JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

# Antiphoto-oxidative Activity of Sesamol in Methylene Blue- and Chlorophyll-Sensitized Photo-oxidation of Oil

JOO YEON KIM, DONG SEONG CHOI AND MUN YHUNG JUNG*

Department of Food Science and Technology, Woosuk University, Samrea-Up, Wanju-Kun, Jeonbuk Province 565-701, Republic of Korea

The effects and mechanism of sesamol on the methylene blue- or chlorophyll-sensitized photo-oxidations of soybean oil have been studied. Sesamol showed strong antiphoto-oxidative activity in both methylene blue-and chlorophyll-sensitized photo-oxidations of soybean oil in a dose-dependent manner. The $1.0 \times 10^{-3}$ M sesamol treatments showed 84.7 and 43.4% inhibitions of methylene blue- and chlorophyll-sensitized photo-oxidations of soybean oil in methylene chloride. The antiphoto-oxidative activity of sesamol was comparable to that of $\delta$-tocopherol in both methylene blue- and chlorophyll-sensitized photo-oxidations, at the same molar basis. Sesamol effectively inhibited rubrene oxidation with a chemical source of singlet oxygen in microemulsion, showing its strong singlet oxygen quenching ability. The results suggested that the antiphoto-oxidative activity of sesamol in the photo-oxidation of oil was, at least in part, due to its singlet oxygen scavenging activity. The singlet oxygen quenching rate constant ($k_{ox-Q} + k_q$) of sesamol was determined to be $1.9 \pm 0.3 \times 10^7$ $M^{-1}$ $s^{-1}$. This represents the first report on the antiphoto-oxidative activity of sesamol in the sensitized photo-oxidation of oil, and its bimolecular singlet oxygen quenching ability.

**KEYWORDS: Sesamol; lipid oxidation; photo-oxidation; singlet oxygen; singlet oxygen quenching; antioxidant**

## INTRODUCTION

Oils, amino acids, proteins, vitamins (ascorbic acid, retinyl palmitate, ergosterol, carotenoids, and tocopherols), cholesterol, limonene, and conjugated terpenes in various types of foods are very susceptible to photo-oxidation, especially when photosensitizers such as chlorophylls and riboflavin are present in the systems (*1*−*8*). Photo-oxidation occurs through a type I or II reaction pathway. Type I photosensitized reaction involves the formation of superoxide anion and other radicals due to the transfer of hydrogen atoms or electrons by interaction of triplet sensitizer with molecular oxygen or other components. The type II process involves the generation of singlet oxygen by the energy transfer from an excited triplet sensitizer to a triplet oxygen. The photochemical processes in the food system are dependent on the types and concentration of sensitizers and substances in the system.

Chlorophylls, myoglobin derivatives, riboflavin, a synthetic food colorant (FD&C red no. 3), and methylene blue are reportedly efficient sensitizers for the conversion of triplet oxygen to singlet oxygen in the presence of light (*6*, *9*−*13*). Singlet oxygen reportedly induces lipid oxidation ~1450 times more rapidly than triplet oxygen (*14*). Thus, foods containing the sensitizers deteriorate rapidly under light-illuminated conditions. Effective radical scavengers such as BHA and BHT do not possess effective antioxidative properties in the photosensitized oxidation of oils due to their lack of ability in scavenging singlet oxygen (*15*−*17*). There are only a few antioxidants that could be used for the protection of foods from singlet oxygen oxidation. These are ascorbic acid, ascorbyl palmitate, carotenoids, and tocopherols (*13*, *18*−*24*). Thus, the need for novel antioxidants for the effective reduction of photosensitized oxidation of food components is obvious, and academia and industry continue to look for natural antiphoto-oxidants. Through continuous monitoring with various natural components for the antiphoto-oxidants, it was found in our laboratory that sesamol effectively reduced the methylene blue- or chlorophyll-sensitized photo-oxidation of oils.

Thus, the objectives of this research were (1) to study the qualitative and quantitative information on the antiphoto-oxidative activity of sesamol in the methylene blue- and chlorophyll-sensitized photo-oxidation of oil and (2) to determine its antiphoto-oxidation mechanism and singlet oxygen quenching activity.

## MATERIALS AND METHODS

**Materials.** Chlorophyll *b*, linoleic acid, methylene blue, 1,4-diazabicylro[2.2.2]octane (DABCO), sesamol, sodium molybdate dihydrate, and $\alpha$- and $\delta$-tocopherols were purchased from Sigma Chemical Co. (St. Louis, MO). The chemical structure of sesamol is shown in **Figure 1**. Sodium dodecyl sulfate (SDS) was purchased from Fluka Chemie AG (Buchs, Switzerland). Rubrene (5,6,11,12-tetra-

* Author to whom correspondence should be addressed (telephone 82-63-290-1438; fax 82-63-291-9312; e-mail munjung@woosuk.ac.kr).

10.1021/jf026056p CCC: $25.00 
Published on Web 04/24/2003

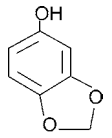

**Figure 1.** Chemical structure of sesamol.

phenylnaphthacene) was purchased from Aldrich Chemical Co. (Milwaukee, WI). Soybean oil without any additive was obtained from Heinz Korea (Inchon, Korea).

**Determination of Antiphoto-oxidative Activity.** For the study of the photo-oxidation of oil, sample solutions containing soybean oil (0.042 g/mL) and methylene blue or chlorophyll (4 $\mu$g/mL), with or without quenchers (sesamol, $\alpha$- and $\delta$-tocopherols, and DABCO), in a solvent (2-propanol, methylene chloride, or chloroform) were prepared. The reason for using the solvent model system in this study was to selectively obtain singlet oxygen lipid oxidation in the reaction media induced by sensitized photochemical reaction. Five milliliters of the prepared sample solutions were, in triplicate, transferred into 30 mL capacity serum bottles. The sample bottles were hermetically sealed with Teflon-coated rubber septa and aluminum caps. The prepared sample bottles were stored in a light storage box for 3 h as described previously (*19*). The light intensity of the sample level was 5500 lx. The temperature within the light box was $20 \pm 1$ °C during light storage. Oil oxidation was determined by measuring peroxides or conjugated dienes according to AOCS Official Methods Cd 8-53 and T1 1a-64, respectively (*25*).

**Rubrene Oxidation in Microemulsion with a Chemical Source of Singlet Oxygen.** Rubrene oxidation in microemulsion with a chemical source of singlet oxygen was performed to check whether sesamol possessed singlet oxygen quenching activity (*26*). The microemulsion is prepared at room temperature by adding an aqueous solution of 0.2 M $Na_2MoO_4 \cdot 2H_2O$ (290.4 mg in 6 mL of water) dropwise to a magnetically stirred slurry of SDS (4.7 g), 1-butanol (9.4 g), and methylene chloride (60 mL). After a few minutes, the turbid suspension is converted into a mobile and transparent liquid. Then, 0.1 g (0.2 mmol) of rubrene is introduced into a small Erlenmyer flask followed by 15 mL of the above microemulsion. The medium is magnetically stirred for 10 min in dimmed light to prevent the autosensitized photo-oxidation of rubrene. Then, 100 $\mu$L of 30% $H_2O_2$ (1 mmol) is added to the red solution, and the reaction medium is stirred at room temperature. The reaction is completed after ~5 min, as indicated by the fading of the solution to colorless. The oxidation of rubrene is monitored by visible spectroscopy at 522 nm by diluting 300 $\mu$L of the reaction medium into 5 mL of methylene chloride.

**Determination of Singlet Oxygen Quenching Rate.** The singlet oxygen quenching rate constant of sesamol was determined with slight modification according to the previously reported method (*27*, *28*). Rate determinations were carried out in chloroform because the relatively long lifetime of $^1O_2$ in this solvent allows measurements to be made at low sesamol concentrations, thus minimizing potential complications that might arise from the quenching of the rubrene-excited state by the added quenchers. The $0.3 \times 10^{-3}$ M rubrene solutions in chloroform with and without $0.5 \times 10^{-3}$ M sesamol (as a quencher) were prepared. Four milliliter rubrene samples with and without quenchers were transferred into 15 mL capacity serum bottles and hermetically sealed with Teflon-coated rubber septa and aluminum caps. Then the solution was stored for 2 min in the light box with fluorescence light illumination. The illumination time of 2 min was adjusted so that ~40−50% of the rubrene was oxidized in the sample. Three different determinations of the singlet oxygen quenching rate of sesamol were made. In each determination, a set of triplicate samples was analyzed.

**Statistical Analysis.** Statistical analysis was accomplished using an SAS method. Duncan's multiple-range test was used to ascertain the sesamol effects on the methlene blue- or chlorophyll-sensitized photo-oxidation of oil.

## RESULTS AND DISCUSSION

**Antiphoto-oxidative Activity of Sesamol in Methylene Blue-Sensitized Photo-oxidation of Soybean Oil.** The effects of sesamol at different concentrations on the methylene blue-sensitized photo-oxidation of soybean oil in 2-propanol, methylene chloride, and chloroform are shown in panels A, B, and C, respectively, of **Figure 2**. Fluorescence light illumination greatly induced peroxide formation in the sample. The initial peroxide value of soybean oil before light illumination was 0.1 mequiv/kg of oil. The mean peroxide values of soybean oil containing no sesamol in 2-propanol, methylene chloride, and chloroform after 3 h of fluorescence illumination were 190, 785,

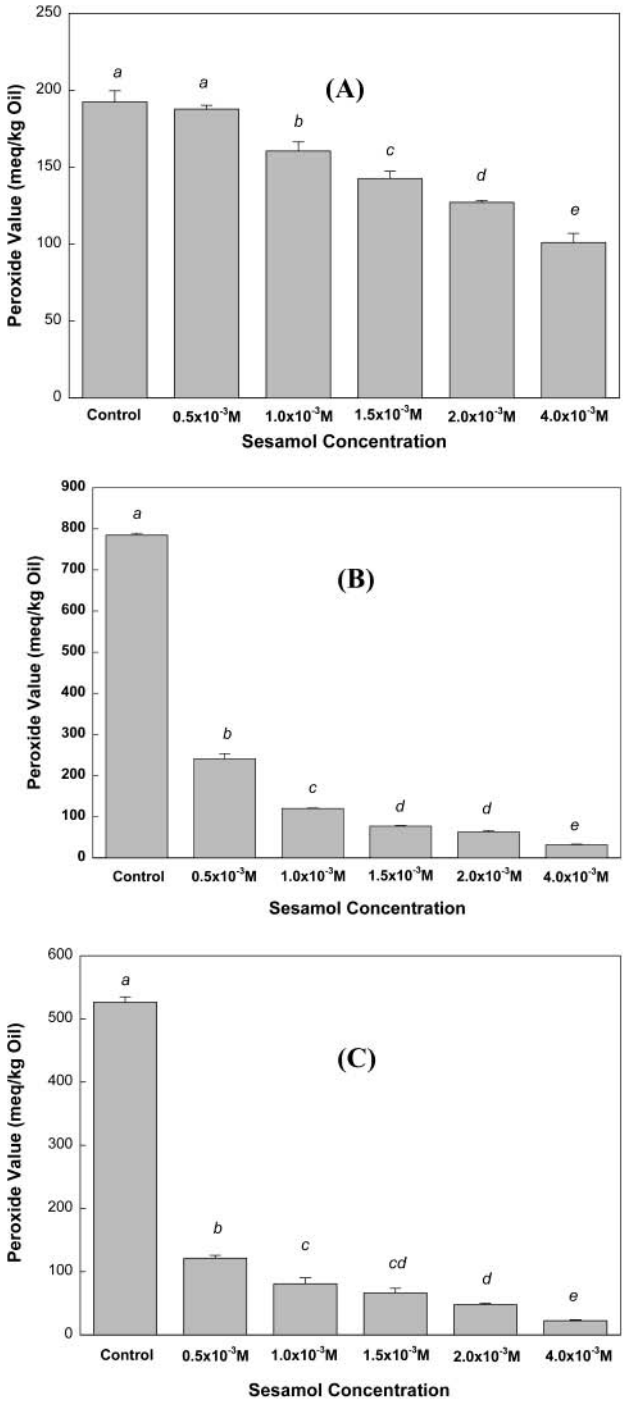


**Figure 2.** Antiphoto-oxidative activities of different concentrations of sesamol on methylene blue-sensitized photo-oxidation of soybean oil in 2-propanol (A), methylene chloride (B), and chloroform (C) during 3 h of fluorescent light illumination (5500 lx) at $20 \pm 1$ °C. The methylene blue concentration in this experimental system was 4 $\mu$g/mL. The antioxidative activities with the same italicized letter at the top of the bar were not significantly different at $\alpha = 0.05$.

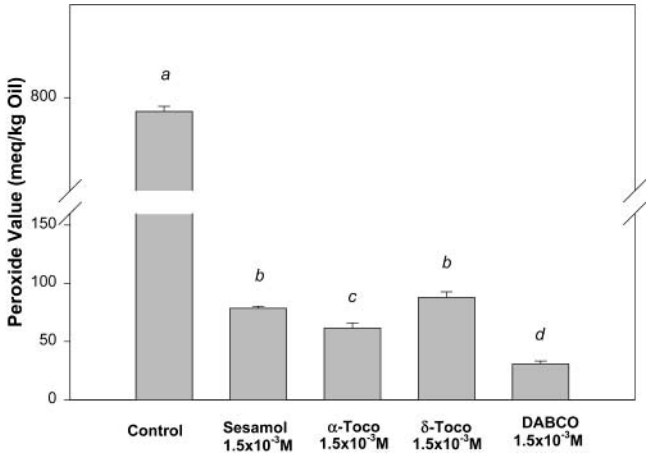


**Figure 3.** Comparison of antiphoto-oxidative activity of sesamol with those of α-tocopherol, δ-tocopherol, and DABCO on methylene blue-sensitized photo-oxidation of soybean oil in methylene chloride during 3 h of fluorescent light illumination (5500 lx) at 20 ± 1 °C. The methylene blue concentration in this experimental system was 4 μg/mL. The antioxidative activities with the same italicized letter at the top of the bar were not significantly different at $\alpha = 0.05$.

and 527 mequiv/kg of oil, respectively, indicating greatly different oxidation rates with different solvents. The samples protected from light with aluminum foil did not develop any peroxide during the 3 h storage under the same conditions (data not shown). Sesamol greatly reduced the methylene blue-sensitized photo-oxidation of soybean oil in all of the tested solvent systems (2-propanol, methylene chloride, and chloroform) during 3 h of fluorescence light illumination, and its effectiveness was dose-dependent. It is interesting to note that the antiphoto-oxidative activities of sesamol were greatly different with different solvent systems. The $0.5 \times 10^{-3}$ M sesamol treatment showed 2.5, 69.4, and 73.0% inhibitions of methylene blue-sensitized photo-oxidation of soybean oil in 2-propanol, methylene chloride, and chloroform, respectively. The reason for this is not clear at this point. With increase in treated sesamol concentration, significantly higher antiphoto-oxidative activity was obtained in all of the solvent systems. The inhibitions of methylene blue-sensitized photo-oxidation of soybean oil in methylene chloride by $0.5 \times 10^{-3}$, $1.0 \times 10^{-3}$, $1.5 \times 10^{-3}$, $2.0 \times 10^{-3}$, and $4.0 \times 10^{-3}$ M sesamol were 69.4, 84.7, 90.1, 92.0, and 96.2%, respectively (**Figure 2B**). The antiphoto-oxidative activity of sesamol on methlene blue-sensitized photo-oxidation of soybean oil in methylene chloride was compared with those of other well-known singlet oxygen quenchers (α- and δ-tocopherols and DABCO) (**Figure 3**). The addition of DABCO at $1.5 \times 10^{-3}$ M showed 96.1% inhibition of the methylene blue-sensitized photo-oxidation of soybean oil. It is well-known that DABCO, an effective singlet oxygen quencher, does not possess other radical scavenging properties. This result indicated that singlet oxygen oxidation was the main oxidation process involved in the reaction system used in this study. The results in **Figure 3** also show that the antiphoto-oxidative activity of sesamol in methylene blue-sensitized photo-oxidation of soybean oil was comparable to that of δ-tocopherol ($p > 0.05$) but lower than those of α-tocopherol and DABCO ($p < 0.05$). We also carried out an experiment to confirm the antiphoto-oxidative activity of sesamol during the methylene blue-sensitized photo-oxidation of soybean oil in methylene chloride by measuring the peroxides and conjugated dienes at several sampling points (**Figure 4**). The results clearly showed that sesamol effectively prevents the formation of not only

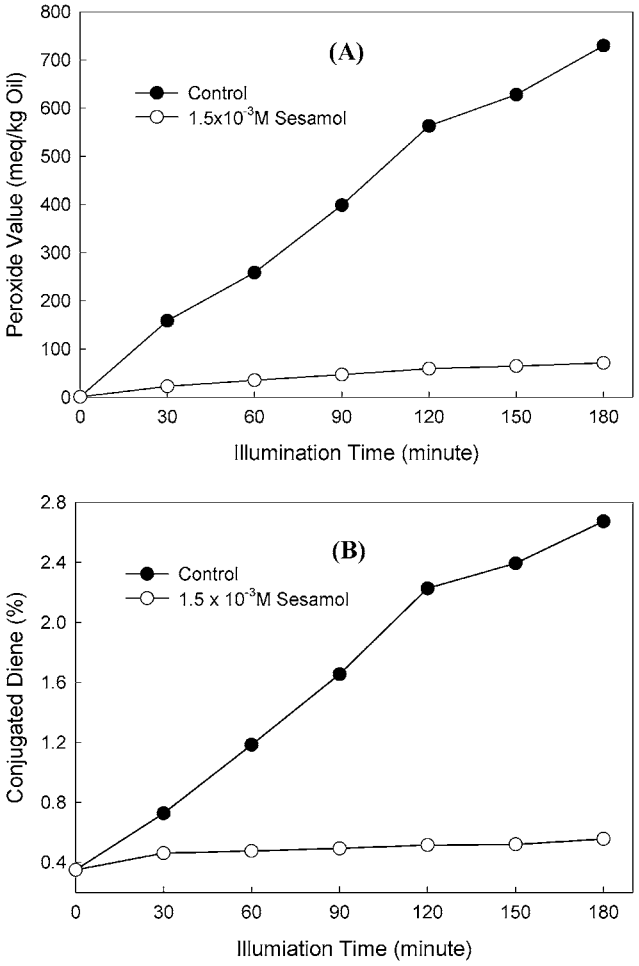


**Figure 4.** Effects of sesamol ($1.5 \times 10^{-3}$ M) on the formation of peroxides (A) and conjugated dienes (B) at different sampling points during the methylene blue-sensitized photo-oxidation of soybean oil in methylene chloride under the fluorescent light illumination (5500 lx) at 20 ± 1 °C. The methylene blue concentration in this experimental system was 4 μg/mL.

peroxides but also conjugated dienes as measured at several different sampling points. The $1.5 \times 10^{-3}$ M sesamol concentration showed inhibitions of 90.2% peroxide formation and 91.1% conjugated diene formation in soybean oil in methylene choride during 3 h of light illumination.

**Antiphoto-oxidative Activity of Sesamol in Chlorophyll-Sensitized Photo-oxidation of Soybean Oil.** We also chose another important sensitizer, chlorophyll *b*, to be used as a photosensitizer in our experiment to determine whether sesamol also reduces chlorophyll *b*-sensitized photo-oxidation of soybean oil (**Figure 5**). Sesamol also reduced the chlorophyll-sensitized photo-oxidation of soybean oil in methylene chloride with a dose-dependent manner. The $0.5 \times 10^{-3}$, $1.0 \times 10^{-3}$, $1.5 \times 10^{-3}$, $2.0 \times 10^{-3}$, and $4.0 \times 10^{-3}$ M sesamol treatments showed 33.9, 43.4, 52.4, 55.4, and 64.9% inhibitions of chlorophyll-sensitized photo-oxidation of soybean oil, respectively (**Figure 5A**). Sesamol was much less effective in the chlorophyll-sensitized photo-oxidation of soybean oil than in methylene blue-sensitized photo-oxidation of soybean oil. The effectiveness of sesamol on chlorophyll-sensitized photo-oxidation of soybean oil was lower than that of α-tocopherol ($p < 0.05$) but comparable to that of δ-tocopherol at the same molar concentration ($p > 0.05$) (**Figure 5B**). In the chlorophyll-sensitized photo-oxidation, DABCO was significantly less effective than sesamol at the same molar concentration ($p < 0.05$). Note that DABCO

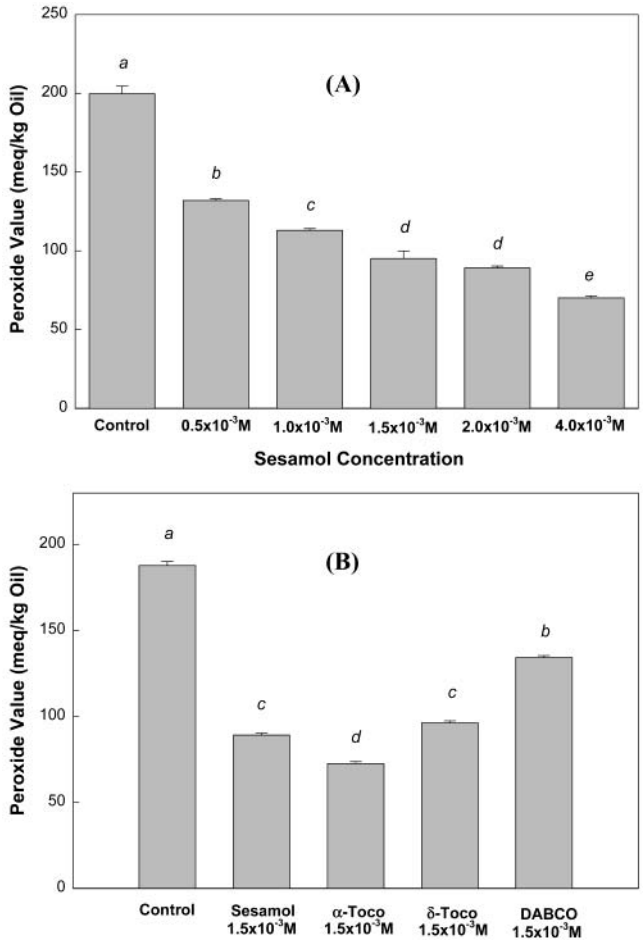


**Figure 5.** Antiphoto-oxidative activity of different concentrations of sesamol (A) and comparison of its activity with those of α-tocopherol, δ-tocopherol, and DABCO (B) on chlorophyll-sensitized photo-oxidation of soybean oil in methylene chloride during 3 h of fluorescent light illumination (5500 lx) at 20 ± 1 °C. The chlorophyll concentration in this experimental system was 4 μg/mL. The antioxidative activities with the same italicized letter at the top of the bar were not significantly different at $\alpha = 0.05$.

was significantly more effective than sesamol, at the same molar concentration, in methylene blue-sensitized photo-oxidation of oil (**Figure 3**). Even though sesamol has been known to have a strong antioxidative activity in the autoxidation of oils (*29*), this is the first report on the antioxidative activity of sesamol in the sensitized photo-oxidation of oil.

**Proposed Mechanism for Antiphoto-oxidative Activity of Sesamol.** Methylene blue and chlorophyll *b* are efficient photosensitizers for the generation of sinlget oxygen in the presence of light (*6*, *11*). To explore the preventive mechanism of sesamol in the photo-oxidation of oil, we studied the inhibitory activity of sesamol on rubrene oxidation induced by chemical source of singlet molecular oxygen in a microemulsion system (*26*, *30*). The water/oil microemulsion used in the present work was water microdroplets coated by an interfacial film of SDS and 1-butanol dispersed in a continuous phase of methylene chloride. Hydrogen peroxide and sodium molybdate were compartmentalized in an aqueous microreactor, where they generated $^1O_2$. Rubrene was localized in a continuous phase of methylene chloride. Because $^1O_2$ is a small and uncharged molecule, it can diffuse freely through the charged (coming from the anionic surfactant SDS) interfacial region (*26*). Rubrene (red color) is selectively oxidized with singlet oxygen but not with other oxygen species to produce its cycloendoperoxide (colorless). **Figure 6** shows the effects of sesamol on the red color fading of rubrene with chemically induced singlet oxygen in the microemulsion. The red color of rubrene was almost gone after 5 min, indicating the complete oxidation of rubrene. However, $0.5 \times 10^{-2}$ and $1.5 \times 10^{-2}$ M sesamol treatments inhibited the singlet oxygen-induced color fading of rubrene in the microemulsion. Treatments with $0.5 \times 10^{-2}$ and $1.5 \times 10^{-2}$ M sesamol showed 18.7 and 47.6% inhibitions of rubrene oxidation after 5 min of reaction as determined by spectrophotometry at 522 nm (**Figure 6**). This result clearly indicated that sesamol possessed singlet oxygen quenching activity. This result also suggested that the inhibition of oil photo-oxidation by sesamol was, at least in part, due to its singlet oxygen quenching ability.

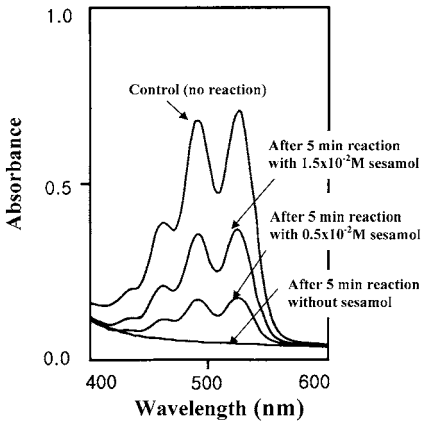


**Figure 6.** Visible spectra for rubrene oxidation in microemulsion containing 0, $0.5 \times 10^{-2}$, or $1.5 \times 10^{-2}$ M sesamol with singlet oxygen generated from the system $H_2O_2/MoO_4^{2-}$.

**Determination of Singlet Oxygen Quenching Rate Constant ($k_{ox-Q} + k_q$).** The singlet oxygen quenching rate constant of sesamol was determined according to a previously reported technique that was based on inhibition of the self-sensitized photo-oxidation of rubrene. In this reaction system, when a solution of rubrene is illuminated in the presence of oxygen and singlet oxygen quencher, the following reactions take place:

$$R + h\nu \rightarrow R^1 \quad (1)$$

$$R^1 + O_2 \xrightarrow{k_s} R^3 + {}^1O_2 \quad (2)$$

$$R^3 + {}^1O_2 \xrightarrow{k_t} R + {}^1O_2 \quad (3)$$

$${}^1O_2 \xrightarrow{k_d} {}^3O_2 \quad (4)$$

$${}^1O_2 + R \xrightarrow{k_{ox}} RO_2 \quad (5)$$

$${}^1O_2 + Q \xrightarrow{k_{ox-Q} + k_q} {}^3O_2 + Q \text{ (and/or } QO_2) \quad (6)$$

R is rubrene, $R^1$ is the rubrene singlet, $R^3$ is the rubrene triplet, $RO_2$ is the rubrene photoperoxide, Q is the quencher, and $QO_2$ is the oxidation product of the quencher. Note that rubrene is both the sensitizer and $^1O_2$ acceptor.

If a quencher is present, $^1O_2$ disappears by three routes: nonradiative decay (reaction 4), reaction with rubrene (reaction 5), and singlet oxygen quenching (reaction 6). In the absence of a quencher, $^1O_2$ disappears by only two routes: nonradiative decay and reaction with rubrene. If two solutions of equal volume, one containing quencher and one without quencher and

**Table 1.** Effects of $0.5 \times 10^{-3}$ M Sesamol on the Self-Sensitized Photo-oxidation of $0.3 \times 10^{-3}$ M Rubrene in Chloroform after 2 min of Illumination

| | test 1 | test 2 | test 3 |
|---|---|---|---|
| [R] (M) | $0.3 \times 10^{-3}$ | $0.3 \times 10^{-3}$ | $0.3 \times 10^{-3}$ |
| [Q] (M) | $0.5 \times 10^{-3}$ | $0.5 \times 10^{-3}$ | $0.5 \times 10^{-3}$ |
| $[R]_F^0$ (M) | $0.181 \times 10^{-3}$ | $0.178 \times 10^{-3}$ | $0.205 \times 10^{-3}$ |
| $[R]_F^Q$ (M) | $0.204 \times 10^{-3}$ | $0.207 \times 10^{-3}$ | $0.182 \times 10^{-3}$ |
| $k_{ox-Q} + k_q$ | $1.67 \times 10^7$ | $2.21 \times 10^7$ | $1.71 \times 10^7$ |
| mean $k_{ox-Q} + k_q$ ($M^{-1}$ $s^{-1}$) | | $1.9 \pm 0.3 \times 10^7$ | |

**Table 2.** Effects of $0.25 \times 10^{-3}$ M DABCO on the Self-Sensitized Photo-oxidation of $0.3 \times 10^{-3}$ M Rubrene in Chloroform after 2 min of Illumination

| | test 1 | test 2 | test 3 |
|---|---|---|---|
| [R] (M) | $0.3 \times 10^{-3}$ | $0.3 \times 10^{-3}$ | $0.3 \times 10^{-3}$ |
| [Q] (M) | $0.25 \times 10^{-3}$ | $0.25 \times 10^{-3}$ | $0.25 \times 10^{-3}$ |
| $[R]_F^0$ (M) | $0.182 \times 10^{-3}$ | $0.184 \times 10^{-3}$ | $0.80 \times 10^{-3}$ |
| $[R]_F^Q$ (M) | $0.215 \times 10^{-3}$ | $0.214 \times 10^{-3}$ | $0.215 \times 10^{-3}$ |
| $k_{ox-Q} + k_q$ | $5.50 \times 10^7$ | $4.93 \times 10^7$ | $5.58 \times 10^7$ |
| mean $k_{ox-Q} + k_q$ ($M^{-1}$ $s^{-1}$) | | $5.4 \pm 0.5 \times 10^7$ | |

each having the same initial concentration of rubrene, are each exposed to the same amount of $^1O_2$, the singlet oxygen quenching rate constant ($k_{ox-Q} + k_q$) can be calculated from the following equation (*27*, *28*):

$$k_{ox-Q} + k_{ox} = \frac{([R]_F^Q - [R]_F^0) + k_d \ln ([R]_F^Q/[R]_F^0)}{[Q] \ln ([R]/[R]_F^Q)} \quad (7)$$

[R] is the initial concentration of rubrene, $[R]_F^Q$ the final concentration of rubrene in the quenched solution, $[R]_F^0$ the final concentration of rubrene in the unquenched solution, and [Q] the quencher concentration.

It is seen that the singlet oxygen rate constant ($k_{ox-Q} + k_q$) can be calculated from four experimentally determinable quantities and two rate constants. Both the lifetime of $^1O_2$ ($1/k_d$) and the rate of addition of $^1O_2$ to rubrene ($k_{ox}$) in a number of solvents can be obtained from previous papers. We adapted $k_{ox} = 5.3 \times 10^7$ $M^{-1}$ $s^{-1}$ and $k_d = 1.7 \times 10^4$ $s^{-1}$ from the literature (*27*, *31*). The initial rubrene content ([R]) used in this experiment was $0.3 \times 10^{-3}$ M. The sesamol concentration [Q] used in this experiment was $0.5 \times 10^{-3}$ M. The rubrene concentrations with sesamol ($[R]_F^Q$) and without sesamol ($[R]_F^0$) after 2 min of illumination are shown in **Table 1**. With the eq 7, the singlet oxygen quenching rate constant ($k_{ox-Q} + k_q$) of sesamol was calculated. The obtained mean $k_{ox-Q} + k_q$ of sesamol was $1.9 \pm 0.3 \times 10^7$ $M^{-1}$ $s^{-1}$. To check the accuracy of our kinetic value, we also determined $k_{ox-Q} + k_q$ of DABCO, a well-known singlet oxygen quencher. The rubrene concentrations with DABCO ($[R]_F^Q$) and without DABCO ($[R]_F^0$) after 2 min of illumination are shown in **Table 2**. The calculated $k_{ox-Q} + k_q$ of DABCO in chloroform was $5.4 \pm 0.5 \times 10^7$ $M^{-1}$ $s^{-1}$. The $k_{ox-Q} + k_q$ value for DABCO is in good agreement with the previously reported value of $5.2 \pm 0.4 \times 10^7$ $M^{-1}$ $s^{-1}$ in chloroform (*27*). The result indicated that the technique used in this research for the determination of the singlet oxygen quenching rate constant of sesamol was acceptable. The singlet oxygen rate constant ($1.9 \times 10^7$ $M^{-1}$ $s^{-1}$) of sesamol was slightly lower than our previously reported value for α-tocopherol ($2.6 \times 10^7$ $M^{-1}$ $s^{-1}$) (*18*). The singlet oxygen quenching abilities of the compounds seemed to be consistent with their antiphoto-oxidative effects in the methylene blue-sensitized photo-oxidation of oil (**Figure 3**). However, the singlet oxygen quenching ability values were not well correlated with their antiphoto-oxidative activities in the chlorophyll-sensitized photo-oxidation of oil as shown in **Figure 5B**. Note that the antiphoto-oxidative activity of sesamol with a lower singlet oxygen quenching ability was higher than that of DABCO with a higher singlet oxygen quenching ability in the chlorophyll-sensitized photo-oxidation of oil (**Figure 5B**). The result indicated that methylene blue and chlorophyll induced different routes of photo-oxidation mechanism. Nevertheless, the present research showed for the first time the antiphoto-oxidative activity and singlet oxygen quenching ability of sesamol.

In brief summary, sesamol possesses a strong antiphoto-oxidative activity in sensitized photo-oxidation of oil. The antiphoto-oxidative property of sesamol was found to be due to its singlet oxygen quenching ability. The singlet oxygen quenching rate constant of sesamol was $1.9 \times 10^7$ $M^{-1}$ $s^{-1}$. The present research showed for the first time the antiphoto-oxidative activity and singlet oxygen quenching rate constant of sesamol in the photosensitized oxidation of oils. On the basis of the present results, it is expected that sesamol could be applied to oils and oil-containing foods to prevent the photo-oxidation of various oils, oil-soluble vitamins, and other oil-soluble components (cholesterol, limonene, conjugated terpenes, etc.).

## LITERATURE CITED


(1) Jung, M. Y.; Lee, K. H.; Kim, S. Y. Retinyl palmitate isomers in skim milk during light storage as affected by ascorbic acid. *J. Food Sci.* **1998**, *63*, 597−600.

(2) Jung, M. Y.; Yoon, S. H.; Lee, H. O.; Min, D. B. Singlet oxygen and ascorbic acid effects on the dimethyl disulfide and off-flavor formation in skim milk exposed to light. *J. Food Sci.* **1998**, *63*, 408−412.

(3) Jung, M. Y.; Kim, J. P.; Kim, S. Y. Methanolic extract of *Coptis japonica* Makino reduces the chlorophyll-sensitized photooxidation of oils. *Food Chem.* **1999**, *67*, 261−268.

(4) Kim, J. S.; Chung, M. S.; Shin, H. S. Oxidative effects of animal-originated porphyrins and riboflavin on cholesterol oxidation in an aqueous model system. *J. Am. Oil Chem. Soc.* **2001**, 78, 1209−1212.

(5) Kim, G. Y.; Lee, J. H.; Min, D. B. Study of light-induced volatile compounds in goat's milk cheese. *J. Agric. Food Chem.* **2003**, *51*, 1405−1409.

(6) Koryck-Dahl, M. B.; Richardson, T. Activated oxygen species and oxidation of food constituents. In *Critical Reviews in Food Science and Nutrition*; CRC Press: Boca Raton, FL, 1978; pp 209−241.

(7) King, J. M.; Min, D. B. Riboflavin-photosensitized singlet oxygen oxidation of vitamin D. *J. Food Sci.* **1998**, *63*, 31−34.

(8) King, J. M.: Min, D. B. Riboflavin-photosensitized singlet oxygen oxidation product of vitamin D2. *J. Am. Oil Chem. Soc.* **2002**, *79*, 983−987.

(9) Berliner, L. J.; Bradley, D. G.; Meinheltz, D.; Min, D. B.; Ogata, T. The detection of riboflavin-photosensitized singlet oxygen formation in milk by electron spin resonance. In *Supramolecular Structure and Function*; Pifat, G., Ed.; Balaban Publishers: Croatia, 1994; pp 67−77.

(10) Bradley, D. G.; Min, D. B. Singlet oxygen oxidation of foods. *Crit. Rev. Food Sci. Nutr.* **1992**, *31*, 211−236.

(11) Whang, K.; Peng, I. C. Detection of singlet oxygen generation by chlorophyll using electron paramagnetic resonance spectroscopy. *J. Food Sci.* **1988**, *53*, 1918−1919.

(12) Whang, K.; Peng, I. C. Electron paramagnetic resonance studies of the effectiveness of myoglobin and its derivative as photosensitizers in singlet oxygen generation. *J. Food Sci.* **1988**, *53*, 1863−1865.

(13) Yang, W. T.; Lee, J. H.; Min, D. B. Quenching mechanism and kinetics of α-tocopherol and β-carotene on the photosensitizing effects of synthetic food colorant FD&C Red No. 3. *J. Food Sci.* **2002**, *67*, 507−510.
(14) Rawls, H. R.; van Santen, P. J. A possible role for singlet oxygen in the initiation of fatty acid autooxidation. *J. Am. Oil Chem. Soc*. **1970**, *47*, 121−125.
(15) Clements, A. H.; Van Den Engh, R. H.; Frost, D. J.; Hoogenhout, K. Participation of singlet oxygen in photosensitized oxidation of 1,4-dienoic systems and photooxidation of soybean oil. *J. Am. Oil Chem. Soc*. **1973**, *50*, 325−330.
(16) Carlsson, D. J.; Suprunchuk, T.; Siles, D. M. Photooxidation of unsaturated oils: Effects of singlet oxygen quenchers. *J. Am. Oil Chem. Soc.* **1976**, *53*, 656−660.
(17) Chan, H. W. S. Photosensitized oxidation of unsaturated fatty acid methyl esters. The identification of different pathways. *J. Am. Oil Chem. Soc*. **1977**, *54*, 100−104.
(18) Jung, M. Y.; Choe, E.; Min, D. B. α-, γ- and δ-tocopherol effects on chlorophyll photosensitized oxidation of soybean oil. *J. Food Sci.* **1991**, *56*, 807−810 and 815.
(19) Jung, M. Y.; Kim, S, K.; Kim, S. Y. Riboflavin-sensitized photooxidation of ascorbic acid: kinetics and amino acid effects. *Food Chem*. **1995**, *53*, 397−403.
(20) Jung, M. Y.; Lee, K. H.; Kim, S. Y. Riboflavin-sensitized photochemical changes in lactoglobulin in an aqueous solution as affected by ascorbic acid. *J. Agric. Food Chem.* **2001**, *48*, 3849−3850.
(21) Jung, M. Y.; Min, D. B. Effects of quenching mechanisms of carotenoids on the photosensitized oxidation of soybean oil. *J. Am. Oil Chem. Soc.* **1991**, *68*, 563−568.
(22) Kim, J. P.; Jung, M. Y.; Kim, J. P.; Kim, S. Y. Antiphotooxidative activity of protoberberine derived from *Coptis japonica* Makino in the chlorophyll-sensitized photooxidation of oil. *J. Agric. Food Chem*. **2000**, *48*, 1058−1063.
(23) Lee, K. H.; Jung, M. Y.; Kim, S. Y. Quenching mechanism and kinetics of ascorbyl palmitate for the reduction of the photosensitized oxidation of oils. *J. Am. Oil Chem. Soc*. **1997**, *74*, 1053−1057.
(24) Li, T. L.; King, J. M.; Min, D. B. Quenching mechanisms and kinetics of carotenoids in riboflavin photosensitized singlet oxygen oxidation of vitamin D. *J. Food Biochem*. **2000**, *24*, 477−492.
(25) AOCS. *Official Methods and Recommended of the American Oil Chemists' Society*, 4th ed.; Firestone, D., Ed.; American Oil Chemists' Society: Champaign, IL, 1990.
(26) Nardello, V. N.; Matini, M.; Pierlot, C.; Aubry, J. Photochemistry without light: Oxidation of rubrene in a microemulsion with a chemical source of singlet molecular oxygen ($^1O_2$, $^1\Delta_g$). *J. Chem. Educ.* **1999**, *76*, 1285−1288.
(27) Monroe, B. M. Quenching of singlet oxygen by aliphatic amines. *J. Phys. Chem*. **1977**, *81*, 1861−1864.
(28) Carlsson, D. J.; Mendenhall, G. D.; Suprunchuk, T.; Siles, D. M. Singlet oxygen quenching in the liquid phase by metal(II) chelates. *J. Am. Chem. Soc.* **1972**, *94*, 8160−8162.
(29) Kikugawa, K.; Arai, M.; Kurechi, T. Participation of sesamol in stability of sesame oil. *J. Am. Oil Chem. Soc*. **1983**, *60*, 1528−1533.
(30) Nardello, V. N.; Markino, J.; Vermeersch, G.; Aubry, J. M. $^{95}$-Mo NMR and kinetic studies of peroxomolybdic intermediates involved in the catalytic disproportionation of hydrogen peroxide by molybdate ions. *Inorg. Chem*. **1995**, *34*, 4950−4957.
(31) Merkel, P. B.; Kearns, D. R. Radiationless decay of singlet molecular oxygen in solution: An experimental and theoretical study of electronic-to-vibrational energy transfer. *J. Am. Chem. Soc.* **1972**, *94*, 7244−7253.

**Received for review October 23, 2002. Revised manuscript received March 11, 2003. Accepted March 27, 2003. This research was supported by Woosuk University.**

JF026056P