxification of many of the same chemicals that require activation and also may be inducible by ethanol (Figure 1). Ultimately, what is probably most critical in the relationship between enzyme activities and carcinogenesis is the relative effect of the inducer such as ethanol on the steady-state level of activated carcinogens. This may be particularly important in the esophagus, where ethanol has a much greater effect on carcinogen-activating systems relative to detoxifying enzymes as compared to the liver (63, 120).

In aflatoxin-B_1-induced hepatocarcinogenesis, for example, chronic ethanol ingestion led to an enhanced activation of the procarcinogen (125, 126) but did not increase the level of DNA-bound aflatoxin B_1 in the liver of male F344 rats (69). This lack of DNA binding is in good agreement with the fact that ethanol does not influence aflatoxin-B_1-induced cancer occurrence in animals (79).

It was suspected that alcohol could enhance the effect of vinyl chloride indirectly by affecting its degradation. The acetaldehyde formed by the metabolism of ethanol could serve as a competitive inhibitor of chloroacetaldehyde catabolism, thereby increasing its carcinogenic effect (45).

Alcohol and Nitrosamine Metabolism

Nitrosamines have been detected in alcoholic beverages (117). Since ethanol and nitrosamines are both metabolized via cytochrome-P-450-dependent microsomal enzymes, it is not surprising that the two compounds interact.

Ethanol induces microsomal DMN-*N*-demethylase activity, which functions at low DMN concentrations (25); this induction is a result of a specific ethanol-induced cytochrome P-450 species with strong affinity to DMN (143). Thus, chronic ethanol ingestion resulted in an increased microsomal capacity to activate DMN to a mutagen in the Ames test (25). Such an enhanced activation had already been observed with DMN concentrations of less than 0.3 mM, which may be of pathophysiologic relevance. However, no increase in the methylation of hepatic DNA was noted when ^{14}C-DMN was administered to ethanol-fed and control animals (53) or when the mutagenicity of DMN was tested in vivo using the host-mediated assay (28). Therefore it is not surprising that the carcinogenicity of DMN as discussed is not enhanced by alcohol in the liver (Table 1). The lack of an ethanol-mediated cocarcinogenicity may possibly be related to the fact that, although DMN-activating microsomal enzymes are induced by alcohol, inactivating enzymes may also be increased following chronic ethanol ingestion. Thus, the actual

concentrations of reactive carcinogens in target tissues may be the same. On the other hand, ethanol inhibits competitively the activity of hepatic low K_m-DMN-demethylase. In vitro a K_i of 0.31 mM was reported (90), and in the perfused rat liver less than 0.5 mM of ethanol was capable of inhibiting this enzyme (124). When DMN is administered orally, the liver can exert a "first-pass clearance" up to a DMN dose of 30 μg per kg of body weight (120). At higher doses, the hepatic enzymes are saturated, and methylation in other organs such as the kidney or esophagus occurs. Ethanol when given to rats in low amounts, equivalent to a man drinking half a liter of beer, prevents the first-pass clearance by competing for the hepatic microsomal enzymes. As a result, more nitrosamine exits from the liver, and nitrosamine-sensitive extrahepatic organs are exposed to more of the procarcinogen. Among these organs, the esophagus is of particular concern, since the administration of DMN and ethanol significantly increases methylation of esophageal DNA compared with the application of DMN alone (120). Similar results have been observed with *N*-nitrosomethylbenzylamine (53). Measurement of DMN metabolism in liver slices and in the esophageal epithelium suggests that the changes in alkylation of esophageal DNA can be the result of selective inhibition of DMN metabolism in the liver and kidney. The alkylation of esophageal DNA was greater relative to that of hepatic DNA after a small compared to a large dose of ethanol. This may be because of the low K_m of the esophageal nitrosamine-metabolizing system relative to that in liver and kidney (120).

In addition, chronic ethanol ingestion also induces esophageal nitrosamine-activating enzyme activities. Farinati et al (19) were able to demonstrate an increased activation of NPY by esophageal and pulmonary microsomes after ethanol feeding in the rat.

These biochemical data on the interaction between ethanol and nitrosamine metabolism in hepatic and extrahepatic tissue may explain, at least in part, why ethanol when given prior to or during initiation does not stimulate nitrosamine-induced hepatocarcinogenesis but enhances the development of extrahepatic tumors such as carcinoma of the esophagus, the nasal cavity, and the trachea.

Alcohol consumption may also affect nitrosamine intake and intragastric production as well as intestinal absorption. Chronic ethanol ingestion leads to chronic atrophic gastritis (133) associated with an increased intragastric pH. It is well known that under such conditions bacterial overgrowth occurs, and this may enhance the reduction of dietary nitrate to nitrite. Nitrites can react with secondary amines to form nitrosamines and this chemical reaction is catalyzed by ethanol (91). In addition, ethanol damages the intestinal mucosa (9) and this is associated with an increased absorption of nitrosamines (9).

Effect of Alcohol and Its Metabolites on DNA Metabolism

There are two effects of ethanol on DNA metabolism that might be associated with cocarcinogenic activity, namely its effects on sister chromatid exchanges (SCEs) and on DNA repair. In some way, ethanol may also affect DNA integration of genetic material from hepatitis B virus.

Obe & Ristow (87) have reported that acetaldehyde induces SCEs in tissue cultures. In addition, they found an elevation of chromosomal aberrations in the lymphocytes of alcoholics (88). The potential significance of these observations with respect to tumor promotion is related to the hypothesis that compounds with SCE activity may act as promoters (63). By increasing the frequency of SCEs, such compounds could theoretically enhance recessive mutations being converted from a heterozygous to a homozygous state and thereby lead to tumor development.

A second mechanism by which alcohol abuse may increase the risk of developing cancer is by inhibiting the capacity of cells to repair carcinogen-induced DNA damage. It was reported that DMN-induced hepatic DNA alkylation persisted for longer periods in ethanol-fed animals than in controls (24). This effect appeared to be specific for O^6-methylguanine (O^6-MeG) repair (24). The enzyme responsible for the repair of O^6-MeG adducts is O^6-MeG-transferase, which transfers methyl or ethyl groups from the O^6 position of guanine to a cysteine residue located in the enzyme, which in turn inactivates the transferase. Chronic ethanol consumption was found to reduce this enzyme activity significantly (24). It should be noted that two other studies failed to detect an effect of dietary ethanol on the repair of DMN-induced O^6-MeG adducts (4, 106). However, these studies have been criticized, and it was suspected that because of the low caloric intake, undernourishment may have occurred (63).

POSSIBLE PATHOGENESIS OF ALCOHOL-ASSOCIATED ORGAN CANCERS

Esophagus

A variety of factors may be involved in esophageal carcinogenesis. The enhanced activation of nitrosamines in the esophageal mucosa due to enzyme induction by ethanol may be further increased by an ethanol-associated zinc deficiency (3). Alcoholism, however, may also cause a deficiency of other substances that putatively protect against cancer, such as riboflavin (139), carotene, vitamin A (59, 134), folic acid (111), and vitamin C (111). Most of the hypotheses relating a specific type of deficiency to cancer development are based on animal experiments only: Chronic ethanol ingestion decreased the concentrations of vitamin A in the liver (59) and in the esophagus (84) and this is associated with an increased toxicity of vitamin A (58). Zinc de-

ficiency, which frequently occurs during chronic ethanol ingestion, significantly enhanced esophageal tumor occurrence induced by nitrosomethylbenzylamine, and this was further increased by ethanol administration (23). Recent observations of low zinc and vitamin A plasma levels in patients with squamous cell carcinoma of the esophagus (78, 134), of an inverse relationship between intake of vitamin A and C and esophageal cancer (80), and of an inverse association of carcinoma of the esophagus with the level of molybdenum in the soil of farming land (144) added supportive data for humans.

Even other mechanisms of alcohol action have to be considered in esophageal carcinogenesis. Concentrated alcoholic solutions directly damage the esophageal mucosa (85). Ethanol reduces esophageal motility and may thus prolong the exposure to procarcinogens (133). Since ethanol changes cell membrane function, including fluidity and permeability of the esophageal mucosa (114), penetration of procarcinogens into the cell can be facilitated (54, 100). This process could even be favored by the decreased production and secretion of saliva observed after chronic ethanol ingestion (68). In addition, the viscosity of the saliva was found to be increased, which resulted both in higher local concentrations of procarcinogens and less rinsing of the mucosal surface. This is particularly important for smokers, who expose their mucosa to a great variety and to high concentrations of tobacco carcinogens.

Alcohol also promotes esophageal reflux (47), which eventually may result in Barrett's syndrome, a condition possibly associated with an increased risk of cancer. Subsequently, ethanol stimulates cell proliferation in the germinative epithelium of the esophagus without any overt mucosal damage (67). Since replicating DNA (because of its partially single-stranded nature) is more reactive with chemical carcinogens than resting DNA, tissue hyperregeneration would be expected to sensitize the esophagus to chemical carcinogens.

Liver

Hepatocellular carcinoma (HCC) is common in the alcoholic. However, whether this is due to ethanol itself, to the ethanol-induced cirrhosis of the liver, to a concomitant hepatitis B infection, or to a combination of all three is not clear. It has been suggested that viral hepatitis is more common in the alcoholic than in corresponding nonalcoholic populations and therefore could contribute to the increased incidence of HCC. Indeed, an increased prevalence of serologic markers of viral hepatitis B was reported in alcoholics (82). These results have been confirmed in alcoholics from the same and different geographic areas (12, 42).

Brechot et al (6) reported that among 51 subjects with various stages of alcoholic liver disease, 19 had one or more serologic markers of HBV in their serum, 8 had HBV DNA in their livers, and in 5 the DNA was integrated in their genome. Whether this increased incidence of positivity merely reflects

the socioeconomic status of the alcoholic, whether it is a consequence of increased exposure to hepatitis infection from blood transfusions, or whether it results from enhanced susceptibility to infection remains to be determined.

Integrated HBV-DNA sequences in the liver have been reported in a number of subjects (6, 113), particularly in chronic HBV carriers. Brechot et al (6) evaluated 20 subjects with alcoholic cirrhosis and HCC, all of whom had HBV-DNA integrated into the genome of the neoplastic liver cells. However, only 9 exhibited serologic markers for hepatitis B. These results are consistent with the data from Shafritz et al (113) in a group of South Africans with HCC and from Ohnishi et al (89), who noted that hepatocarcinogenesis was hastened significantly in HB_sAg carriers if they continued to drink. These results are in contrast to autopsy studies and to prospective epidemiologic studies, both of which have been criticized on methodological grounds (63).

In addition to the association between HBV infection and hepatocellular carcinoma, associations between alcohol abuse and HCC and between cirrhosis and HCC have also been observed. Clinically, hepatocellular carcinoma in alcoholics commonly occurs in conjunction with cirrhosis of the liver, and cirrhosis may contribute to the development of cancer independently of alcohol. Indeed, numerous etiologies have been proposed for carcinoma in cirrhotic patients, mostly in conjunction with the regeneration process associated with cirrhosis.

The alcoholic, then, seems to be at jeopardy for hepatocellular carcinoma for several reasons. These include the activation of carcinogens, the production of cirrhosis and the possible facilitation of infection with hepatitis B virus. It may not be coincidental that the first reported case of spontaneous regression of well-documented hepatocellular carcinoma was associated with cessation of alcohol intake (29).

Rectum

Chronic ethanol ingestion enhances rectal carcinogenesis induced by the local application of the primary carcinogen AMMN, which does not need metabolic activation. This suggests that alcohol acts by local mechanisms in the rectal mucosa and not solely by enhancing the activation of the procarcinogen (26). One important feature in intestinal carcinogenesis is the change in mucosal cell renewal modulating response to chemical carcinogens. Utilizing the metaphase arrest technique with vincristine, Simanowski et al (115) demonstrated selectively increased cell proliferation in the rectal mucosa of ethanol-fed rats when compared to controls (115). They also found a concomitant increase in proliferative compartment size. Such a hyperproliferation and expansion of the proliferative compartment of the rectal crypt toward the intestinal lumen appears to be predictive of increased susceptibility to chemical carcinogens. The observed rectal hyperproliferation may be of secondary

compensatory nature, since light microscopy of rectal mucosa from alcoholics revealed superficial cell damage that returned to normal after two weeks of alcohol abstinence (7).

It has been suspected that acetaldehyde, a rather toxic metabolite of ethanol, may cause this tissue injury. Significantly high concentrations of acetaldehyde have been found in the distal colon after alcohol application to rats (112). These acetaldehyde concentrations were significantly increased compared to the proximal colon and the liver when calculated per gram of tissue. In addition, various aldehydes have been detected following the in vitro incubation of feces with ethanol (60). It was therefore hypothesized that bacterial production of acetaldehyde, especially in the distal colon (where the highest counts of bacteria occur), may be responsible for the acetaldehyde formation (112).

Most recent data on the effect of ethanol on AMMN-induced rectal cancer support the concept that acetaldehyde may be involved in carcinogenesis. In animals that received ethanol and disulfiram, a potent acetaldehyde-dehydrogenase inhibitor, rectal tumors appeared earlier than in animals receiving ethanol alone. Acetaldehyde concentrations were significantly elevated in serum and colonic mucosa following the application of disulfiram. To what extent the ethanol-mediated hyperenterogluconemia may further stimulate cell regeneration in the rectal mucosa remains to be determined.

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