J Food Sci Nutr
Vol 11, p 278-284 (2006)

J Food Science
and Nutrition

Anticancer and Antimutagenic Activities after Simulated Digestion of Ethanol Extracts from White, Red and Yellow Onions

Mi-Yae Shon[1] and Seok-Kyu Park[2†]

[1]*Department of Food Science and Nutrition, Gyeongsang National University, Jinju 660-701, Korea*
[2]*Department of Food and Nutrition, Sunchon National University, Suncheon 540-742, Korea*

Abstract

The beneficial effects of digested onion extracts have been assessed by antimutagenic and anticancer activities by Ames test and SRB test. The total phenolic acids and flavonoids in onion extracts were determined. Red and yellow onions contain more phenolic acids and flavonoids than those in the white onion. Digested, extracts showed antimutagenic activity and anticancer activity, and it appears that the antimutagenic activity of digested extracts of onion against mutagens and anticancer activities were related to their phenols and flavonoids contents. Moreover, the extracts inhibited the proliferation of four human tumorigenic cell lines such as HT-29 (colon), MCF-7 (breast), DU-145 (prostate) and HepG2 (liver), in a dose-dependent manner. Phenolic acids and flavonoids caused oxidative damage to the cancer cell lines and induced apoptosis. Generally, red onion extracts showed effective antimutagenic and anticancer activity, and the digested red onion extracts elicited stronger antimutagenic activity than those of the onion extracts without digestion.

Key words: antimutagenic activity, anticancer activity, phenols, flavonoids, simulated digestion of onions

INTRODUCTION

Flavonoids and phenolic compounds that occur in food plants and are common components in the human diet have some therapeutic effects on anti-mutagenic and anti-carcinogenic activities (1). Food-derived flavonoids and phenolic compounds expected to be promising drugs for combating superoxide anion radical pathologies and being generally non-toxic natural compounds. Protective effect is attributed to flavonoids and phenolic compounds present in these foods. Therefore, increasing ingestion of such diets may help in maintaining good health. To investigate the mechanisms of these effects in animal bodies, several studies have been conducted to determine their absorption and distribution (2). There is increasing evidence about absorption of phenolics and some of them have been detected in human plasma and other biological fluids (3).

Cancer incidence in the population group eating lots of vegetables on a daily bases are considerably lower than those eating less as discovered in an epidemiological studies that the plant part of the diet protects against colon cancer, in particular consumption of vegetables and cruciferous vegetables (4). A high intake of fruits and vegetables seems to be protective with respect to the final outcome of hormone associated cancers such as breast and prostate (5). Inhibitors of carcinogenesis possibly present in dietary fruits and vegetables may interfere at all stages of this multistage process which at least comprises initiation, promotion, and progression. The mutagenic properties such as IQ and MNNG are acting mainly as initiators of carcinogenesis. 2-amino-3-methylimidazo-[4,5-f] quinoline (IQ) and 1-methyl-3-nitro-1-nitrosoguanidine (MNNG) are highly mutagenic in the Salmonella/reversion assay. These induce DNA damage in mammalian cells (6) and are potent multiorgan carcinogens in rodents (7). In addition, IQ was found to be a potent liver carcinogen in monkeys (8) and MNNG induce gastric cancer (9).

Onions have been shown to contain large amounts of flavonoids such as quercetin and sulfur compounds that have perceived benefits to human health. Sulfur compounds in onion possess various biological effects. In addition to volatile substances in onion, there are non-volatile sulfur-containing peptides and proteins which have been shown to have potential health benefits (10). Onion, a vegetable member of the genus Allium, has been reported to promote cardiovascular health exhibit-

†Corresponding author. E-mail: bestmeju@sunchon.ac.kr
Phone: +82-61-750-3652, Fax: +82-61-750-3650

ing decreased rates of atherosclerosis or thrombotic disease in populations with increased onion intake (2). Dietary inhibitors of mutagenesis and carcinogenesis are of particular interest because they may be useful for human cancer prevention on recently, several flavonoids and phenolic compounds have been demonstrated to have an antimutagenic effect on various mutagens or carcinogens. Also, dietary flavonoids, a group of polyphenols, suppressed the metabolism of the widespread food carcinogen IQ and MNNG (11). Therefore, the purpose of the present study was to examine the influence of digestion of onion extracts in simulated gastric and intestinal juice under various conditions with mutagens on the levels of antimutagenic and anticancer activities and whether the digestion procedure alters their activities.

MATERIALS AND METHODS

Materials

All solvents used were of HPLC grade. Reagents and chemicals were purchased from Sigma-Aldrich, Wako and Fluka. Pepsin (from hog stomach), pancreatin (from porcine pancreas), DMSO and MNNG were purchased from Sigma Chemical Co, St. Louis, Mo, USA. IQ was purchased from Wako Chemical Inc. The *Salmonella* Typhimurium strains TA98 and TA100 were purchased from KCTC (Korean Collection for Type Cultures). Aroclor 1254-induced hepatic S9 was made for the activation system in the case of IQ and MNNG. Agar and Nutrient Broth No. 2 were purchased from the Difco Laboratories, Detroit, USA and Oxoid, Hampshire, UK, respectively. DMEM (Dulbeco's modified Eagle's medium, FBS (fetal bovine serum), 0.05% trypsin-0.02% EDTA and 100 units/mL penicillin-streptomycin were purchased from GIBCO Co. (USA).

Preparation of onion extracts

The onion such as white skinned (Albion), yellow skinned (Rijnsburger) and red skinned (variety Red Baron) was purchased from local markets. Each onion was skinned, chopped and lyophilized. The lyophilized onions were then ground to a fine power. The ground onion powers (50 g) were extracted with hot water at room temperature for 12 hr at 85°C (3 times with 500 mL). The extracts were concentrated in a vacuum evaporator (EYELA, Japan) at below 40°C. All of the concentration were lyophilized and stored at -20°C.

Determination of biological materials

The contents of total phenols were analyzed by the method of Folin and Denis (12), by reading samples on a UV/Vis spectrophotometer at 760 nm. The total phenol content of each solvent extract was estimated by comparison with a standard curve generated from analysis of caffeic acid. The contents of total flavonoids were analyzed colorimetrically by the Davis method using myricetin as a standard (13).

Preparation of the gastric and intestinal juice

The simulated gastric and intestinal fluids were prepared as follows USP 23 (14). The gastric fluid contained the following: 2.0 g NaCl and 3.2 g pepsin were dissolved in 7.0 mL of HCl. Sufficient water was added to make 1,000 mL (pH 1.2). The intestinal fluid contained the following: 6.8 g KH_2PO_4 was dissolved in 250 mL of water, and mixed with 190 mL of 0.2 N NaOH and 400 mL of water. 10.0 g pancreatin was added, mixed and adjusted pH of 7.5 ± 0.1 and dilute with water to 1,000 mL.

Preparation of digested sample solution

For antimutagenic activity test, the lyophilized onion extracts were each subjected to gastric or intestinal juice and a mutagen, such as IQ or MNNG. A 0.1 mL aliquot of each solution of extract and mutagen was subjected to an equal volume of gastric juice for 30 min. Another 0.1 mL aliquot of each solution of extract and mutagen was subjected to an equal volume of intestinal juice for 2 hr. Digested samples were stored at -20°C until the analysis of the mutagenic activity. For anticancer activity test, the lyophilized onion extracts were each subjected to gastric or intestinal juice. A 0.1 mL aliquot of each solution of extract was subjected to an equal volume of gastric juice for 30 min. Another 0.1 mL aliquot of each solution of extract was subjected to an equal volume of intestinal juice for 2 hr. This process was performed in triplicate. Digested samples were stored at -20°C until the analysis of the anticancer activity.

The antimutagenic activity of the lyophilized onion extracts used in the experiments with the in vitro model was tested in the Ames test. The antimutagenic activity were used in the concentration of 3 mg per plate and the mutagenicities were induced by 0.02 μg-IQ (TA98), 0.2 μg-IQ (TA100), 0.1 μg-MNNG (TA98) and 0.01 μg-MNNG (TA100). Anticancer activity was evaluated against the cancer cell lines: HT-29 (human colon), MCF-7 (human breast), DU-145 (human prostate) and HepG2 (human liver) by SRB test.

Antimutagenic activity of onion extracts after digestion in simulated gastric and intestinal juice

Antimutagenic activity was measured using the Ames assay. The assays were performed in the presence of a rat liver activation system (S9 mix). The method was based on the 20 min preincubation procedure described by Maron and Ames (15). All samples were sterilized

by filtration through a 0.45 μm filter prior to analysis in the antimutagenic activity test. The test strains used in this assay were *Salmonella* Typhimurium TA98 and TA100. IQ and MNNG were used as the positive control chemicals. Mutagen, extracts and gastric or intestinal fluids were simultaneously digested to investigate the protection of the extracts against mutagenesis. Digested mixtures, *Salmonella* Typhimurium TA98 or TA100 and S9 mixture were used for antimutagenic activity. All experiments were performed in triplicate. Data presented in the tables are means from three independent series.

Anticancer activity of onion extracts after digestion in simulated gastric and intestinal juice

Anticancer activity was measured using the SRB assay (16). All samples were sterilized by filtration through a 0.2 μm filter prior to analysis in the anticancer activity test. The test-cells used in this assay were HT-29 (colon), MCF-7 (breast), DU-145 (prostate) and HepG2 (liver) cancer cells. Cells were maintained as adherent cell cultures in DMEM medium supplemented with 10% FBS and 10 units of penicillin and 10 μg/mL streptomycin at 37°C in a humidified incubator containing 5% CO_2. Cells were transferred into 96 well plates and incubated for 24 hr prior to the addition of test samples. Samples were added and incubated for 48 hr. The effects of the samples on the growth of human cancer cell lines were evaluated for their cytotoxic activity using a SRB (Sulforhodamine B).

RESULTS

The contents of total phenols and total flavonoids obtained by using ethanol extract are shown in Table 1. The total phenols and total flavonoids content, expressed as caffeic acid equivalents and myricetin as a standard, were 120.0 mg/g, 107.0 mg/g, 97.0 mg/g, 584.1 μg/g, 197.7 μg/g, and 250.8 μg/g in red onion, yellow onion and white onion, respectively. The ethanol extracts from onions investigated for the presence of antimutagenic activities with respect to mutagenicity induced by IQ or MNNG in *Salmonella* Typhimurium TA98 and TA100. The results obtained are summarized in Table 2; typical examples of dose-response correlation observed. Onion has already demonstrated its effective antimutagenic activity (17). Amounts of 3 mg per plate of onion extracts were sufficient to inhibit the mutagenicity induced by any of the IQ and MNNG concentrations used, red, white and yellow onion antimutagenic activity observed. These results suggest that ethanol extracts have antimutagenic effects on both IQ and MNNG. In Tables 3 and 4, ethanol extracts were digested with IQ or MNNG through the in vitro gastric juice or intestinal juice model and were analyzed in the Ames test. To determine the influence of a mutagen and digestion solutions on the in vitro antimutagenic activity of ethanol extracts, the in vitro gastric and intestinal juice model were loaded with ethanol extracts mixed with IQ or MNNG. In all cases, a concentration dependent inhibition was observed. There were no significant differences in antimutagenicity on the digestion solutions recovered with ethanol extracts. Red onion extracts showed a slightly stronger antimutagenic activity than yellow and white onion extracts when added simultaneously to the model with IQ and MNNG. Addition of a mutagen showed a small reduction of the antimutagenic capacity of ethanol extracts during the whole experiment. Purified polyphenols were prepared from onion extracts by ethanol extraction have

Table 1. Contents of total phenolic compounds and flavonoids in ethanol extract from onions

Onions	Total phenols[1)] (mg/g)	Total flavonoids[2)] (μg/g)
White	97.0±3.2[3)]	250.8±12.1
Red	120.0±9.2	584.1±18.3
Yellow	107.0±5.6	197.7±10.9

[1)]Total phenol contents based a standard curve generated by caffeic acid.
[2)]Total flavonoids contents based a standard curve generated by myricetin.
[3)]Each sample analyzed in triplicate.

Table 2. Antimutagenic activity of ethanol extract of onion against IQ (0.02 μg, 0.2 μg/plate) and MNNG (0.1 μg, 0.01 μg/plate) on *Salmonella* Typhimurium TA98 and TA100 at 3,000 μg/plate

Onions	IQ				MNNG			
	Revertants/plate[1)]		Percent inhibition (%)		Revertants/plate[1)]		Percent inhibition (%)	
	TA98	TA100	TA98	TA100	TA98	TA100	TA98	TA100
Control	435.2±2[2)]	330.0±6	0	0	153.3±10	228.2±9	0	0
Red	200.0±5*	197.7±2*	54.0±2.9*	40.1±3.3*	49.0±13*	113.4±5*	68.0±2.5*	50.3±3.6*
White	197.7±1*	205.0±10*	54.6±0.8*	38.9±5.6*	42.0±6*	102.0±2*	72.6±2.3*	55.3±0.4*
Yellow	143.3±12*	150.1±9*	67.1±0.5*	54.5±1.0*	44.7±9*	87.2±20*	70.8±1.7*	61.8±1.1*

[1)]Triplicate plates were tested per dose per experiment.
[2)]Each data is presented as the mean±SD of triplicate determinations. *$p<0.05$, compared with control.

Table 3. Antimutagenic activity of ethanol extract from three kinds of onion against IQ (0.02 μg, 0.2 μg/plate) on *Salmonella* Typhimurium TA98 and TA100 under simulated gastric juice or intestinal juice

Onions (μg/plate)		Gastric juice				Intestinal juice			
		Revertants/plate[1)]		Percent inhibition (%)		Revertants/plate[1)]		Percent inhibition (%)	
		TA98	TA100	TA98	TA100	TA98	TA100	TA98	TA100
Red	0	265.0±5[2)]	245.3±3	0	0	257±7	285.0±1	0	0
	1000	153.3±2*	116.5±6*	42.2±7*	52.5±1.0*	131.5±4*	119.0±15*	48.8±0.9*	58.3±1.0*
	2000	92.5±13*	97.0±17*	65.1±1.6*	60.5±0.7*	73.5±12*	87.7±3*	71.4±1.7*	69.2±1.3*
	3000	44.0±10*	73.0±11*	70.2±2.6*	72.1±0.7*	55.0±9*	83.0±9*	78.6±2.0*	71.8±0.8*
White	0	167.7±3	197.7±5	0	0	124.7±8	243.0±5	0	0
	1000	73.0±7*	144.3±2*	56.5±1.0*	27.0±0.4*	75.5±5*	150.1±4*	39.4±2.0*	38.2±0.9*
	2000	54.0±19*	118.5±1*	67.8±0.4*	40.0±1.1*	62.7±16*	145.0±13*	49.7±0.7*	40.3±0.8*
	3000	35.1±2*	103.5±4*	79.1±0.4*	47.6±1.7*	54.0±20*	131.1±7*	56.7±0.8*	46.0±1.6*
Yellow	0	114.7±5	314.5±5	0	0	147.7±6	311.3±3	0	0
	1000	48.0±7*	200.0±3*	58.1±1.5*	36.4±1.8*	68.7±4*	210.1±7*	53.5±1.1*	32.5±1.8*
	2000	40.0±19*	150.0±11*	65.1±1.5*	52.3±0.8*	50.0±2*	140.5±25*	66.1±0.9*	54.9±1.3*
	3000	25.7±20*	129.5±16*	77.6±0.9*	58.8±1.6*	39.7±11*	125.2±11*	73.1±1.6*	59.8±0.8*

[1)]Triplicate plates were tested per dose per experiment.
[2)]Each data is presented as the mean±SD of triplicate determinations. *$p<0.05$, compared with control.

Table 4. Antimutagenic activity of ethanol extract from three kinds of onion against MNNG (0.1 μg, 0.01 μg/plate) on *Salmonella* Typhimurium TA98 and TA100 under simulated gastric juice or intestinal juice

Onions (μg/plate)		Gastric juice				Intestinal juice			
		Revertants/plate[1)]		Percent inhibition (%)		Revertants/plate[1)]		Percent inhibition (%)	
		TA98	TA100	TA98	TA100	TA98	TA100	TA98	TA100
Red	0	244.0±3[2)]	340.9±2	0	0	350.3±1	355.7±12	0	0
	1000	165.3±5*	226.5±6*	33.6±1.2*	33.6±1.0*	274.0±3*	230.2±3*	21.8±0.4*	35.3±1.0*
	2000	121.4±11*	153.0±15*	50.3±1.6*	55.1±0.4*	204.3±23*	194.9±5*	41.7±0.7*	45.2±0.9*
	3000	83.3±8*	127.6±3*	65.9±0.4*	62.6±0.3*	173.1±10*	151.0±7*	50.6±1.3*	57.6±0.7*
White	0	229.3±3	228.5±9	0	0	238.5±3	207.5±2	0	0
	1000	180.4±2*	163.6±5*	21.3±1.0*	28.5±0.7*	210.0±7*	143.7±7*	11.9±1.1*	30.7±2.1*
	2000	135.9±10*	114.5±14*	40.7±0.4*	49.9±0.4*	153.0±9*	119.0±15*	35.9±0.8*	42.7±0.9*
	3000	120.2±17*	96.0±20*	47.6±1.3*	58.0±1.4*	115.3±10*	110.5±6*	51.7±0.3*	46.7±0.2*
Yellow	0	258.1±9	191.3±11	0	0	269.3±3	272.7±9	0	0
	1000	160.7±4*	152.0±7*	37.7±0.6*	20.4±1.7*	184.3±21*	233.3±23*	31.6±0.9*	14.5±0.6*
	2000	130.3±1*	119.0±4*	49.5±2.5*	37.8±1.1*	133.9±6*	173.1±4*	50.3±1.4*	36.4±0.8*
	3000	114.0±17*	82.1±7*	55.8±0.2*	57.1±0.3*	119.0±9*	107.2±2*	55.8±1.5*	60.7±1.6*

[1)]Triplicate plates were tested per dose per experiment.
[2)]Each data is presented as the mean±SD of triplicate determinations. *$p<0.05$, compared with control.

been tested for inhibition of the IQ and MNNG in *Salmonella* Typhimurium TA98 and TA100. In Fig. 1, 2, 3 and 4, anticancer activity was observed against HT-29 (colon), MCF-7 (breast), DU-145 (prostate) and HepG2 (liver) cancer cells. Treatment with onion extracts caused a dose-dependent reduction in cell numbers for cancer cells. The observed inhibition rate (%) for red onion extracts were 54.4%, for HT-29, at 2 mg, whereas those for white and yellow onion extracts were 48.9% and 41%, respectively.

DISCUSSION

In the antimutagenicity assays, we have found that the protective effects at 2 mg of digested red, white and yellow onions in gastric juice against the known positive mutagen IQ were about 65.1%, 67.8% and 65.1% in TA 98, 60.5%, 40.0% and 52.3% in TA100 with S9 activation. In the protective effects of digested red, white and yellow onions in gastric juice against the known positive mutagen MNNG were about 50.3%, 40.7% and 49.5% in TA 98, 55.1%, 49.9% and 37.8% in TA100. In the protective effects of digested red, white and yellow onions in intestinal juice against the known positive mutagen IQ were about 71.4%, 49.7% and 66.1% in TA 98, 69.2%, 40.3% and 54.9% in TA100 with S9 activation. In the protective effects of digested red, white and yellow onions in intestinal juice against the known

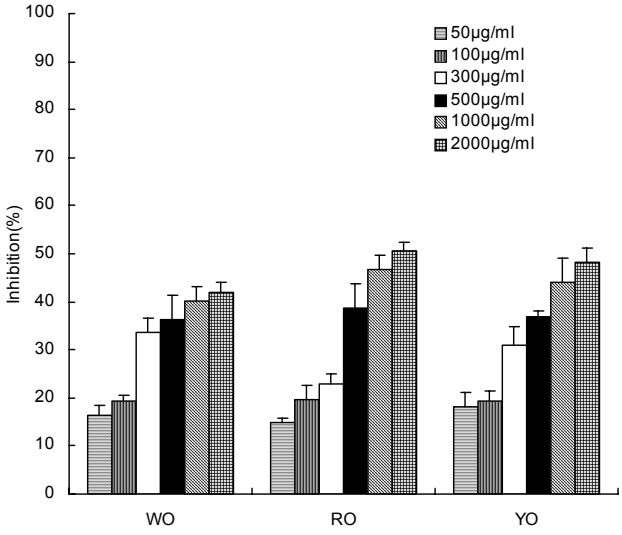

Fig. 1. In vitro inhibitory effects of WO (white onion), RO (red onion) and YO (yellow onion) on proliferation of HT-29 (colon) cancer cell. Each data is presented as the mean±SD of triplicate determinations.

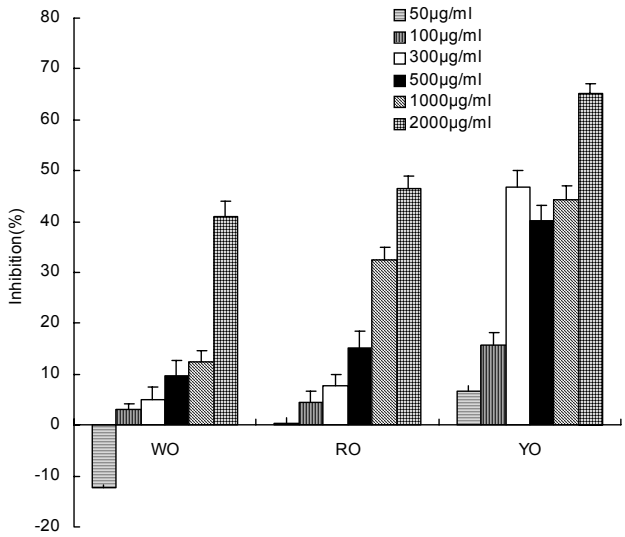

Fig. 3. In vitro inhibitory effects of WO (white onion), RO (red onion) and YO (yellow onion) on proliferation of DU-145 (prostate) cancer cells. Each data is presented as the mean±SD of triplicate determinations.

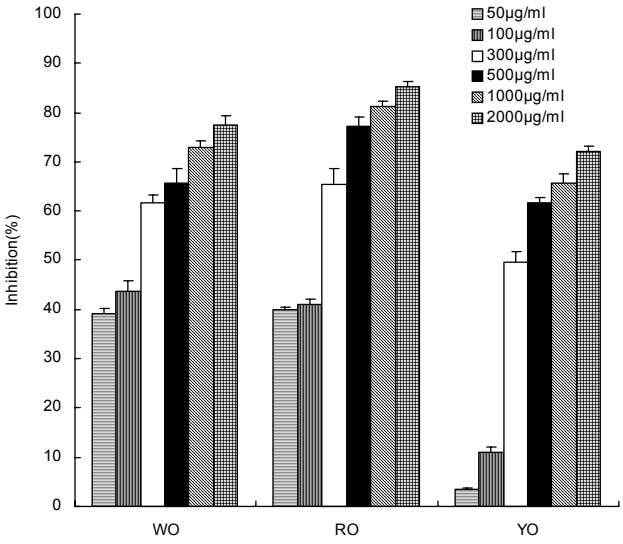

Fig. 2. In vitro inhibitory effects of WO (white onion), RO (red onion) and YO (yellow onion) on proliferation of MCF-7 (breast) cancer cells. Each data is presented as the mean±SD of triplicate determinations.

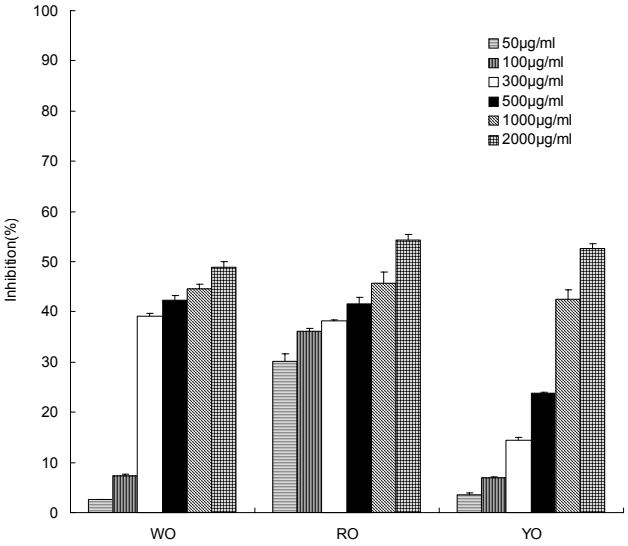

Fig. 4. In vitro inhibitory effects of WO (white onion), RO (red onion) and YO (yellow onion) on proliferation of HePG2 (liver) cancer cells. Each data is presented as the mean±SD of triplicate determinations.

positive mutagen MNNG were about 41.7%, 35.9% and 50.3% in TA98, 45.2%, 42.7% and 36.5% in TA100. These results showed that digested extracts of onions exhibit almost similar type of protective effects in vitro on bacterial systems at these concentrations tested. Also the digested extracts of red onion shows more protective effects than digested extracts of white and yellow onions. In anticancer activity, were found to be active against the cancer cell lines and showed a concentration dependent effectivity. The digested extracts of onions was the most active against breast, colon and prostate cancer cells, although digested extracts of red onion was much more effective. The contents of flavonoids on red onion were higher than white and yellow onion. Kamei et al. (18) also demonstrated that a crude anthocyanin fraction prepared from red red wine was an effective inhibitor of the growth of HCT-15 human colon tumor cells in vitro. Hagiwara et al. (19) demonstrated that anthocyanins in purple corn color reduced the promotion of colon tumors caused by 2-amino-1-methyl-6-phenylimidazo [4, 5-*b*] pyridine (PhIP) in rats initiated with dimethyl hydrazine. Since this experiment used only the crude ex-

tract of onion, it is necessary to use purified flavonoids in order to elucidate a structure-activity relationship. Any physiological significance of dietary flavonoids depends upon their availability for absorption and their subsequent interaction with target tissues, but little is known about their transport across the intestine. Flavonoids are abundant in onions flavonoids in onion are present as glycosides, it has been suggested that their hydrophilic nature and relatively high molecular weight preclude their absorption in the small intestine. Further, that by the nature of their β-glycoside linkages, they are resistant to degradation by intestinal hydrolases flavonoid glycosides pass unaltered into the large intestine. Flavonoids have been shown to be highly effective scavengers of most types of oxidizing molecules, including singlet oxygen and various free radicals (20), which are possibly involved in DNA damage and tumor promotion (21). Addition of H_2O_2 to cells in culture can lead to transition metal ion-dependent OH mediated oxidative DNA damage (22). Levels of H_2O_2 at or below about 20～50 μm seem to have limited cytotoxicity to many cell types (23).

Anticancer activity was observed against HT-29 (colon), MCF-7 (breast), DU-145 (prostate) and HepG2 (liver) cancer cells. The observed inhibition rate (%) for red onion extracts were 54.4%, 85.4%, 46.6% and 50.5% for HT-29, MCF-7, DU-145 and HepG2 at 2 mg, respectively. Red onion extracts was more effective in inhibiting the growth of these cancer cell lines than white and yellow onion extracts. Polyphenols and biomaterials in onion extracts may be beneficial to human health by resulting in a lower cancer risk.

Antimutagenic and anticancer activity of digested extracts of onion is mostly due to their antioxidant activity that inactivates direct carcinogens extracellularly. Some flavonoid and non-flavonoid phenolic compounds have been reported to also show alkylperoxyl radical scavenging activity thus reducing radical-mediated pathogenesis, e.g. carcinogenesis (24). Most chemical carcinogens require transformation by phase Ⅰ metabolizing enzymes into a more reactive form able to bind to DNA. If the resulting mutation is not repaired, it may initiate or promote the carcinogenesis process. The reactive chemical group introduced by phase Ⅰ enzymes (or the original carcinogen) can be detoxified through conjugation by phase Ⅱ enzymes metabolizing enzymes into a water-soluble compound which can then be eliminated from the body. The flavonols have been shown to inhibit cytochrome P450 enzymes (25), these enzymes play a major role in the activation of a number of suspected human carcinogens, such as polycyclic hydrocarbons and heterocyclic amines (26), flavonols plays an important role in the antimutagenic and anticancer activity of onions by cytochrome P450 dependent bioactivation of the carcinogens (27). Onions extracts also acts by scavenging radicals to produce antimutagenic effects (28).

Flavonoids can inhibit the mutagenicity of some of the cooking mutagens, the mechanism is postulated to involve inhibition of the metabolic activation process rather than binding the activated mutagen or affecting DNA repair (29), also inhibit the mutation or initiation caused by inhibition of activation of promutagens and trap the electrophiles by chemical reaction or conjugation (30). Following the Anderson et al. (30), at low doses of flavonoids, there was enhancement of the DNA damage above the effect of the mutagen. Eventually human dietary studies will be required to demonstrate that a coexposure to cooking mutagens and flavonoids actually inhibits the activation and/or DNA binding of the mutagens. The effect of dietary flavonoids is that they can reduce the intestinal uptake of benzo-α-pyrene and IQ. Antimutagenic activity of extracts against MNNG invested in the present study was very low. This observation indicates that these flavonoids are unable to interact and neutralize electrophiles such as MNNG. Other papers have also noted this failure and/or weak protective effect against direct acting mutagens (31).

In the present study, we demonstrated that digested onion extracts elicit considerable antimutagenic as well as anticancer effects against the known mutagen IQ and MNNG as observed in vitro in bacterial mutagenicity assay and in cancer cell lines. Also, Pedraza-Chaverri et al. (32) reported that onion was effective in preventing oxidative stress, indicating that onion can reduce the mutagenicity and cytotoxicity. In general, phenols are more stable to gastrointestinal conditions in ethanolic phenolic solution, and thus are effective against antimutagenic and anticancer activity. Their crude extracts in various foods and drinks, may be beneficial to human health by lowering cancer risk. Also, more detailed work is required to know their exact mechanism of the antimutagenic and anticancer actions.

REFERENCES

1. Vang OB, Rasmussen F, Andersen O. 1997. Combined effects of complex mixtures of potentially anticarcinogenic compounds on antioxidant enzymes and carcinogen metabolizing enzymes in the rat. *Cancer Lett* 114: 283-286.
2. Kendler BS. 1987. Garlic (*Allium sativm*) and onion (*Allium cepa*): a review of their relationship to cardiovascular disease. *Print Med* 16: 670-685.
3. Bourne LC, Rice-Evans CA. 1999. Detecting and measuring bioavailability of phenolics and flavonoids in humans:

pharmacokinetics of urinary excretion of dietary ferulic acid. In *Methods of enzymology*. Packer L, ed. Academic Press, Berkeley, California. p 91-106.
4. Greenwald P. 1992. Colon cancer overview. *Cancer* 70: 1206-1215.
5. Armstrong B, Doll R. 1975. Environmental factor, cancer incidence and mortality in different countries with special reference to dietary practices. *Int J Cancer* 15: 617-631.
6. Aeschbacher HU, Tursky RJ. 1991. Mutagenicity of mammalian cell and metabolism of heterocyclic aromatic amines. *Mutat Res* 259: 235-250.
7. Ohgaki H, Takayama S, Sugimura T. 1991. Carcinogenicities of heterocyclic amines in cooked food. *Mutat Res* 259: 399-410.
8. Adamson RH, Thorgeirsson VP, Snyderwine EG, Thorgeirsson SS, Reeves J, Dalgard DW, Takayama S, Sugimura T. 1990. Carcinogenicity of 2-amino-3-methyl-imidazo (4,5-f) quinoline in nonhuman primates: induction of tumors in three Macaques. *J Jpn Soc Food Science* 54: 321-326.
9. Yamada J, Tomita Y. 1994. Antimutagenic activity of water extracts of black and oolong tea. *Biosci Biotech Biochem* 58: 2197-2200.
10. Prasad K, Laxdal VA, Yu M, Raney BL. 1996. Evaluation of hydroxyl radical scavenging property of garlic. *Molecular Cellular Biochem* 154: 55-63.
11. Edenharder R, Petersdorff VI, Rauscher R. 1993. Antimutagenic effects of flavonoids, chalcones and structurally related compounds on the activity of 2-amino-3-methylimidazo[4,5-f]quinoline (IQ) and other heterocyclic amine mutagens from cooked food. *Mutat Res* 287: 261-274.
12. Folin O, Denis W. 1915. Colorimetric method for the determination of phenols (and phenol derivatives) in urine. *J Biol Chem* 22: 305-308.
13. AOAC. 1995. *Official Methods of Analysis*. 12th eds. Association of official analytical chemists, Washington, DC. p 950.
14. United States Pharmacopeia (USP23). 1995. The National Formulary (NF18). United States Pharmacopeial Convention.
15. Maron DM, Ames BN. 1983. Revised methods for the Salmonella mutagenicity test. *Mutat Res* 113: 314-321.
16. Skehan P, Storeng R, Scudiero D, Monks A, McMahon I, Vistica D, Waren JT, Bokesch H, Kenney S, Boyd MR. 1990. New colorimetric cytotoxicity assay for anticancer drug screening. *J National Cancer Inst* 82: 1107-1112.
17. Shon MY, Choi SD, Kahng GG, Nam SH, Sung NJ. 2004. Antimutagenic, antioxidant and free radical scavenging activity of ethyl acetate extracts from white, yellow and red onions. *Food Chem Toxicol* 42: 659-666.
18. Kamei H, Hashimoto Y, Koide T, Kojima T, Hasegawa M. 1998. Antitumor effect of methanol extracts from red and white wines. *Cancer Biother Radiopharm* 13: 447-452.
19. Hagiwara A, Miyashita K, Nakanishi T, Sano M, Tamano S, Kadota T, Koda T, Nakamura M, Imaida K, Ito N, Shirai T. 2001. Pronounced inhibition by a natural anthocyanins, purple corn color, of 2-amino-1-methyl-6-phenylimidazo [4,5-b] pyridine (PhIP)-associated colorectal carcinogenesis in male F344 rats pretreated with 1, 2-dimethylhydrazine. *Cancer Lett* 171: 17-25.
20. Bravo L. 1998. Polyphenols: chemistry, dietary sources, metabolism and nutritional significance. *Nutr Rev* 11: 317-333.
21. Cerutti PA. 1985. Prooxidant statse and tumor promotions. *Science* 227: 375-381.
22. Spencer J, Jenner PE, Aruoma OI, Cross CE, Wu R, Halliwell B. 1996. Oxidative DNA damage in human respiratory tract epithelial cells. Time course in relation to DNA strand breakage. *Biochem Res Commum* 224: 17-22.
23. Wang X, Martindale JL, Liu Y, Holbrook NJ. 1998. The cellular response to oxidative stress: influences of mitogen-activated protein kinase signalling pathways on cell survival. *Biochem J* 333: 291-300.
24. Sawa T, Nakao M, Akaike T, Ono K, Maeda H. 1999. Alkylperoxyl radical-scavenging activity of various flavonoids and other polyphenolic compounds: implications for the anti-tumor-promoter effect of vegetables. *J Agric Food Chem* 47: 397-402.
25. Lautraite S, Musonda AC, Doehmer J, Edwards GO, Chipman JK. 1999. Flavonoids inhibit genetic toxicity produced by carcinogens in cells expressing CYP1A2 and CYP1A1. *Mutagenesis* 17: 45-53.
26. Muto S, Fujita KI, Yamazaki T. 2001. Inhibition by green tea catechins of metabolic activation of procarcinogens by human cytochrome P450. *Mutat Res* 479: 197-206.
27. Cattral F, Copeland E, Clifford MN, Loannides C. 1998. Contribution of theaflavins to the antimutagenicity of black tea: their mechanism of action. *Mutagenesis* 13: 631-636.
28. Shiraki M, Hara Y, Osawa T, Kumon M, Nakayama T, Kawakishi S. 1994. Antioxidative and antimutagenic effects of theaflavins from black tea. *Mutat Res* 323: 29-34.
29. Lee H, Tsai SJ. 1991. Effect of emodin on cooked food mutagen activation. *Food Chem Toxicol* 29: 765-770.
30. Anderson D, Dobrzynska MM, Yn TW. 1997. Modulating effects of silymarin and myricetin on food mutagens and doxorubicin in assays with different endpoints. *J Environ Pathol Toxicol Oncol* 16: 313-327.
31. Jeanine L, Marnewick T, Wentzel CA, Gelderblom T, Elizabeth J. 2000. An investigation on the antimutagenic properties of south African herbal teas. *Mutat Res* 471: 157-166.
32. Pedraza-Chaverri J, Maldonado PD, Medina-Campos ON, Olivares-Corichi IM, Granados-Silvestre MA, Hernandez-Pando R, Ibarra-Rubio ME. 2000. Garlic ameliorates gentamicin nephrotoxicity: relation to antioxidant enzymes. *Free Radic Biol Med* 29: 602-611.

(Received November 4, 2006; Accepted December 5, 2006)