Available online at www.sciencedirect.com
SCIENCE DIRECT®
Food and Chemical Toxicology 43 (2005) 1753–1762
www.elsevier.com/locate/foodchemtox

Modulation of cytochrome P450 enzymes by organosulfur compounds from garlic

Destiny M. Davenport *, Michael J. Wargovich

Department of Pathology and Microbiology, University of South Carolina, School of Medicine and South Carolina Cancer Center, 14 Richland Medical Park, Suite 500, Columbia, SC 29208, USA

Received 29 June 2004; accepted 24 May 2005

Abstract

Organosulfur compounds (OSCs) derived from garlic have been studied for the ability to inhibit experimental cancer in various animal models, primarily through modification of carcinogen detoxification enzymes, such as cytochrome P450 (CYP) enzymes. OSCs vary in structural and physical properties, and a detailed analysis of these properties has not been performed with respect to their ability of inhibit chemically-induced colon cancer development. Gastric intubation of rats with a single dose of 200 mg/kg diallyl sulfide (DAS), diallyl disulfide (DADS), and allyl methyl sulfide (AMS) decreased hepatic CYP2E1 protein by 45%, 25% and 47%, respectively, and this inhibition was sustained after 1, 4 and 8 weeks of treatment by these compounds. Dipropyl sulfide (DPS), dipropyl disulfide (DPDS), propyl methyl sulfide (PMS) and *S*-allylcysteine (SAC) did not inhibit hepatic CYP2E1 protein expression, nor did any of the OSCs affect CYP2E1 mRNA levels. A single dose of 200 mg/kg DAS and AMS increased hepatic CYP1A2 protein (but not mRNA) by 282% and 70%, and DAS increased CYP1A1 protein levels by 684%. Daily treatment for 1, 4 and 8 weeks with 200 mg/kg DAS and AMS resulted in time-dependent increases in hepatic CYP1A1 and CYP1A2 protein levels to a maximum of 600% and 50% for DAS, and 1600% and 240% for AMS after 8 weeks. Dosing with 200 mg/kg of each of the OSCs used in this study increased hepatic CYP3A2 protein levels at all time points. Dosing for 8 weeks with 200 mg/kg DAS, but not AMS or lower doses of DAS, induced bile duct obstruction and focal areas of necrosis. These results indicate that OSCs present in garlic, including DAS and AMS, may be beneficial in inhibiting chemically-induced colon cancer, but that longer dosing with higher concentrations of DAS may elicit minor hepatic toxicity.

Keywords: Cytochrome P450; Diallyl sulfide; Allyl methyl sulfide; Liver; Toxicity

1. Introduction

Colorectal cancer (CRC) remains one of the most common solid malignancies in Western developed countries. Unequivocal evidence exists which suggests that a diet rich in fruits and vegetables can modify cancer risk in the human population though individual components of the diet that are necessary for this protection are still a matter of intense debate. Strong evidence exists for the experimental efficacy of garlic and its associated organosulfur compounds (OSCs) (Amagase et al., 2001). These OSCs have been shown to inhibit chemically-induced cancers of the skin, forestomach, lung, breast, colon and esophagus (reviewed in Khanum et al., 2004), as well as suppress the proliferation of cancer cells in culture and inhibit the growth of transplanted tumor xenographs in vivo (Herman-Antosiewicz and Singh, 2004). Mechanisms proposed to explain this chemoprotective

Abbreviations: AMS, allyl methyl sulfide; AOM, azoxymethane; CRC, colorectal cancer; CYP, cytochrome P450; DADS, diallyl disulfide; DAS, diallyl sulfide; DASO, diallyl sulfone; $DASO_2$, diallyl sulfoxide; DPDS, dipropyl disulfide; DPS, dipropyl sulfide; GAPDH, glyceraldehyde-3-phosphate dehydrogenase; OSC, organosulfur compound; PMS, propyl methyl sulfide; SAC, *S*-allylcysteine.

* Corresponding author. Tel.: +1 9195412694; fax: +1 9195411885.
E-mail address: matthew2@niehs.nih.gov (D.M. Davenport).

0278-6915/$ - see front matter © 2005 Elsevier Ltd. All rights reserved.
doi:10.1016/j.fct.2005.05.018

activity include inhibition of the bioactivation of procarcinogens by phase I metabolic enzymes, induction of phase II detoxification enzymes, and scavenging of ultimate electrophilic carcinogenic species by the sulfur atom (reviewed in Yang et al., 2001). The phase I detoxification system, composed mainly of the cytochrome P450 (CYP) family of enzymes, is frequently the first enzymatic defense against exogenous compounds.

Inhibition of metabolic activation may be linked to the protective activity of DAS against the carcinogenicity of azoxymethane (AOM), dimethylhydrazine and nitrosodimethylamine, all of which are activated by the cytochrome P450 2E1 enzyme (CYP2E1) (Yang et al., 1994). Several compounds present in garlic are capable of inhibiting CYP2E1 in the liver of rats, which could partially explain the efficacy of these compounds in inhibiting AOM-induced colon tumors (Kwak et al., 1994; Reddy et al., 1993). However, the efficacy of these compounds with respect to structure-activity relationships has not been adequately evaluated. Previous studies suggest that the presence of an allylic side chain in certain garlic-derived OSCs may be a key determinant of the mechanism of anticarcinogenic action in that the allylic forms suppressed dimethylhydrazine cytotoxicity in the colon, but the saturated analogs of these compounds did not (Wargovich and Eng, 1989). Wattenberg's laboratory developed an alternative hypothesis suggesting that the number of sulfur atoms in the structure of a garlic-derived OSC was most indicative of the protective potential of the agent (Sparnins et al., 1988). However, these authors did not rigorously assess the role of the allylic group. Identification of the most efficient inhibitors of CYP2E1 could lead to the development of dietary supplements enhanced in these compounds, or genetic engineering of the garlic plant to produce more of these compounds.

Cytochromes P450 1A1 and 1A2 (CYP1A1 and CYP1A2) represent other cytochrome P450 enzymes important in human CRC. These enzymes are responsible for the metabolic activation of heterocyclic amines and polycyclic aromatic hydrocarbons. Several epidemiological studies have suggested that consumption of a diet containing well-done, browned or chargrilled meat, which can contain large quantities of heterocyclic amines, may be a risk factor for the development of various malignancies, including colon cancer (de Verdier et al., 1991; Giovannucci et al., 1994; Probst-Hensch et al., 1997; Schiffman and Felton, 1990). Ingestion of a diet enriched with chargrilled meat for 7 days induced hepatic CYP1A2 in humans (Fontana et al., 1999), which is the major enzyme implicated in the liver metabolism of these carcinogens. OSCs present in garlic have inhibited the formation of tumors in animals treated with various carcinogenic substrates of CYP1A1 and CYP1A2 (Huber et al., 1997; Mori et al., 1999; Sparnins et al., 1988).

Cytochrome P450 enzymes are also involved in the metabolism of a variety of pharmacological agents. In that respect, an increase in garlic consumption with the intent of preventing colon cancer may lead to adverse side effects if consumed concomitantly with other herbal supplements or pharmaceutical drugs. There are reports of dietary constituents or supplements interfering with drug metabolism, most notably those of grapefruit juice and St. John's Wort (reviewed in Sparreboom et al., 2004). CYP3A4 is the most abundant cytochrome P450 in the human liver and intestine and it is known to metabolize the majority of drugs whose biotransformation is known (Anzenbacher and Anzenbacherova, 2001). There are a few case reports claiming adverse drug interactions of garlic and warfarin (a CYP3A4 substrate) (Evans, 2000), but there does not seem to be adequate evidence to support these claims. Determination of the potential interactions of OSCs derived from garlic is important in establishing the potential for adverse reactions when taken daily as a supplement, or when increasing dietary consumption.

Although inhibition of cytochrome P450 enzymes by OSCs may be an important event in the prevention of chemically induced colon cancer, there is some evidence to suggest that cells can adapt to prolonged treatment by these compounds by changing the rate of absorption or altering metabolic activity (Knowles and Milner, 1998). This adaptation can result in diminishing activity of

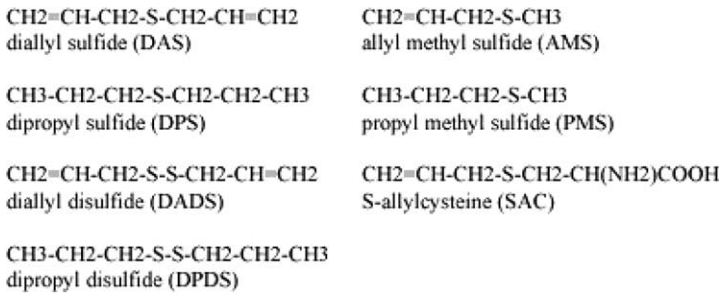

Fig. 1. Structures of the various OSCs tested.