COMMENTARY

Increase Intake of Water for Prevention of Cancer: an Enzymological Lesson?

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Abstract

Extensive studies have so far been principally made using hydrophobic carcinogens in the field of chemical carcinogenesis. However, the species of glutathione S-transferases (GSTP1-1) linked to neoplasia of rat and human were recently shown to be selective for hydrophilic carcinogens such as acrolein and hydroxyalkenals (Satoh, 1998; Satoh et al., 1999) in accord with the finding of a water-network in the active site of the human GSTP1-1 by X-ray analysis (Hu et al.; Ji et al., 1997). These results indicate that water-soluble carcinogens may be more significant than their hydrophobic counterparts in vivo. Of note, half-times for excretion of hydrophilic compounds are as short as several hrs, while those for hydrophobic ones are as long as several months or years. These available enzymological data suggest on importance of consuming water to prevent cancer.

Key Words: cancer prevention - tumor marker enzyme - glutathione S-transferase - GSTP1-1 - water - soluble carcinogens - hydrophobic carcinogens

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1. Outlook

For primary prevention of cancer, it is important to limit ingestion of carcinogens as far as possible, although this is very difficult given the fact that such compounds are so numerous (De Vita et al., 1989; Tannock and Hill, 1992; Fukushima et al., 2000). Aside from taking specific medicines or foodstuffs for health, it may be better for us to become aware of the importance of excreting exogenous and endogenous carcinogens efficiently and rapidly from our bodies. With this point in mind, it is worthwhile to reassess the importance of drinking water in the light of recent biochemical data on a tumor marker enzyme.

2. The Tumor Marker Problem

Carcinogens are first bioactivated into 'proximate' or 'ultimate' forms by the catalytic action of phase I drug-metabolizing enzymes such as mixed function oxidase cytochromes P450 as shown in Fig. 1 (Miller and Miller, 1981; Levin et al., 1982). The reactive forms, which give rise to injury to DNA bases in the initial step within the prevailing paradigm for transformation of normal into preneoplastic cells, are then mostly inactivated by phase II (detoxication) enzymes. While phase I enzymes are generally down-regulated, phase II enzymes are known to be up-regulated in preneoplastic and neoplastic lesions. Various marker enzymes are particular isozymic species of phase II enzymes such as glutathione S-transferase (GST), epoxide hydrolase, UDP-glucuronyltransferase and aldehyde dehydrogenase (Table I) (Farber, 1980; Moore and Kitagawa, 1986; Sato, 1989). One may expect that enzyme forms linked to neoplasia must have particular catalytic powers. However, the isozymic species in question generally demonstrate lower catatytic efficiencies against various substrates than non-neoplastic forms. In short, tumor marker species might appear to be physiologically of little significance. What are the real functions of the neoplastic species, if any? This is one of the most basic questions regarding tumor marker enzymes attracting interest over the last two decades.

3. GSTP1-1 is Selective for Water-soluble Carcinogens

Functional characteristics of the Pi class glutathione S-transferases, exemplified by rat GST-P and human GSTP1-1, which are most markedly increased in preneoplastic and neoplastic cells, also remain to be clarified in detail (Sato et al., 1984; Satoh et al., 1985).

The glutathione S-transferase (GST) family of multifunctional enzymes catalyzes GSH conjugation of a number of electrophilic and hydrophobic compounds including carcinogens, and is classified into three major classes, Alpha, Mu and Pi (Mannervik et al., 1985;

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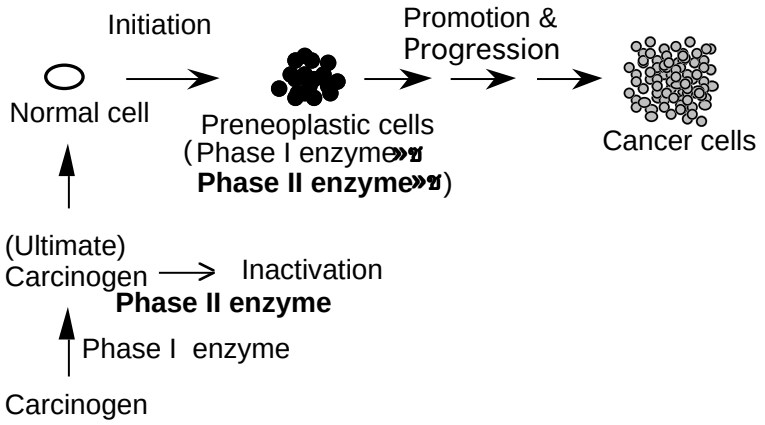


Figure 1. Participation of Drug-metabolizing Enzymes in Chemical Carcinogenesis

Mannervik and Danielson 1988). Hydrophobic interaction may play key roles in enzyme-catalyzed GSH conjugation. The active region of GST is composed of a GSH binding site (the G-site) as well as a hydrophobic compound binding site (the H-site) (Mannervik 1985; Reinemer et al., 1991). The hydrophobic force, which is one of the basic chemical interaction forces in solution, together with hydrophilic forces, electrostatic force and hydrogen bonding, is, intrinsically intricate especially with interactions among multiple hydrophobic and hydrophilic amino acid residues of proteins and enzymes (Kauzmann, 1959; Tanford, 1962). It was recently noted that the H-site hydrophobicity of the rat and man GSTP1-1 forms is the lowest among the three classes when examined by an approach incorporating theories of physical organic chemistry (Fig. 2)(Satoh, 1998; Satoh et al., 1999). The quantitative data agree well with qualitative data of crystallography obtained by Ji et al. (1997; Hu et al., 1997) who found a water-network of five water molecules in the H-site of human GSTP1-1. The results clearly indicate that Pi class species are selective for weakly hydrophobic i.e. water-soluble carcinogens rather than strongly hydrophobic and electrophilic ones. While strong electrophiles are detoxifiable by Alpha and Mu species, their water-soluble counterparts, such as acrolein and hydroxyalkenals produced in vivo, are more readily targeted by the neoplasia-associated species. The available data may, thus, shed light on the physiological role of the 'neoplastic' Pi class GSTs as specific detoxifiers of endogenous water-soluble carcinogens. Other tumor marker species such as type I UDP-glucuronyltransferase, EHb epoxide hydrolase and class 3 aldehyde dehydrogenase also appear to be selective for relatively polar substrates when compared with their respective 'non-neoplastic' isoenzymes, judging from the limited published data (Table 1). These imply that preneoplastic and neoplastic cells recognize hydrophilic carcinogens are more harmful than hydrophobic ones. It should be noted that hydrophilic compounds are rapidly metabolized in vivo. Biological half-times for excretion of hydrophilic compounds are 2.3-18h when administered orally to men, while those of hydrophobic compounds, which are incorporated into and accumulate in adipocytes, are as long as several months or years, as seen in Table 2. Taking account of the ingestion time, hydrophilic compounds may be metabolised much faster in vivo. Paradoxical, but it should also be recognized that enzymatically detoxifiable compounds themselves are fairly stable ones bacause enzyme catalyzes essentially slow a reaction (Jencks, 1969; Dixon and Webb, 1979). For example, 4-hydroxynonenal is dialysable and can be purifed by ordinary chemical techniques (Beneditti et al., 1980; Esterbauer et al., 1991). In contrast, the unstable 'ultimate carcinogens' of hydrophobic carcinogens are known to react with various macromolecules nonenzymatically (Sims and Crober, 1974; Miller and Miller, 1981; Levin et al., 1982).

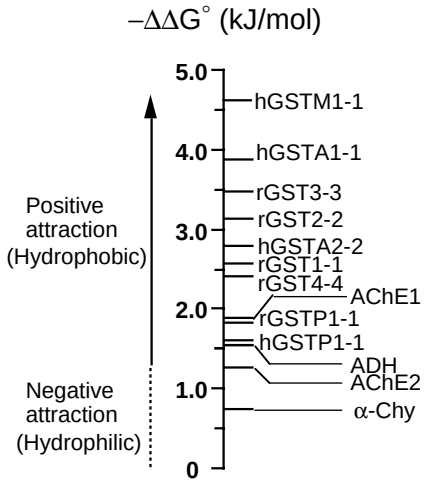


Figure 2. Active Site Hydrophobicity of Human and Rat GST Species. -ΔΔG° values are shown as a measure of the active site (the H-site) hydrophobicity of GSTs. r and h denote rat and human GST species, respectively. The active site or appropriate binding site hydrophobicities of other enzymes are also shown for comparison in the histogram. ADH, yeast alcohol dehydrogenase; AChE1 and AChE2, acetylcholine esterases, α-Chy, α-chymotrypsin (Satoh et al., 1999).

Table 1. Substrate Specificity of Preneoplastic Marker Enzymes Involved in Phase II Detoxification, Expressed in Hyperplastic Nodules and Hepatomas

Phase II Enzyme	Isozymic Species	Substrate Specificity	References
Epoxide hydrolase	EHb	broad	Batt et al., 1984
Glutathione S-transferase	GSTP1-1	broad	Satoh et al., 1991
UDP-Glucuronyl-transferase	pGT	(broad)	Yin et al., 1982
Aldehyde dehydrogenase	Type 3	broad	Lindahl et al., 1984
Quinone reductase	NQO1	(broad)	Schor et al., 1978
Carboxyesterase	LI	non-specific	Kaneko et al., 1979

Accordingly, it may be helpful to drink sufficient water to detoxify endogenous carcinogens through urination.

4. Endogenous Carcinogens

The water-soluble nature of endogenous carcinogens is, however, in contrast to the hydrophobic character of most well known carcinogens thought to be principal causatives of cancer. As a matter of fact, for example, drinking water is referred to neither in cautionary "Twelve articles for cancer prevention" of the National Cancer Center of Japan, nor in those from world Anti-Cancer Foundations. This is probably due to the historical fact that chemical carcinogenesis began with cancer of chimney sweeps followed by identification of various aromatic hydrocarbons and dyes in the 1930's, most or all of which are extremely hydrophobic (Fig. 3). Although hydrophilic carcinogens were subsequently found, most extensive studies have been made principally on hydrophobic carcinogens. However, recent investigations have revealed that endogenous lipid peroxidation end products are causally involved in the pathogenesis of numerous diseases including cancers (Hallwell, 1989., Esterbauer, 1990). Unsaturated aldehydes such as acrolein and 4-hydroxyalkenals selectively modify DNA bases to give propane or etheno adducts as demonstrated in tissue DNA of humans and untreated rodents as seen in Fig. 4 (Nath and Chung, 1994; Nair et al., 1995; Chaudhary et al., 1994). Functional amino acid residues such as cysteine, lysine and histidine of proteins and enzymes are also modifiable giving rise to Michael adducts and various other ones (Fig. 5) (Beneditti et al., 1980; Esterbauer et al., 1991; Uchida et al., 1998). Modification of macromolecules thus accounts well for the cytotoxic, mutagenic and carcinogenic activities of the aldehydic compounds detoxifiable through conjugation with GSH. Exogenous carcinogens could also be the stimulators of peroxidation. It should be borne in mind that in vivo modifications are occurring in micro or ultramicroquantities, judging from the results of microanalyses, as well as from the fact that it ordinarily takes more than 10 years to develop cancer in men i.e. the carcinogenic stimuli for initiation/promotion must be very weak. The lower the substrate concentration, the less efficient the enzymatic action, so that non-enzymatic factors may be of importance in detoxification of endogenous carcinogens. Accordingly, it may be efficient to intake more water to reduce carcinogenic stimuli. This is in accord with the epidemiological data showing bladder cancer risk to be inversely-correlated with the consumption of water per day i.e. the risk for those with an intake of fluid of >2.5l/day was 0.51 as compared with unity for an intake of 1.3l/day, on follow-up of 47,900 participants over 10 years (Michaud et al., 1999).

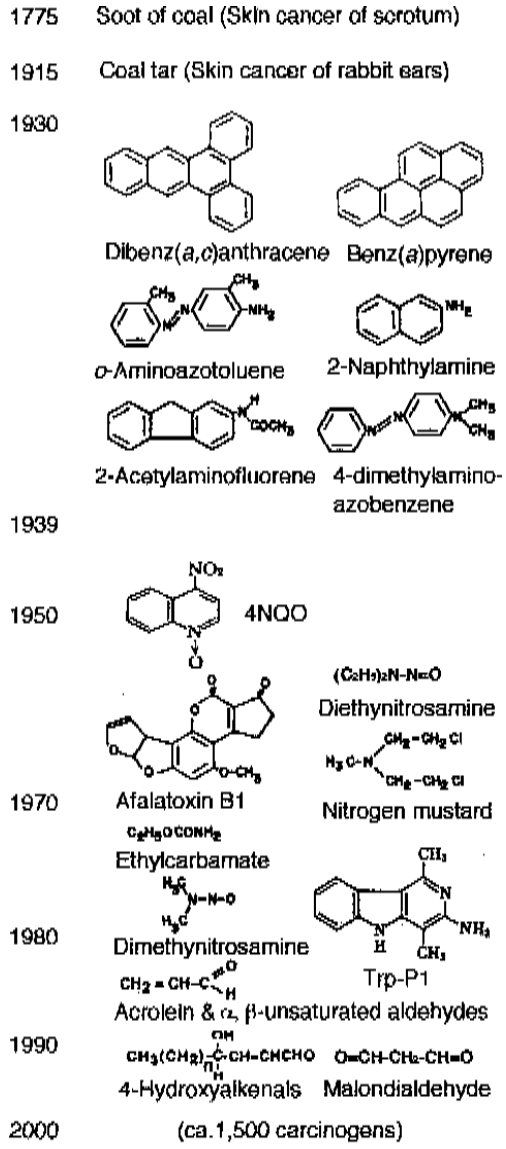
5. Additional Meaning of a Cup of Water or Tea

"Intake water for prevention and therapy of cancer" is thus an unforeseen solution to the puzzle of cancer with a firm enzymological basis, which requires further examination from various aspects e.g. 'awash with alcohol' (Moore, 2000), tea consumption (Fujiki et al., 2000),

Table 2. Excretion Rates of Hydrophobic and Hydrophilic Compounds in Humans

Compound	Biological half time	Intake/excretion routes	References
(Hydrophilic)			
Ethyl alcohol/acetaldehyde	1.6-3.5h	Oral/blood clearance	Mardones, 1963
Methanol	1.4-3.3h	Oral/blood clearance	Haffner et al., 1997
N-Acetylcysteine	2.3h	Oral/urine	Borgstrom et al ., 1986
L-Alloisoleucine	9.2h	Oral/urine	Schadewaldt et al., 1991
Methylethyl ketone	10h	Oral/urine	Sakata et al.,1989
Acetone	18h	Oral/urine	Sakata et al.,1989
Hexane	64h	Gas exposure/urine	Perbellini et al ., 1986
TCDD[a]	93.7±15.5d[b]	i.p./urine & feces	Olson et al., 1986
TCDD &PCDF[c]	5-10y	Environmental/ urine & feces	Jones and Benet 1989;
(Hydrophobic)			Kreuzer et al., 1997

2,3,7,8-Tetrachlorodibenzo-p-dioxin, [b]guinea pig data, [c]Polychlorinated dibenzofurans.



physical activity for sweating (Tang et al., 1999), and epidemological data on the risk of sleeping longer than 9 hours a day (Tamakoshi and Ohno, 2000). These could be the subjects of tea breaks to chatting about the history, chemistry, and biochemistry of carcinogens and taking broad view classifying them into hydrophobic and hydrophilic speces (Fig. 6). It should also be reminded that water has the strongest physiological activity amongst liquids, and that man and the rat are pouches of water.

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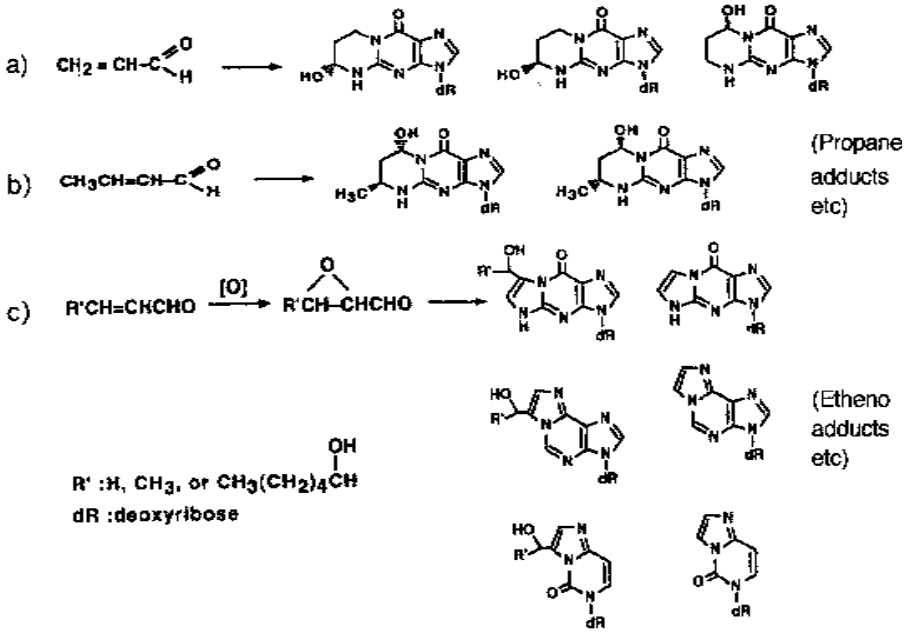

Figure 3. Upper left. A Chronology for Typical Carcinogens (Pitot, 1985; Sims and Crover, 1974; Chung, 1996).

Figure 4. Lower left. DNA Base Modification with α,β-unsaturated Adehydes (Chung et al., 1996).

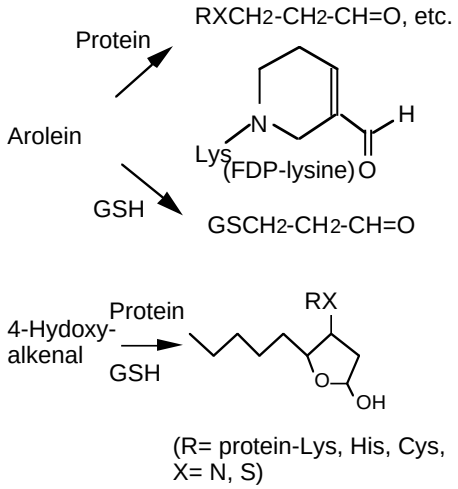

Figure 5. Amino Acid Modification of Enzymes and Proteins with a,b-unsaturated Aldehydes (Beneditti et al., 1980; Esterbauer et al., 1991; Uchida et al., 1998).

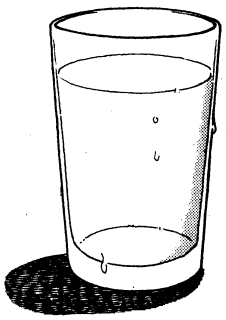

Figure 6. Enzymological but Non-enzymatic Prevention of Cancer with a Cup of Water.

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