

Reactivity on Photooxidation of Selected Five-Membered Heterocyclic Flavor Compounds

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Conjugated five-membered heterocyclic compounds including 2-ethylfuran, 2-furfural, 2-acetylfuran, 5-(hydroxymethyl)-2-furfural (HMF), 2-ethylthiophene, 2-formylthiophene, 2-acetylthiophene, 2-ethylpyrrole, 2-formylpyrrole, 2-acetylpyrrole, 2,4,5-trimethyloxazole, and 2,4,5-trimethylthiazole in absolute alcohol containing chlorophylls were bubbled with oxygen under light for 1 h. The reactivity of these heterocyclic compounds with singlet oxygen was studied using a high-performance liquid chromatographic method, in which the percentage of substrate remaining was determined. The photooxidation reactivity of these selected five-membered flavor compounds was as follows: 2-ethylpyrrole > 2-ethylfuran > 2-ethylthiophene; 2,4,5-trimethyloxazole > 2,4,5-trimethylthiazole; 2-ethylfuran > HMF > 2-acetylfuran > 2-furfural; 2-ethylpyrrole > 2-acetylpyrrole > 2-formylpyrrole. The electron density at C-1 and C-4 may explain the photooxidation reactivity of these five-membered heterocyclic compounds.

Keywords: Five-membered heterocyclic compounds; singlet oxygen; photooxidation reactivity; chlorophyll

INTRODUCTION

The photooxidation reaction, which occurs in the presence of light, triplet oxygen, and photosensitizers, is the most common way to generate singlet oxygen in foods. Chlorophylls, rose bengal, and methylene blue are efficient photochemical sensitizers used to generate singlet oxygen (Matsuura and Saito, 1976; Mensah, 1984). Singlet oxygen easily initiates the photooxidation of flavor or lipid compounds because of its low energy of only 22.4 kcal above the ground state, its relatively long lifetime, and its highly electrophilic nature which allows easy attack of moieties of high electron density (Bradley and Min, 1992; Rawls and Van Santen, 1970).

The reaction of singlet oxygen with organic compounds has been studied and may be classified into five categories including 1,4-addition to conjugated dienes, 1,2-addition to electron-rich olefins, the ene reaction with olefins, the oxidation of sulfides to sulfoxides, and the photosensitized oxidation of phenols to unstable hydroxydienes (Chan, 1977; Corey and Taylor, 1964; Matsuura and Saito, 1976; Thomas and Foote, 1978). The flavor compounds which contain conjugated double bonds as part of homo- and heterocyclic ring systems undergo 1,4-cycloaddition reaction when the singlet oxygen reacts with these compounds. In this reaction, singlet oxygen acts as a dienophile and adds to conjugated dienes, resulting in six-membered or 1,4-endo-peroxide compound formation (Matsuura and Saito, 1976). These peroxides are extremely unstable and undergo decomposition reactions to yield different kinds of products depending on the solvent system, temperature, or other conditions. At room temperature, the solvent methanol can participate in this reaction, resulting in methoxy compound formation (Foote, 1967; Matsuura and Saito, 1976; Mensah et al., 1986; Wasserman and Strehlow, 1970).

The mechanisms of the photosensitized oxidation of heterocyclic compounds are known. However, the reactivity between these compounds and singlet oxygen has not been well studied. The purpose of this research was to investigate the reactivity of singlet oxygen with selected five-membered heterocyclic flavor compounds including furan, pyrrole, thiophene, oxazole, and thiazole compounds. The present study reports how the different chemical structures affect the reactivity of photosensitized oxidation.

EXPERIMENTAL PROCEDURES

Materials. 2-Formylpyrrole and 5-(hydroxymethyl)-2-furfural (HMF) were obtained from Sigma Chemical Co. (St. Louis, MO). 2-Ethylpyrrole, 2-acetylpyrrole, 2-ethylfuran, 2-furfural, 2-acetylfuran, 2-ethylthiophene, 2-formylthiophene, 2-acetylthiophene, 2,4,5-trimethyloxazole, and 2,4,5-trimethylthiazole were obtained from Aldrich Chemical Co. (Milwaukee, WI). Structures of the above compounds are shown in Figure 1. The chlorophylls used as a photosensitizer were prepared and purified in our laboratory from spinach according to the procedure described by Omata and Murata (1980). Absolute alcohol as a solvent was purchased from Pharmco Products Co. (Brookfield, CT).

Photooxidation of Five-Membered Heterocyclic Compounds and Detection of Five-Membered Heterocyclic Compound Residues. Solutions (1×10^{-3} M) of the five-membered heterocyclic compounds to be tested (Figure 1) were prepared in 100 mL of absolute alcohol containing 10 ppm of chlorophylls in a 250 mL of flask as shown in Figure 2. This reaction flask was placed in a beaker that contained water. The water temperature was kept at 15–20 °C. Two 150 W Philips projector lamps were used as the light source. The distance from the light source to the reaction flask was 20 cm. The photooxidation reaction of these five-membered heterocyclic compounds was carried out under the light and while bubbling with oxygen at 260 mL/min for 1 h. The condenser on the top of the reaction flask was used to keep the solvent from evaporating while the oxygen supply was being applied. The reactivity of singlet oxygen with these compounds was measured using a high-performance liquid chromatographic (HPLC) method in which the percentage of substrate remaining was calculated by dividing the peak area of substrate by that of internal standard.

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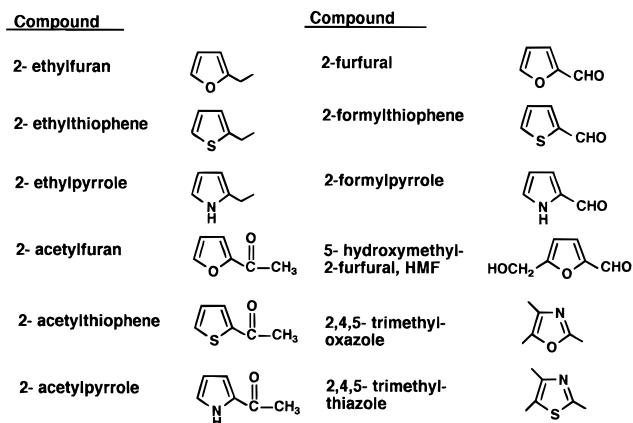


Figure 1. Structures of the furan, thiophene, pyrrole, oxazole, and thiazole compounds.

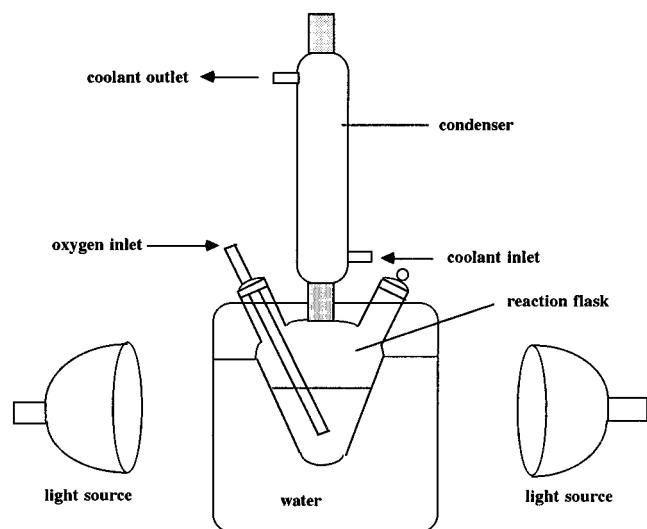


Figure 2. Apparatus for the photooxidation of flavor compounds.

HPLC analysis was performed using a Varian 5000 liquid chromatograph equipped with a Varian 2050 detector at 265 nm for detection of 2-formylthiophene, 2-acetylthiophene, 2-furfural, and 2-acetylfuran, at 285 nm for detection of 2-formylpyrrole, 2-acetylpyrrole, and HMF, at 230 nm for detection of 2-ethylthiophene and 2,4,5-trimethyloxazole, at 220 nm for detection of 2-ethylfuran and 2-ethylpyrrole, and at 254 nm for detection of 2,4,5-trimethylthiazole. A Whatman Partisphere 5 ODS-2 reversed phase column (10 cm \times 0.49 cm i.d.) was used. The mobile phase was 35% methanol solution with 0.5% acetic acid and was pumped at a flow rate of 1.5 mL/min isocratically.

RESULTS AND DISCUSSION

Twelve five-membered heterocyclic flavor compounds including four furans (2-ethylfuran, 2-furfural, 2-acetylfuran, and HMF), three thiophenes (2-ethylthiophene, 2-formylthiophene, and 2-acetylthiophene), three pyrroles (2-ethylpyrrole, 2-formylpyrrole, and 2-acetylpyrrole), 2,4,5-trimethyloxazole, and 2,4,5-trimethylthiazole were separately dissolved in absolute alcohol containing chlorophylls and bubbled with oxygen under light for 1 h. The HPLC method was used to determine the percentage of substrate remaining during the photooxidation.

The reactivity of singlet oxygen with 2-ethylthiophene, 2-ethylpyrrole, and 2-ethylfuran is shown in Figure 3A. The results showed that the reactivity of singlet oxygen with 2-ethylpyrrole and 2-ethylfuran was

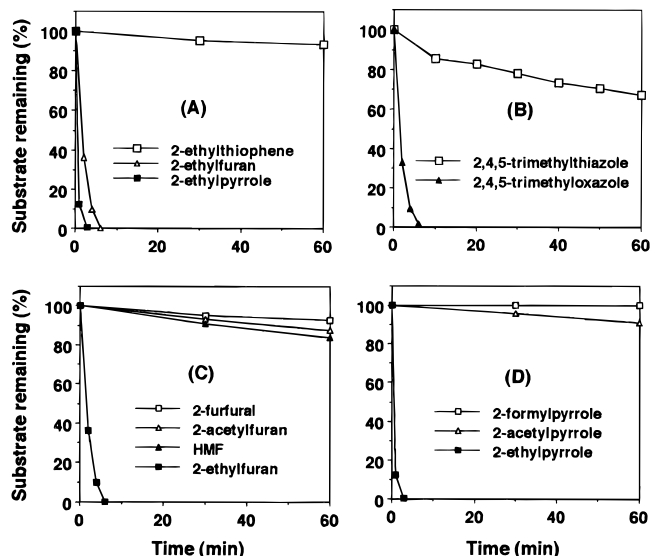


Figure 3. Stability of selected five-membered heterocyclic compounds in absolute alcohol containing 10 ppm of chlorophylls under light and bubbling with oxygen.

so fast that most of these substrates were oxidized within 5 min. However, there was little photooxidation effect on 2-ethylthiophene in 1 h. As shown in Figure 3A, the photooxidation reactivity of these three compounds was in the order 2-ethylpyrrole > 2-ethylfuran > 2-ethylthiophene. It has been reported that thiophenes, unlike the corresponding furans and pyrroles, are unreactive toward photosensitized oxidation. However, when thiophene is substituted with alkyl groups at both the C-1 and C-4 positions, it does react to yield oxidation products (Matsuura and Saito, 1976; Skold and Schlessinger, 1970; Wasserman and Strehlow, 1970). The results of our study showed that the photooxidation reactivity of 2-ethylthiophene, which has only one alkyl group attached at the C-1 position, was very slow compared to the reactivity of 2-ethylpyrrole and 2-ethylfuran (Figure 3A).

It is well-known that the reaction of singlet oxygen with conjugated five-membered ring compounds is via a 1,4-cycloaddition reaction (Matsuura and Saito, 1976). Singlet oxygen with a vacant molecular orbital is highly electrophilic and readily seeks electrons to fill this orbital (Bradley and Min, 1992). Therefore, the conjugated five-membered ring compounds that contain high electron density at the C-1 and C-4 positions can easily react with singlet oxygen. The π -electron densities of pyrrole, furan, and thiophene at the C-1 or C-4 position were 1.084, 1.067, and 0.912, respectively (Clark, 1968a,b; Newkome and Paudler, 1982a). The order of photooxidation reactivity (2-ethylpyrrole > 2-ethylfuran > 2-ethylthiophene) is in agreement with these π -electron density calculations. Newkome and Paudler (1982b) also indicated that the electrophilic reaction of cyclic five-membered ring compounds depends on the atom in this ring structure. Generally, the more electronegative heteroatoms such as nitrogen and oxygen, for which d-orbital participation is not possible, facilitate these reactions more significantly than do those heteroatoms such as sulfur, for which d-orbital participation is possible.

Figure 3B shows that the 2,4,5-trimethyloxazole had higher photooxidation reactivity than 2,4,5-trimethylthiazole. π -Electron density calculations or electronegative atom participation explained as above may be used to elucidate this result. The photooxidation reac-

tivity of 2,4,5-trimethylthiazole as shown in Figure 3B was higher than that of 2-ethylthiophene (Figure 3A). In the structure of 2,4,5-trimethylthiazole (Figure 1), there are two alkyl groups separately located at C-1 and C-4 positions that make it possible to undergo the photooxidation reaction (Matsuura and Saito, 1976), and in addition to a sulfur atom, there is participation of one nitrogen atom in this ring structure that increases its electrophilic substitution ability (Newkome and Paudler, 1982b).

The substituent effect on the photooxidation reactivity is decreasingly reactive in the order 2-ethylfuran > HMF > 2-acetylfuran > 2-furfural for furans (Figure 3C) and 2-ethylpyrrole > 2-acetylpyrrole > 2-formylpyrrole for pyrroles (Figure 3D). In general, the more electron density at the C-1 or C-4 position of conjugated five-membered ring compounds, the higher the photooxidation reactivity the compounds possess (Mensah, 1984). Therefore, the presence of electron-donating groups at C-1 or C-4 tends to increase the reactivity, while electron-withdrawing substituents at C-1 or C-4 tend to slow the reactivity. Thus, 2-ethylfuran and 2-ethylpyrrole, which have one electron-donating ethyl group attached to C-1, showed highest reactivity of the furans and pyrroles, respectively (Figure 3C,D). The lowest reactivity of the furans and pyrroles was observed for 2-furfural and 2-formylpyrrole, which contain an electron-withdrawing carbonyl group in the C-1 position. The reactivities of 2-acetylfuran and 2-acetylpyrrole were slightly higher than those of 2-furfural and 2-formylpyrrole, respectively, because the electron-donating methyl group which is bonded to this carbonyl group increases the electron density at the C-1 position. HMF showed higher photooxidation reactivity than 2-acetylfuran. This may be due to the methyl group that is attached directly to C-4 instead of bonded to the carbonyl group at the C-1 position. This results in an increase of the overall electron density at the C-4 position.

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