Antioxidative Effects of Sulfur Containing Compounds in Garlic on Oxidation of Human Low Density Lipoprotein Induced by Macrophages and Copper Ion

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Sulfur containing compounds in garlic have all be used as one of the traditional folk medicine as well as food source. The present study was performed to investigate the antioxidative compounds of 1-methyl-1-cysteine, dimethyl trisulfide and 2-vinyl-4*H*-1,3-dithiin. The antioxidative activity of sulfur containing compounds on human LDL was investigated by monitoring a thiobarbituric acid substances (TBARS). Sulfur containing compounds inhibited on oxidation of LDL mediated by $CuSO_4$ and macrophages in dose dependent manner with almost completely inhibition at 80 μg/ml. Antioxidant activities of sulfur containing compounds on LDL oxidation were 2-vinyl-4*H*-1,3-dithiin, 1-methyl-1-cysteine, and dimethyl trisulfide in order. Inhibitory effects of sulfur containing compounds on oxidation of LDL mediated by $CuSO_4$ and macrophages were degraded at much greater rate than native LDL, the LDL oxidation process was arrested as shown by the lower conjugated dienes formation at the concentration of 60 μg/ml. Sulfur containing compounds in garlic revealed at high antioxidative activity at low physiological concentration for human LDL oxidation *in vitro* specially, it was indicated that the antioxidative activity of 3-viny l-4*H*-1,2-dithiin was higher than that of the other sulfur containing compounds.

Key words : Garlic, antioxidant, low density lipoprotein

Introduction

An early event in atherogenesis is the accumulation of lipid-laden foam cells in the arterial intima, which can progress to fatty streaks and plaques. Most of foam cells are likely derived from resident tissue macrophages, which can lead to cellular cholesterol accumulation [12,28]. Oxdized LDL that has entered the artery wall and then accumulated in foam cells would affect atherosclerotic progress [25]. LDL oxidation was used to test the effectiveness of antioxidants to slow atherosclerosis in animal models [7]. It was observed that antioxidants or drugs with antioxidant activity were consistently able to reduce the extent of atherosclerosis [3,4,10]. The exact mechanism by which LDL undergoes oxidatioin *in vivo* is not clear yet but there is little doubt that it involves free radical peroxidation of LDL [11,29]. Since oxidized LDL seems to play a role in the development of atherosclerosis, prevention by antioxidants such as probucol [4], β-carotene [20], vitamin E [1,9,17,18,20], vitamin C [19], and flavonoids [36] may be a therapeutic option. The recent discovery of natural antioxidants may lead to replacement of the synthetic antioxidants which are widely used at present. Antioxidants from natural substances such as edible plants, species, and herbs have been widely investigated. A number of naturally occurring antioxidant compounds have been found to strengthen the resistance of LDL to oxidative modification *in vitro* and *in vivo* [2,19,26]. Attention has been focused recently on the importance of the protective defense systems in living cells against damage caused by LDL [5,11,15,29].

Garlic (*Alliium sativum L.*) has been universally as a flavoring ingredient, functional food, and traditional medicine. Historically, garlic have been utilized for folk medicine for the treatment of such varied physical disorders as wounds, chest colds, and heart disease. Using modern scientific techniques, the biological effects of garlic have been studied for a long times [6,22].

The properties of garlic have been often attributed to sulfur-containing volatile compounds such as allicin and its derivatives, which have been examined as potential anticarcinogens, antimutagens, and antimicrobial and antioxidant activity [6,24]. Many of the health benefits associated with garlic consumption have been attributed to its organosulfur compounds, particularly the thiosulfinates [R-S-S(O)-R], the single most abundant class of organosulfur

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compounds found in freshly chopped or crushed garlic [27]. Sulfur-containing components in *Allium* species consist of volatiles (disulfides and trisulfides, etc.) and nonvolatiles (sulfur-containing amino acids). The volatile sulfur compound such as dialk(an)yl disulfides and dialk(en)ly trisulfides are produced by enzymic reaction of alk(en)yl-L-cysteine sulfoxides. Allicin (2-propene-1-sulfinothioic acid S-2-propenyl ester) is the most abundant thiosulfinate, typically accounting for 70% (w/w) of the total thiosulfinates found in fresh garlic.

Another possible indicate of sulfur containing compounds bioavailability, it seemed possible to have antioxdative activity on low density lipoprotein (LDL) oxidation.

Therefore, this paper focuses antioxidant activity for LDL by sulfur containing compounds derived from garlic, which might have an important role in the benefits of garlic to human health.

Materials and Methods

Materials

Sulfur containing compounds S-methyl-L-cysteine sulfoxide, dimethyl trisulfide, 2-vinyl-4*H*-1,3-dithiin were obtained from Dr. Greenspan (College of Pharmacy, The University of Georgia, USA).

Isolation of human LDL

Lipoproteins : Human LDL was isolated from the blood of healthy man by ultracentrifugation [14] and dialyzed extensively against 0.9% (w/v) NaCl, 0.004% (w/v) EDTA, pH 7.4. Prior to oxidation, LDL was dialyzed against phosphate-buffered saline, pH 7.4, to remove the EDTA.

Cell culture : Transformed mouse macrophage J774 cells were maintained in Ham's F-10 supplemented with 10% (v/v) fetal calf serum, $NaHCO_3$ (2 g/l), and 4 mM Hepes, pH 8.1. A series of antibiotics was included in rotation in the medium. The cells were cultured routinely in large dishes (90 mm diameter) in 10 mL of medium and plated out into smaller dishes (60 mm diameter) containing 2 ml of medium. Cultures were maintained in a humidified incubator at 37°C and the medium was changed every 48 hr [12].

Oxidation of LDL

Two different methods were used to examine the effect of the sulfur containing compound on the oxidation of LDL. In the first method, LDL (100 mg protein/ml) was incubated in the presence of 5 μM $CuSO_4$ in phosphate buffered saline, pH 7.4, at 37°C for 18 hr [15].

In the second method, LDL (100 μg protein/ml) was incubated with J774 macrophages in Ham's F-10 culture medium for 24 hr at 37°C. To examine the effect of antioxidant on LDL, the reaction was stopped by the addition of EDTA (final concentration of 10 μM) and placing the lipoproteins on ice [22].

Assay of thiobarbituric acid-reactive substances (TBARS)

TBARS levels were determined spectrophotometically [30]. 0.1 ml aliquots of post incubation mixture and tetramethoxypropane standards were added into the tubes containing 1 ml of 20% trichloroacetic acid and 1 ml of 1% thiobarbituric acid containing EDTA. Tubes were placed in a boiling water bath for 30 min. After cooling, tubes were centrifuged at 1500 g for 15 min. Absorbance of the supernatant was measured at 532 nm.

Detection of conjugated dienes

The formation of conjugated dienes associated with oxidized LDL was measured by monitoring the absorption at 234 nm using a UV-VIS spectrophotometer [8]. Briefly, 1 ml of an LDL solution (100 mg LDL, protein/ml) in phosphate-buffered saline, pH 7.4, was incubated with 5 μM $CuSO_4$ at 37°C in the presence or absence of test agents, and then the absorbance at 234 nm was measured every 30 min. The formation of conjugated dienes in control solutions containing antioxidant in the absence of LDL and 5 μM $CuSO_4$ was also determined.

LDL oxidation by endothelial cells

Primary cultures of human umbilical vein endothelial cells were obtained from a cord vein, after 15 min digestion by a 0.2% collagenase solution. The cells were plated into tissue culture flask and allowed to grow to confluence in RPMI 1640 containing 20% fetal calf serum. 10 mM penicillin, 10 μg/ml streptomycin, and 2 mM L-glutamine at 37°C in a humidified atmosphere of 95% air 5% CO_2. The resulting confluent human endothelial cell cultures in multiwell clusters (1.5×10^5 cells/cm^2) were then washed three times with a serum-free medium, supplemented with 5 μM $CuSO_4$, and incubated with LDL (200 μg/ml) in a serum-free medium containing 1% human serum albumin. Before being added to the endothelial cells, the LDL was

loaded (30 min at 37°C) sulfur containing compounds or its vehicles (DMSO or ethanol respectively), and sterilized by passage through 0.22 μm milipore filters. After 18 hr incubation at 37°C the medium was aspirated, centrifuged to remove any cell debris, and finally processed for the lipid peroxidation assay by the thiobarbituric acid reaction as described above.

Determination of cellular protein

Cell protein was measured using bovine serum albumin as standard [23].

Statistics

Data in the text and figures are given as mean±SEM. Statistical significance was examined through one-way analysis of variance and Duncan's multiple range test [31]. Significant differences were accepted at $p<0.05$.

Results and Disussion

Antioxidative activities of sulfur containing compounds

Antioxidative activities of sulfur containing compounds were evaluated by comparison with ascorbic acid. Antioxidant activity of sulfur containing compounds such as S-methyl-1-cysteine sulfoxide, dimethyl trisulfide, and α-vinyl-4*H*-1,3-dithiin was comparable with ascorbic acid. The order of antioxidant activity were ascorbic acid > 2-vinyl-4*H*-1,3-dithiin > S-methyl-1-cysteine sulfoxide > dimethyl trisulfide (Fig. 1). The antioxidant test with 1, 1-diphenyl-2-picrylhydrazy (DPPH) used in this experiment is based on a portion radical scavenging activity, sulfur containing compound was also found to be positive by the thiocyanate method. Antioxidants activity of sulfur containing compounds such as S-methyl-1-cysteine sulfoxide, dimethyl trisulfide, and 2-vinyl-4*H*-1,3-dithiin have been widely investigated, although the antioxidant on LDL in these studies were not always clear. The role of nutrition and diet in the development of atherosclerosis is becoming increasingly recognized. Along with the multiple studies attempting to correlate carotenoids or vitamin C with LDL oxidizability and coronary ischemic cardiopathy [19,20], The interesting *in vitro* studies appeared recently concerning inhibition of LDL oxidation by substances. In one study, the authors demonstrated the antioxidant activity of phenolic substances. Flavonoid [2] and vitamin E [17] were shown to exhibit potent *in vitro* inhibition of LDL oxidation. These evidences prompted us to study the LDL oxidation inhibiting properties of one species, garlic commonly used in large quantities a natural product.

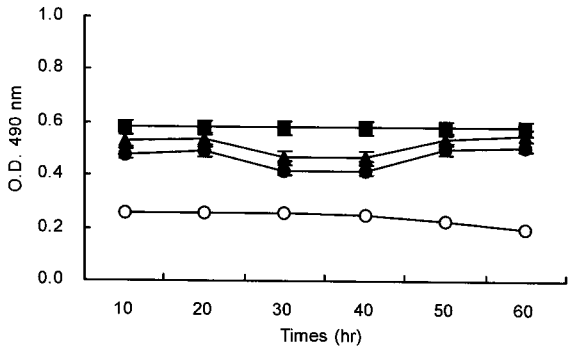

Fig. 1. Antioxidative activity of sulfur containing compounds compared to that of ascorbic acid (▲: S-methyl-1-cysteine sulfoxide, ■: dimethyl trisulfide, ●: 2-vinyl-4*H*-1,3-dithiin, ○: ascorbic acid). The concentration of each sample was tested at a dose of 40 μg/ml. Result represent average of two seperately experiments.

Antioxidative effects on human LDL

Antioxidative effects of sulfur containing compounds on the oxidation of LDL, as measured by production of TBARS, was initially examined at various concentrations of sulfur containing compounds, S-methyl-1-cysteine sulfoxide, dimethyl trisulfide, and 2-vinyl-4*H*-1,3-dithiin demonstrated a concentration-dependent inhibition of the production of oxidized LDL by Cu^{2+}-mediated LDL oxidation (Fig. 2). Sulfur containing compounds showed a dose-dependent inhibition of Cu^{2+}-mediated LDL oxidation after 24 hr of incubation. At a concentration of 80 μg/ml sulfur containing compounds such as S-methyl-1-cysteine sulfoxide, dimethyl trisulfide, 2-vinyl-4*H*-1,3-dithiin on the oxidation of LDL was almost completely inhibited than that observed in the obsence of sulfur containing compounds. Although research supports the *in vivo* existence of oxidized LDL, the most persuasive data on the role of oxidized LDL in atherogenesis derives from studies showing that antioxidants prevent atherosclerosis in animal models while some antioxidants such as BHA and BHT prevented atherosclerosis in animals [3], but their side effects preclude their use in human subjects [7]. Oxidation of LDL in the presence of copper was maximal between 2 and 3 hr of incubation; oxidation for 24 hr of incubation was almost four fold greater than at 4 hr [13]. This may reflect the level

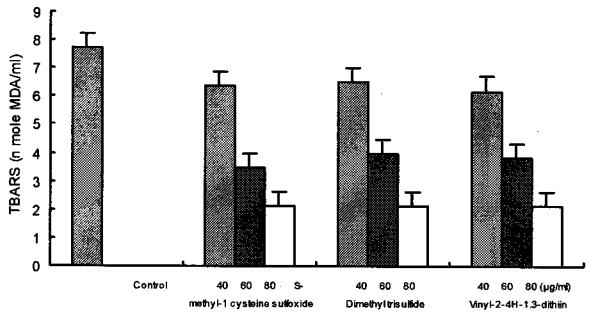

Fig. 2. Concentration-dependent inhibition effects of sulfur containing compounds on LDL oxidation mediated by Cu^{2+}. LDL (100 μg protein/ml) was incubated for 24 hr at 37°C in phosphate-buffered saline containing 5 μM $CuSO_4$ in the presence or absence of increasing concentration of sulfur containing compound. Results are presented as means±SEM of there to five independent experiments.

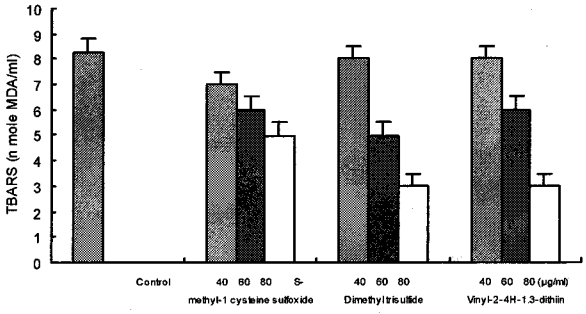

Fig. 3. Concentration-dependent inhibition effects of sulfur containing compounds on LDL oxidation mediated by macrophage. LDL (100 μg protein/dish) was incubated for 24 hr with macrophages in Hams F-10 in the presence or absence (control) of the sulfur containing compound. Results are presented as mean±SEM of three to five independent experiments.

of endogenous antioxidants present in the LDL preparation, which may vary with individual donors. For example, vitamin E as dietary antioxidant contained to partition into the LDL fraction which protection against LDL oxidation, but LDL from donors who smoker occurred more susceptible to oxidation than non-smoker [13].

As shown in Fig. 3, human LDL was oxidized by mouse J774 macrophages, in a time-dependent manner and the production of TBARS reached a plateau after 24 hr of incubation. Therefore, all data relating to macrophages presented here were obtained following 24 hr incubation. The production of TBARS in the presence or absence of the macrophages was 7.4±0.2 nmol MDA/ml and 2.20±0.04 nmol MDA/ml protein, respectively. Increasing the number of macrophages per dish increased the production of TBARS. The oxidation of LDL by J774 macrophages was inhibited in the presence of 80 μg/ml sulfur containing compounds S-methyl-1-cysteine sulfoxide, dimethyl trisulfide, and 2-vinyl-4*H*-1,3-dithiin in the culture medium. Although the mechanism of oxidation is not known, LDL oxidation may involve cellular lipoxygenases. The inhibition of macrophage induced oxidation by sulfur containing compounds is consistent with the role of lipoxygenase [26]. Macrophages on endocytose and degrade oxidatively modified LDL via scavenger receptors at a much greater rate than native LDL [23], and this property was used to assess, the protection afforded LDL by coincubation with sulfur containing compounds during the oxidation period. Many other cells types have been shown to oxidize LDL *in vitro*, e.g. mouse peritoneal macrophages [15].

It was observed that protection by sulfur containing compounds on cell-induced LDL oxidation may be, in part, through its capacity to scavenge $0_2^{\cdot}$ radicals. However, inhibition of Cu^{2+}-mediated LDL oxidation by sulfur containing compounds was marked than the oxidation by macrophages. The differences in sulfur containing compounds potency in the two systems might be due to the fact that oxidative modification is more complex than a simple free radical reaction.

Effects of sulfur containing compounds on conjugated diene formation

Fig. 4 shows the effects of sulfur containing compounds, S-methyl-1-cysteine sulfoxide, dimethyl trisulfide, and 2-vinyl-4*H*-1,3-dithiin on the formation of conjugated dienes, a measurement of the LDL oxidative process. The conjugated dienes formed were significantly lower in the concentration of each 60 μg/ml sulfur containing compounds than the control. Incubation of LDL with Cu^{2+} produced a lag phase of 120 min before the onset of the propagation phase where polyunsaturated fatty acid underwent conversion to conjugated lipid hydroperoxides. However, in the presence of sulfur containing compounds at each 60 μg/ml, the lag phase and propagation phase were inhibitory. In agreement with the findings of Esterbauer et at. [9], there was an initial lag period in the formation of conjugated dienes. The antioxidant activity of sulfur containing compounds in LDL, plasma, or arterial walls may be important in preventing or reducing the progression of atherosclerosis by inhibiting the peroxidation of lipoproteins.

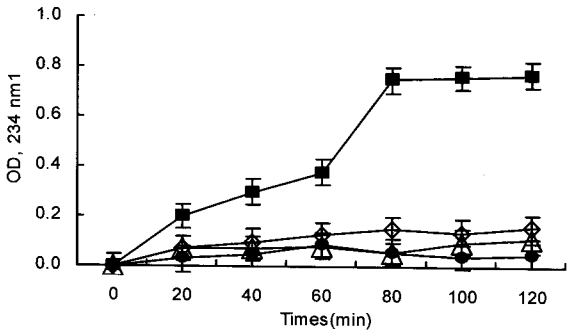

Fig. 4. Antioxidative effect of sulfur containing compounds on the formation of conjugated dienes observed during the oxidation of LDL (■: LDL+5 μM $CuSO_4$, ◇: 60 μg/ml dimethyl trisulfide, △: 60 μg/ml S-methyl-1-cysteine sulfoxide, ●: 60 μg/ml vinyl-2-4M- 1,3-dithiin). LDL (100 μg protein/ml) was incubated by the addition of 5 μM $CuSO_4$. The formation of conjugated dienes was measured at 234 nm on LDL oxidation. Results are presented as means±SEM of three to five independent experiments.

Inhibitory effects of sulfur containing compounds on LDL oxidation mediated endothelial cells

The antioxidant agent is also acting under more physiological conditions, experiments at cellular level were undertaken using human endothelial cells to oxidize LDL sulfur containing compound, S-methyl-1-cysteine sulfoxide, dimethyl trisulfide and 2-vinyl-4-*H*-1.3-dithiin successfully inhibited endothelial cell-mediated LDL oxidation in a dose-dependent manner (Fig. 5), and 50% inhibition was calculated to require about 60 μg/ml. The each concentrations of antioxidant and vehicle applied did not induce any toxic effect to endothelial cells. No dettachment was observed after the incubation period and cells washed at the end of the incubation period were able to oxidized LDL again. Decisively, sulfur containing compounds inhibited LDL oxidation at the cellular level. Endothelial cells have been proposed as one of the sources responsible for LDL modification *in vivo* by a free radical mediated mechanism. Inhibition of the oxidative modification of LDL is a crucial event in the suggested mechanism of atherosclerosis.

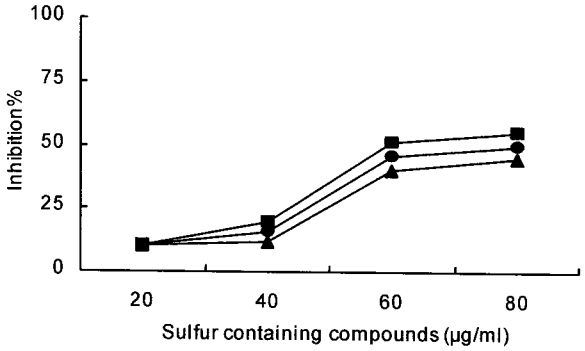

Fig. 5. Effects of sulfur containing compounds on LDL oxidation mediated artery endothelial cells (■: dimethyl trisulfide, ▲: S-methyl-1- cysteine, sulfoxide, ●: vinyl-2-4*H*-1,3-dithiin). LDL (100 μg protein/ml) was incubated with endothelial cells.

To support further the physiological relevance of these results, sulfur containing compounds were compared to ascorbic acid, which can be considerate reference antioxidants in biological systems.

From these reports, it appears that sulfur containing compounds is important in preventing the oxidative modification of LDL. Sulfur containing compounds may also play an important role in preventing the peroxidation of LDL *in vivo*, perhaps through regenerating lipid soluble antioxidants such as ascorbic acid. The antioxidant activity of sulfur containing compounds in LDL, plasma, or arterial walls may be important in preventing or reducing the progression of atherosclerosis by inhibiting the peroxidation of lipoproteins.

Acknowlegement

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References

1. Babiy, A. V., J. M. Gebicki and D. R. Sullivan. 1990. Vitamin E content and low density lipoprotein oxidizability induced by free radicals. *Atherosclerosis* **81,** 175-182.
2. Baumann, J., F. Bruchhausen and G. Wurm. 1980. Flavonoids and related compounds as inhibitors of arachidonic acid peroxidation. *Prostaglandins* **20,** 627-639.
3. Bjorkhem, I., F. Henricheson-Freychuss, O. Breuer, V. Diczfaiusy, L. Berflund and P. Henrikson. 1991. The antioxidant butylated hydroxytoluene protects against atheroscler. *Thromb.* **11,** 15-22.
4. Bridges, A. B., N. A. Scott and J. F. Belch. 1991. Probucol, a superoxide free radical scavenger *in vitro*. *Atherosclerosis* **89,** 263-265.
5. Brown, M. S. and J. L. Goldstein. 1983. Lipoprotein metabolism in the macrophage: Implications for cholesterol deposition in atherosclerosis. *Ann. Rev. Biochem.* **52,** 223-261.
6. Cavallito, C. J., J. S. Buck and C. M. Suter. 1944. Allicin,

the antibacterial principles of Allium sativim. II Determination of the chemical structure. *J. Am. Chem. Soc.* **66,** 1952-1954.
7. Cristol, L. S., I. Jialal and M. Grundy. 1992. Effect of low dose probucol therapyon LDL oxidation and the plasma lipoprotein profile in male volunteers. *Atherosclerosis* **97,** 11-20.
8. Esterbauer, H., G. Striegl, H. Puhl and M. Rotheneder. 1989. Continuous monitoring of *in vitro* oxidation of human low density lipoprotein. *Free Rad. Res. Commun.* **6,** 67-75.
9. Esterbauer, H., M. Dieber-Rotheneder, G. Striegl and G. Wage. 1991. Role of vitamin E in preventing the oxidation of low density lipoprotein. *Am. J. Clin. Nutr.* **53,** 314-321.
10. Esterbauer, H., J. Gebicki, H. Puhl and G. Jürgens. 1992. The role of lipid peroxidation and antioxidative modification of LDL. *Free Rad. Biol. Med.* **13,** 341-390.
11. Goldstein, J. L. and M. S. Brown. 1983. Lipid metabolism in the macrophage. *Annu. Rev. Biochem.* **52,** 223-262.
12. Goldstein, J. L., Y. K. Ho, S. K. Basu and M. S. Brown. 1979. Binding site on macrophages that mediates uptake and degradation of acetylated low density lipoprotein, producing massive cholesterol deposition. *Proc. Nat. Acad. Sci. U.S.A.* **76,** 333-337.
13. Harats, D., M. Ben-Naim, Y. Dabach, G. Hollander, O. Stein and Y. Stein. 1989. Cigarette smoking renders LDL susceptible to peroxidative modification and enhanced metabolism by macrophages. *Atherosclerosis* **79,** 245-252.
14. Havel, R. J., H. A. Eder and J. H. Bragdon. 1955. The distribution and chemical composition of ultracentrifugally separated lipoproteins in human serum. *J. Clin Invest.* **34,** 1345-1352.
15. Heinecke, J. W. 1987. Free radical modification of low-density lipoprotein: Mechanisms and biological consequences. *Free Rad. Biol. Med.* **3,** 65-73.
16. Morel, D. W., P. E. DiCorleto, G. M. Chisholm. 1984. Endothelial and smooth muscle cells after low density lipoproteins *in vitro* by free radical oxidation. *Arteriosclerosis* **4,** 357-364.
17. Janero, D. R. 1991. Therapeutic potential of vitamin E in the pathogenesis of spontaneous atherosclerosis. *Free Rad. Biol. Med.* **11,** 129-144.
18. Jessup, W., S. M. Rankin, C. V. De Whalley, R. S. Hoult, J. Scott and D. S. Leake. 1990. α-tocopherol consumption during low-density-lipoprotein oxidation. *Biochem. J.* **265,** 399-405.
19. Jialal, I., G. L. Vega and S. M. Grundy. 1990. Physiologic levels of ascorbate inhibit the oxidative modification of low density lipoproteins. *Atherosclerosis* **82,** 185-191.
20. Jialal, I., E. P. Norkus, L. Cristol and S. M. Grundy. 1991. β-Carotene inhibits the oxidative modification of low-density lipoprotein. *Biochim. Biophys. Acta.* **1086,** 134-138.
21. Kendler, B. S. 1987. Garlic (Allium sativum) and onion (Allium cepa): A review of their ralationship to cardiovascular disease. *Prev. Med.* **16,** 670-685.
22. Leake, D. S. and S. M. Rankin. 1990. The oxidative modification of low density lipoproteins by macrophages. *Biochem. J.* **270,** 741-748.
23. Lowry, O. H., N. J. Rosebrough, A. L. Farr and R. J. Randall. 1951. Protein measurement with the Folin phenol reagent. *J. biol. Chem.* **193,** 265-275.
24. Moore, G. S. and R. D. Atkins. 1977. The fungicidal and fungistatic effects of aqueous garlic extract on medically important yeast fungi. *Mycologia.* **69.** 341-348.
25. Ross, R. 1993. The pathogensis of atherosclerosis: perspective for the 1990s. *Nature* **362,** 801-809.
26. Robak, J., F. Shridi, M. Wolbis, M. Krolikowska and J. Sol. 1988. Screening of the influence of flavonoids of lipoxygenase and cycloxygenase activity as well as on non-enzymatic lipid oxidation. *Pharmacol. Pharm.* **40,** 451-458.
27. Srivastava, K, C., A. Bordia and S. K. Verma. 1995. Garlic (Allium sativum) for disease prevention. *South African J. Sci.* **91,** 68-77.
28. Steinbrecher, U. P., H. Zhang and M. Lougheed. 1990. Role of oxidatively modified LDL in atherosclerosis. *Free Rad. Biol. Med.* **9,** 155-178.
29. Witztum, J. L. and D. Steinberg. 1991. Role of oxidized low density lipoprotein in atherogenesis. *J. Clin. Invest.* **88,** 1785-1792.
30. Yaki, K. 1976. A simple fluoimetric assay for lipoprotein in blood plasma.*Biochem. Med.* **15,** 212-217.
31. Tallarida, R. J. and R. B. Murray. 1987. Manual of pharmacological calculations with computer programs. pp. 145-148. 2th eds., Springer Verlag, New York.

초록 : 마크로파아지 및 구리 이온으로 유도한 사람 low density lipoprotein의 산화에 대한 마늘 유황 화합물의 항산화 효과

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마늘의 주성분인 유황 함유 화합물을 이용하여 사람 low density lipoprotein (LDL)의 산화에 대하여 항산화 활성을 실험하였다. 유황함유화합물인 l-methyl-l-cysteine, dimethyl trisulfide 및 l-vinyl-4*H*-1,3-dithiin의 농도를 각각 40, 60, 80 μg/ml 씩 첨가하여 Cu^{2+} 및 macrophages 유도로 LDL을 산화할 때 항산화 효능을 TBARS로 측정한 결과 용량 의존형으로 나타났으며 유황 함유 화합물이 모두 효능이 있었으며 항산화력은 2-vinyl-4*H*-1,3-dithiin > l-methyl-l-cysteine > methyl trisulfide 순이었다. 이 때 유황 함유 화합물의 LDL에 대한 공액 2중결합에 대한 항산화 실험에서도 항산화 효과가 있었으며 60 μg/ml의 농도에서 거의 억제되었다. 유황 함유 화합물 중에서는 2-vinyl-4*H*-1,3-dithiin이 다른 유황 함유 화합물에 비하여 약간 높은 항산화 효능을 나타내었다. Endothelial cell을 이용한 LDL의 산화에 대한 억제율은 2-vinyl-4*H*-1,3-dithiin이 가장 높게 나타났다.