Chemoprevention of Carcinogenesis by Organoselenium Compounds

Shigeyuki Sugie,* a,b Takuji Tanaka, c and Karam El-Bayoumy d

*Department of Pathology, a Institute of Laboratory Animals, Gifu University School of Medicine, 40 Tsukasa-machi, Gifu-city 500–8705 Japan, b Department of Pathology, Kanazawa Medical University, Ishikawa 920–0263 Japan, and c American Health Foundation, Valhalla, NY 10595, U.S.A.

(Received June 30, 2000)

The modifying effects of novel synthesized organoselenums on carcinogenesis have been examined in several organs. p-Methoxybenzeneselenol (MBS), benzylselenocyanate (BSC) and 1,4-phenylenebis(methylene)selenocyanate (p-XSC) have been synthesized, respectively. MBS reduced benzo[a]pyrene (B[a]P)-induced forestomach tumors in female CD-1 mice and azoxymethane (AOM)-induced colon, liver and kidney neoplasms in female F344 rats. BSC has been effective on AOM-induced colon neoplasms and liver preneoplastic lesions in male F344 rats, and dimethylbenz[a]anthracene (DMBA)-induced mammary tumors in female SD rats. p-XSC reduced AOM induced colon neoplasms, 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK)-induced lung neoplasms in female A/J mice, DMBA induced mammary neoplasms in female SD rats, and 4-nitroquinoline oxide(4-NQO)-induced tongue carcinogenesis in male F344 rats. BSC increased selenium-dependent glutathione peroxidase in the kidney, colon and small intestine. An increase in total liver cytochrome P-450 was also found in BSC-treated rats. Following AOM treatment, significantly less O6-methylguanine and 7-methylguanine was present in the colon DNA from rats consuming the BSC diet than in the rats fed control diets. Those results indicate that dietary BSC induces the enzymes to hydroxylate or oxidate the carcinogens and decrease DNA alkylation. p-XSC inhibited NNK-induced oxidative damage in the lung of A/J mice or F344 rats, DMBA-DNA adducts in rat mammary tissue, and induced apoptosis. These mechanisms may account for their chemopreventive activities.

Key words —— organoselenium compound, chemoprevention, carcinogenesis, animal model

INTRODUCTION

Studies examining the relationship between the intake of dietary selenium and the risk of various cancers have shown that low selenium intake is associated with higher cancer rates, including liver cancer.1–4) Epidemiological studies and a human intervention trial indicate that selenium may have chemopreventive activity in humans.3,5,6) In laboratory animal assays, chemopreventive effects of inorganic and organic selenium compounds have been observed in the mammary gland, colon, lung, pancreas, and skin.7) Earlier studies had led to the conclusion that chronic feeding of selenium in its inorganic form inhibits carcinogenesis, but that doses >5 ppm selenium are toxic in animals. Naturally-occurring selenium-containing amino acids, such as selenomethionine and selenocysteine, have about the same efficacy as inorganic selenium in cancer prevention and have comparable toxicity.7) Therefore, the development of selenium compounds with higher anticarcinogenic efficacy but better tolerance continues to be a priority in chemoprevention research. A series of novel organoselenium compounds, p-Methoxybenzeneselenol (MBS), benzylselenocyanate (BSC) and 1,4-phenylenebis(methylene)selenocyanate (p-XSC), (Fig. 1) has been synthesized in efforts to obtain improved chemoprevention, yet lower toxic activity.8–10) Effects of those compounds on carcinogenesis have been examined in several organs and found chemopreventive effects in animal models.

MBS

Chemopreventive effects of organoselenums, MBS, BSC, and phenoselenazine (PS), and their
sulfur analogue compounds, benzyl thiocyanate (BTC), p-Methoxyphenol (MP) and Phenothiazine, in the initiation phase, were examined on benzo[a]pyrene (B[a]P)-induced forestomach carcinogenesis using female CD-1 mice. Dietary administration of MBS, BSC or MP significantly inhibited, and phenothiazine enhanced, the number of forestomach tumors per animal.8) The effects of dietary MBS in initiation phase on azoxymethane (AOM)-induced carcinogenesis were studied in female F344 rats. Colon, liver and kidney neoplasms were decreased by MBS administration.9,10) BSC The effects of feeding BSC and its sulfur analogue, BTC, in the initiation phase, have been examined on AOM-induced carcinogenesis in male F344 rats.11) BSC in the diet significantly inhibited the incidence and multiplicity of total colon tumors as well as adenocarcinomas in the colon. The incidence and density of glutathione S-transferase placental form (GST-P) was reduced by BSC.12) In this experiment, selenium-dependent glutathione peroxidase (GSH-Px) was measured in some organs. BSC increased GSH-Px in the kidney, colon and small intestine. The effects of BSC, BTC and sodium selenite (Na2SeO3) in the initiation phase were studied in dimethylbenz[a]anthracene (DMBA)-induced mammary tumors using female SD rats.13) BSC inhibited mammary tumor incidence, although the other compounds did not. Fiala et al. investigated the mechanism of the chemopreventive effect of BSC on some parameters. An increase in total liver cytochrome P-450 and the increased rate of AOM metabolism was also found in BSC treated rats.14) Following AOM treatment, significantly less O\textsuperscript{6}-methylguanine and 7-methylguanine was present in the colon DNA from rats consuming the BSC diet than in the rats fed control diets.14) Those results indicate that dietary BSC induce the enzymes to hydroxylate or oxidate the carcinogens and decrease DNA alkylation. In the rat liver agitated by 2-nitropropane (2-NP), pretreatment with BSC increased the denitification activity of liver microsomes, increased liver P4502B1, decreased the levels of 2-NP-induced modifications in liver DNA and RNA, and increased the 24-h urinary excretion of nitrate.15,16) These results suggest that BSC inhibits 2-NP-induced liver nucleic acid modifications in part by increasing its detoxication through the induction of denitification.

p-XSC

El-Bayoumy et al. synthesized p-XSC, a novel organoselenium possessing low toxicity by comparison with inorganic selenium, and several other synthetic organoselenium compounds. Then they tested the effect of p-XSC (80 ppm) treatment on DMBA-induced mammary carcinogenesis during the initiation phase.17) The development of mammary tumors in those rats that received p-XSC-supplemented diets was significantly inhibited when compared with the control group. Ip et al. also studied different lower doses of p-XSC (10, 20 and 30 ppm, corresponding to 5, 10 and 15 ppm as selenium) treatment during the initiation or post-initiation phase on DMBA-induced mammary carcinogenesis.18) A suppressing effect of p-XSC and a dose-response effect was found in both phases. p-XSC treatment during the initiation phase on DMBA-induced mammary carcinogenesis was more evident than in the post-initiation phase. They also compared the chemopreventive effects of different kinds of Se compounds (sodium selenite (Na2SeO3), methyl selenocyanate (CH3SeCN), potassium selenocyanate (KSeCN), BSC and p-XSC in DMBA-induced mammary carcinogenesis.18) All five selenium compounds are active in mammary cancer prevention. With the exception of p-XSC, which has a higher effective dose range, the other four compounds are clustered around a lower effective dose range. Based on the limited data available, it appears that Na2SeO3 is equal to KSeCN in efficacy, while CH3SeCN appears to be similar in potency to BSC. Para substitution of a second methylene selenocyanate side chain to the aromatic ring of BSC to form p-XSC affects the biological activity to a much greater extent than the addition of an aromatic ring to CH3SeCN to form BSC. Reddy et al. examined the chemopreventive
effect of 40% and 80% of the maximum tolerated dose level of \( p \)-XSC (20 ppm and 40 ppm, respectively) administered in the diet during the initiation phase and the post-initiation phase of AOM-induced colon carcinogenesis in male F344 rats.\(^{19}\) \( p \)-XSC reduced AOM-induced colon neoplasms. Colonic mucosal selenium-dependent GSH-Px activity was increased, and prostaglandin \( E_2 \) was reduced in animals fed the \( p \)-XSC diet compared to animals fed the control diet. The chemopreventive effects of 10, 20 and 30 ppm (5, 10 and 15 ppm as selenium, respectively) of dietary \( p \)-XSC on tobacco-specific 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK)-induced lung neoplasms was examined in female A/J mice.\(^{20}\) Sodium selenite (5 ppm as selenum) was given in the same manner for comparison. Experimental diets were given in whole term in this study. \( p \)-XSC significantly inhibited lung tumor multiplicity in a dose dependent manner. In contrast, 5 ppm. sodium selenite had no protective effect against lung tumor induction. The results of this study clearly indicate that the structure of selenium-containing compounds is important in determining their efficacy as chemopreventive agents. We also examined whether \( p \)-XSC inhibits pulmonary neoplasia induced by NNK in female A/J mice during the initiation phase of carcinogenesis or during the post-initiation phase.\(^{21}\) Naturally occurring selenomethionine was also included in this study. Selenomethionine did not show chemopreventive activity when administered in either phase of tumorigenesis. In contrast, \( p \)-XSC significantly reduced lung tumor multiplicity regardless of whether it was given during the initiation or post-initiation. The chemopreventive effect of \( p \)-XSC was more prominent in the initiation phase than in the post-initiation phase. \( p \)-XSC can effectively inhibit NNK-induced DNA methylation and oxidative DNA damage in female A/J mice and in male F344 rats.\(^{22,23}\) Tanaka et al. studied the inhibitory effects of dietary \( p \)-XSC (5 and 15 ppm as selenium) during the initiation phase (1 week before, during, and up to 1 week after the carcinogen exposure) and during the post-initiation phase (1 week after carcinogen administration until termination) on 4-nitroquinoline oxide(4-NQO)-induced tongue carcinogenesis in male F344 rats.\(^{24}\) Dietary \( p \)-XSC, administered at selenium levels of 5 and 15 ppm during either the initiation or post-initiation phases, significantly reduced the incidence of carcinoma of the tongue. \( p \)-XSC was especially effective when it was administered at 15 ppm selenium during the post-initiation phase, in which case it completely inhibited the development of tongue carcinoma. Glutathione S-transferase (GST) activities in the liver and tongue of rats treated with 4-NQO and \( p \)-XSC were significantly elevated compared to those in rats treated with 4-NQO alone. Similarly, quinone reductase (QR-NADH) activity was significantly elevated in the liver but decreased in the tongue (posterior portion). Such modulation by \( p \)-XSC in the phase II enzyme activities of the liver and tongue might be related to inhibition of the initiation. In addition, the expression of cell proliferation biomarkers, such as polyamine level, ornithine decarboxylase (ODC) activity, 5-bromodeoxyuridine (BrdU)-labeling index, and argyrophilic nucleolar organizer regions’ protein (AgNORs), in the epithelium of the tongue was significantly reduced in rats that were fed the \( p \)-XSC diets compared to those who were fed the basal diet. Such alterations in cell proliferation through the modulation of ODC activity and polyamine biosynthesis in the tongue epithelium might be related to inhibition occurring in the post-initiation phase of carcinogenesis.

Some mechanisms of the chemopreventive effect of \( p \)-XSC have also been reported. \( p \)-XSC inhibited DNA, RNA and protein synthesis and induced apoptosis in mammary carcinoma cell lines.\(^{25,26}\) Shimada et al. studied the effects of \( \sigma \)-XSC, \( \omega \)-XSC and \( m \)-XSC on the activation of pro-carcinogens by recombinant human CYP1A1, 1A2, and 1B1 enzymes using a Salmonella typhimurium NM2009 tester strain for the detection of DNA damage. The three XSCs were found to be very potent inhibitors of metabolic activation of 3-amino-1,4-dimethyl-5H-pyrido[4,3-b]indole, 2-amino-3,5-dimethylimidazo[4,5-f]quinoline, and 2-aminoanthracene, catalyzed by CYP1A1, 1A2, and 1B1, respectively.\(^{27}\) Sohn et al. reported that those three XSCs induced GST in some organs of female CD rats.\(^{28}\) Those results indicate that \( p \)-XSC inhibits the metabolic activating enzymes, induces phase II enzymes, inhibits cell proliferation, and induces the apoptosis. These mechanisms are the reason that \( p \)-XSC is chemopreventive in both the initiation and post-initiation phase.

**CONCLUSION**

The chemopreventive effects of three novel synthesized organoseeleniums, MBS, BSC and \( p \)-XSC, were introduced. These compounds inhibit carcino-
genesis in some organs using animal models and are effective in both the initiation and post-initiation phase. These organoselenions are less toxic and more chemopreventive than inorganoseleniums and natural organoseleniums. These organoseleniums can be candidates of chemopreventive agents for human cancers.

REFERENCES