

REVIEW ARTICLE

The role of acetaldehyde in upper digestive tract cancer in alcoholics

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Chronic excessive alcohol consumption is the strongest risk factor for upper aerodigestive tract (UADT) cancer. Multiple mechanisms are involved in alcohol-associated cancer development of the UADT, including acetaldehyde (AA) effects. AA is toxic, mutagenic, and carcinogenic. Evidence of the role of AA in alcohol-associated carcinogenesis derived from genetic linkage studies in alcoholics. Polymorphism or mutation in genes coding for AA generation or detoxification enzymes are associated with increased cancer risk. It has been clearly shown in Asians that individuals carrying the acetaldehyde dehydrogenase 2*2 (ALDH2*2) allele have a significantly increased cancer risk when they consume alcohol. In Caucasians, alcohol dehydrogenase 1*1 (ADH1C*1) allele encodes for an alcohol dehydrogenase (ADH) isoenzyme, which produces 2.5 times more AA than the corresponding allele ADH1C*2. The authors found that the ADH1C*1 allele frequency and rate of homozygosity was significantly associated with an increased risk for alcohol-related cancer. AA seems to be an important factor in alcohol-associated carcinogenesis of the UADT. (*Translational Research* 2007;149:293–297)

Abbreviations: AA = acetaldehyde; ADH = alcohol dehydrogenase; ADH1C*1 = alcohol dehydrogenase 1*1; ALDH = acetaldehyde dehydrogenase; ALDH2*2 = acetaldehyde dehydrogenase 2*2; CYP 2E1 = cytochrome P4502E1; N²-Et-dG = N²-ethyl-2-deoxyguanosine; PdG = propano-dG; ROS = reactive oxygen species; UADT = upper aerodigestive tract

Alcoholism is a frequently observed disease, and in some societies, up to 3% of the adult population reveal alcohol dependency.¹ However, even in heavy drinkers, the occurrence of certain alcohol-associated organ injuries is rather low. Only 10–15% of heavy drinkers develop, for example, cirrhosis of the

liver² and only a small percentage develop cancer. Chronic alcohol consumption is indeed a risk factor for cancer of the upper aerodigestive tract (UADT), including the oral cavity, oropharynx, hypopharynx, and esophagus, of the liver in the presence of cirrhosis, and of the large intestine and the breast.³ As the amount of alcohol is obviously not the only determinant for organ injury, genetic and environmental factors may modulate and determine organ damage or carcinogenesis. Although various factors contribute to alcohol-associated cancer development, it has been shown that acetaldehyde (AA) rather than alcohol itself is carcinogenic.⁴ Thus, the amount of AA to which cells or tissues are exposed after alcohol ingestion may be of great importance and may, among others, influence carcinogenesis. The AA concentration in tissues depends on its production and degradation. (Fig 1). In the current overview, the role of AA in carcinogenesis

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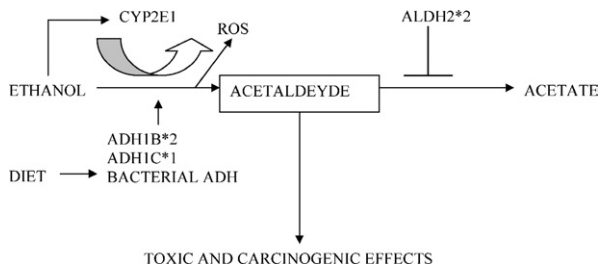


Fig 1. Ethanol metabolism and its influence by various ADH alleles and ALDH: ADH1B and ADH1C show polymorphism with the alleles ADH1B*2 and ADH1C*1, which code for rapid metabolizing enzymes, leading to increased acetaldehyde concentrations. ALDH2*2 codes for an enzyme with low activity, resulting in a decreased metabolism of acetaldehyde, and causes elevated acetaldehyde concentrations. Gastrointestinal bacteria may also contain ADH and contribute to luminal ethanol metabolism, which is influenced by diets. Finally, chronic ethanol consumption results in the induction of CYP2E1, which metabolizes ethanol to acetaldehyde and, in addition, produces reactive oxygen species (ROS).

and genetic factors modulating AA levels will be discussed in detail. With respect to other factors involved in alcohol-associated carcinogenesis, recent review articles are referenced.³⁻⁵

ACETALDEHYDE, A CARCINOGEN

Increasing evidence exists that AA rather than alcohol itself may be one important factor to explain the cocarcinogenic effect of alcohol.⁶ AA is highly toxic, mutagenic, and carcinogenic. AA interferes at many sites with DNA synthesis and repair and can, consequently, result in tumor development.⁷ Numerous *in vitro* and *in vivo* experiments in prokaryotic and eukaryotic cell cultures as well as in animal models have shown that AA has direct mutagenic and carcinogenic effects. It causes point mutations in the hypoxanthine-guanine-phosphoribosyl transferase locus in human lymphocytes, and it induces sister chromatid exchanges and gross chromosomal aberrations.⁸⁻¹⁰ It induces inflammation and metaplasia of tracheal epithelium, delays cell cycle progression, and enhances cell injury associated with hyper-regeneration.^{6,11} Thus, when AA was administered in drinking water to rodents,¹² the mucosa lesions of the UADT observed resembled those after chronic alcohol ingestion.¹³ It also been shown that AA interferes with the DNA repair machinery. AA directly inhibits O6 methyl-guanyltransferase, an enzyme important for the repair of adducts caused by alkylating agents.¹⁴ Moreover, when inhaled, AA causes nasopharyngeal and laryngeal carcinoma.¹⁵ AA also binds rapidly to cellular proteins and DNA, which results in morphological and functional impairment of the cell and in an immunologic cascade reaction. The binding to DNA and the formation of stable adducts

represent one mechanism by which AA could trigger the occurrence of replication errors or mutations in oncogenes or tumor suppressor genes.¹⁶ The occurrences of stable DNA adducts has been shown in different organs of alcohol-fed rodents and in leukocytes of alcoholics.¹⁷ In addition, it has been shown that the major stable DNA adduct, N²-ethyl-2-deoxyguanosine (N²-Et-dG), can be used by eukaryotic DNA polymerase.¹⁸ Although N²-Et-dG has been shown to form in DNA samples from white blood cells of human alcoholics and in the liver of rats given ethanol in the drinking water, relatively little evidence exists that this lesion is mutagenic, and the biological significance of the lesion is unclear. However, this lesion can be detected in human urine samples, suggesting that it may be useful as a biomarker of AA-related DNA damage.¹⁹ More recent data have shown that, in the presence of basic amino acids or histones, AA reacts with deoxyguanosine in DNA to form a different DNA adduct, 1,N²-propano-dG (PdG).¹⁹ In contrast to N²-Et-dG, PdG has been shown to be a mutagenic DNA lesion in mammalian cells *in vivo*. These AA-associated effects occurred at AA concentrations from 40 to 1000 μ M. According to the International Agency for Research on Cancer, sufficient evidence exists to identify AA as a carcinogen in experimental animals.⁷

AA is produced from ethanol by alcohol dehydrogenase (ADH) or cytochrome P4502E1 (CYP 2E1). Seven isoenzymes for ADH exist, and almost every tissue contains ADH activity. ADH2 is only present in the liver and ADH4 only in the upper gastrointestinal mucosa (for review, see Seitz and Oneta²⁰). ADH1B and ADH1C are polymorphic and thus code for enzymes capable of producing different amounts of AA.²¹ The amount of AA produced by CYP2E1 is relatively small. However, in chronic alcoholics, CYP2E1 is induced, and this pathway contributes up to 30% of the overall ethanol metabolism.²² This CYP2E1-dependent microsomal ethanol oxidizing system also produces ROS, which may be of importance in alcohol-associated carcinogenesis.^{23,24}

In addition, CYP2E1 activates various procarcinogens present in diets and tobacco smoke to their ultimate carcinogens.²⁵ For more details, recent review articles are recommended.²⁶

Increased AA concentrations may also occur when its detoxification is inadequate. AA is metabolized by acetaldehyde dehydrogenases (ALDHs) (for review, see Pares and Farres²⁷). The most important ALDH is ALDH2 with a high affinity to AA. Mutations of ALDH2 as observed in more than 40% of Asians result in elevated serum AA concentrations and cancer.⁶

ACETALDEHYD DEHYDROGENASE 2 MUTATION AND ITS ROLE IN ALCOHOL-ASSOCIATED CARCINOGENESIS

Recent and striking evidence of the causal role of AA in ethanol-associated UADT carcinogenesis derives from genetic linkage studies in alcoholics. Individuals who accumulate AA, because of polymorphism or mutation in the gene coding for enzymes responsible for AA generation and detoxification, have been shown to have an increased cancer risk. In Japan as well as in other Asian countries, a high percentage of individuals carry a mutation of the ALDH2 gene. In humans, at least 4–5 classes of ALDH isoenzymes exist.²⁰ Mitochondrial class 2 ALDH, or ALDH2, is primarily responsible for AA oxidation. Human ALDH2 enzyme is polymorphic, with 2 distinct alleles: ALDH2*1 and ALDH2*2, which results from a single-point mutation in chromosome 6 coding the normal ALDH2*1 allele. Individuals homozygous for the mutated ALDH2*2 allele are completely devoid of ALDH2 activity, whereas heterozygous individuals showing the ALDH2*1,2 genotype reveal only 30–50% of the normal ALDH activity. Blood AA levels of ALDH2*2 homozygous individuals are 6–20 times higher compared with ALDH2*1 individuals in which AA is hardly detectable after alcohol consumption. Yokohama et al²⁸ were the first to report that the heterozygous mutation of the ALDH2 gene (ALDH2*1,2) is a strong risk factor for esophageal cancer both in everyday drinkers and in alcoholics.^{28,29} A comprehensive study of the ALDH2 genotype and cancer prevalence in Japanese alcoholics showed that the frequency of inactive ALDH2 increased remarkably among alcoholics with cancer of the oral cavity, oropharynx, hypopharynx, larynx, esophagus, and colorectum.³⁰ For example, the relative risk to develop synchronous esophageal cancer in 1 individual was calculated to be over 50. These data underline the important role of AA in UADT cancer as ALDH2 heterozygotes have significantly increased AA levels after drinking alcohol. It is important to note that these individuals also have high AA in their saliva and thus deliver AA directly to the surface mucosa of the UADT.³¹

POLYMORPHISM OF ALCOHOL DEHYDROGENASE AND ITS POSSIBLE ROLE IN ALCOHOL-ASSOCIATED GASTROINTESTINAL CARCINOGENESIS

Studies on ADH1C polymorphism in Caucasians and UADT cancer have shown contradictory results. Whereas an increased risk of oropharyngeal and laryngeal cancer in individuals with the ADH1C*1 allele has been reported,^{32,33} others could not confirm such an association in case control studies.^{34–38} One reason for

this discrepancy is that in all these studies the percentage of cancer patients with high alcohol intake was rather low, sometimes extremely low. In the study by Sturgis et al,³⁵ the amount of alcohol ingested was not even reported. Thus, it is not surprising that a pooled analysis of all studies published so far came to the conclusion that the ADH1C allele is not a risk factor for alcohol-associated carcinogenesis.³⁹ Therefore, 107 alcoholic patients have been studied with oropharyngeal, laryngeal, hypopharyngeal, and esophageal cancer and a high alcohol ingestion to compare their ADH1C genotype with 103 age-matched alcoholics with a similar alcohol consumption but without cancer, and a significantly increased cancer risk was found in individuals with the ADH1C*1 allele.⁴⁰ A recent study extended this observation in a cohort of a total of 818 alcoholic patients. An odds ratio of 2.2 was found in patients with alcohol-associated UADT cancer as compared with controls (48) ($P < 0.024$), which was found to be associated with significantly elevated AA levels in the saliva of individuals homozygous for ADH1C*1 up to 100 μM .⁴⁰ Increased salivary AA levels in these individuals that were similar to individuals with ineffective ALDH activity may explain their increased cancer risk, as AA comes into direct contact with the mucosa. In this context, it is interesting to note that AA-fed rats showed a severe hyper-regeneration of the upper gastrointestinal mucosa,¹² which is very similar to the morphological changes observed after chronic alcohol consumption.¹³ These changes were only observed when the animals had functionally intact salivary glands. After sialoadenectomy, this proliferation disappeared, which supports the hypothesis that salivary AA is involved in carcinogenesis. In this context, it has to be pointed out that chronic alcohol consumption alters salivary morphology and function.⁴¹

Morphometric analysis in rats who were fed alcohol over 6 months have shown an enlargement of the size of the nuclei of the basal cells of the oral mucosa associated with an increased percentage of cells in the S-phase and a reduction of the epithelial thickness indicating mucosal atrophy and hyperproliferation.⁴² A similar finding of hyperproliferation was reported for the esophageal mucosa in rats chronically fed ethanol.¹³

AA can also be produced by oral bacteria. Significant amounts of AA can be detected in the saliva of healthy volunteers after ingestion of a moderate dose of alcohol, which is 10–20 times higher compared with systemic blood AA levels even at a higher alcohol intake.⁴³ Salivary AA concentrations after ethanol ingestion can be significantly reduced by using the antiseptic chlorhexidine before alcohol intake, emphasizing the important role of oral bacteria in AA production.⁴³ It has been shown that alcoholics with oropharyngeal

cancer had very high salivary AA concentrations,⁴⁴ which may be because smoking⁴⁵ and poor oral hygiene,⁴⁶ both frequently observed in alcoholics, result in high salivary AA concentrations because of bacterial AA production. Very recently, it has been shown that smoking changes the oral bacterial flora rapidly from Gram-negative to Gram-positive bacteria, which leads to AA concentrations 50–60% higher compared with those observed without smoking.⁴⁷ Indeed, Gram-positive bacteria are capable of producing higher amounts of AA than Gram-negative bacteria. In this context, it is important to note that nonpathogenic *Neisseria species* isolated from oral cavity can metabolize ethanol to AA.⁴⁷ In addition, *Candida albicans* also frequently belongs to the microbial environment of smokers and converts alcohol to AA. The data imply that smokers exposed to moderate amounts of alcohol produce higher AA concentrations compared with non-smokers. Apart from that, poor oral hygiene is associated with bacterial overgrowth, parodontitis, and caries and increases salivary AA concentrations.

SUMMARY AND CONCLUSION

Chronic alcohol consumption and heavy smoking are the major risk factors for UADT cancer, including the oropharynx, hypopharynx, larynx, and esophagus. Evidence has accumulated that AA is predominantly responsible for the alcohol-associated carcinogenesis because AA is carcinogenic, mutagenic, binds to DNA and protein, destructs folate, and results in secondary hyperregeneration. AA is produced by various alcohol dehydrogenases in the liver and in the gastrointestinal tract and by gastrointestinal bacteria. AA is degraded by ALDHs to acetate. Both generation and degradation of AA are modulated because of polymorphisms or mutations of the genes responsible for the enzymes involved. In addition, cigarette smoke and some alcoholic beverages also contain AA.

Other mechanisms by which alcohol stimulates carcinogenesis may include the induction of CP450E1 associated with an enhanced production of free radicals and enhanced activation of various procarcinogens present in alcoholic beverages and tobacco smoke, nutritional deficiencies, and local mechanisms.^{3–6}

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