lowing bromine atoms anisotropic and carbon atoms isotropic vibration. Hydrogen atoms were not located. Atomic fractional coordinates are given in Table III.

Synthesis of 2,3,6,7-Tetrabromonaphthalene. To an ice-cooled solution of 624 mg (1.5 mmol) of 2,3,6,7-tetrakis(trimethylsilyl)naphthalene (Funk and Vollhardt, 1976) in 5 mL of CCl_4 and 250 μL of pyridine was added 14 mL of 1.9 M Br_2 in CCl_4 solution. The reaction mixture was stirred for 10 h. The course of the reaction was followed by GLC (OV-101; 250 °C). The first-formed product was the dibromobis(trimethylsilyl) derivative, the structure of which was assigned by mass spectral analysis. The next product to appear was the desired tetrabromonaphthalene. Aqueous $Na_2S_2O_3$ was added to remove excess bromine and the reaction mixture extracted twice with ether. The organic phase was dried and evaporated to yield 320 mg of product. One recrystallization from benzene yield 235 mg of 2,3,6,7-tetrabromonaphthalene, mp 259–261 °C, with the expected NMR and MS spectral properties [for X-ray structural work, see Singh et al. (1980)].

Synthesis of 1,2,3,4,6,7-Hexabromonaphthalene. To a solution of 624 mg (5.0 mmol) of naphthalene in 25 mL of dibromoethane and 2.0 mL of liquid bromine was added 40 mg of iron powder. The reaction mixture was refluxed for 4 h with stirring. After the mixture was cooled at room temperature, the crude product was isolated by suction filtration and dried in vacuo. One crystallation from hot dibromoethane afforded chromatographically (GLC; 3% OV-101) pure 1,2,3,4,6,7-hexabromonaphthalene, mp 260–263 °C, with the expected NMR and MS spectral properties.

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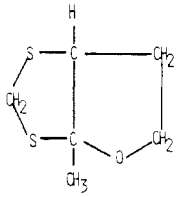
Odor Threshold of Thiamin Odor Compound 1-Methylbicyclo[3.3.0]-2,4-dithia-8-oxaoctane

An odor threshold of 4 parts of compound per 10^{13} parts of water was found for the thiamin odor compound (1-methylbicyclo[3.3.0]-2,4-dithia-8-oxaoctane or 2,3-methylenedithio-2-methyltetrahydrofuran, I) believed to be responsible for the characteristic odor of thiamin hydrochloride (vitamin B_1) preparations. This threshold is among the lowest ever reported for any odor compound in water solution. A possible mechanism pathway for the formation of the thiamin odor compound from thiamin is proposed.

Some of the authors (Seifert et al., 1978) had already reported on the determination of the structure of the thiamin odor compound as 1-methylbicyclo[3.3.0]-2,4-dithia-8-oxaoctane (I, Figure 1) which they had found from the photolysis of thiamin hydrochloride (vitamin B_1). This compound appeared to be responsible for the characteristic aroma of commercial thiamin and multivitamin preparations. The structure was recently confirmed by its synthesis from simple starting materials (Gygax, 1979).

The present study was undertaken because of the need



I

Figure 1. Structure of thiamin odor compound.

Table I. Threshold Determination of Thiamin Odor Compound (I)

concn, mL of I/ 10^{12} mL of water	% correct judgements	total no. of judgements
10^6	100	16
10^3	100	16
10^2	98	49
25	98	47
5	100	28
2	97	58
1	93	70
0.5	81	91
0.25	65	91
0.125	47	91
0.0625	50	49

for data on the effectiveness of this compound as an odorant in vitamin preparations and in foods which contain natural thiamin.

EXPERIMENTAL SECTION

Materials. The thiamin odor compound (I) was prepared by the ultraviolet light (UV) irradiation of thiamin hydrochloride and purified by gas–liquid chromatography separation as described previously (Seifert et al., 1978).

Odor Threshold Determination. This was carried out by using the procedure described by Guadagni et al. (1963) and Guadagni and Buttery (1978) using odor-free Teflon squeeze bottles equipped with Teflon tubes.

RESULTS AND DISCUSSION

The odor threshold of I was determined by a trained panel consisting of 16 judges. As in previous threshold determinations (Guadagni et al., 1963), the odor judges were presented with two Teflon squeeze bottles, one containing the solution and the other the odor-free water. The task for each judge was to determine which of the coded bottles contained the odorant. Table I lists the results obtained. Plotting this data as outlined by Guadagni et al. (1973) gives a threshold of 4 parts (mL) per 10^{13} parts (mL) of water. To the authors' knowledge, this is one of the lowest thresholds in water solution determined by reliable panel methods. It is lower than that found for the aroma compound of bell peppers, 2-isobutyl-3-methoxypyrazine, and related compounds (Seifert et al., 1970). Some lower thresholds have been reported in the literature, but there is some question whether these are correct. The compound 2,3,6-trichloroanisole had been reported to have an odor threshold of 3 parts per 10^{16} parts of water (Curtis et al., 1972), but when this was repeated with careful panel methods by some of the authors, it was found to be 7.4 parts per 10^{12} parts of water (Guadagni and Buttery, 1978). Similarly, the compound *tert*-amylmercaptan (2-methyl-2-butanethiol) was reported (Meilgaard, 1975) to have an odor threshold of 7 parts per 10^{14} parts of water. On repeating this threshold in our laboratories with careful panel methods (Guadagni, 1979), it was found to be 2 parts per 10^{12} parts of water. If we consider concentrations of these compounds in the air (a measure of the amount of compound actually reaching the olfactory senses), probably

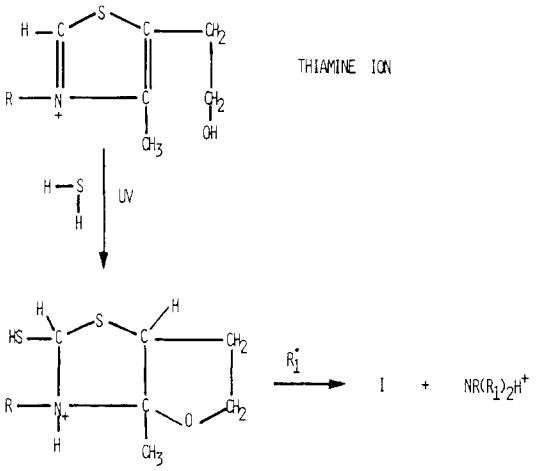


Figure 2. Possible mechanism pathway in the formation of I by UV light irradiation of thiamine hydrochloride. R is the normal (4-amino-2-methyl-5-pyrimidinyl)methyl moiety of thiamin. R_1· is any free radical.

the alkylmethoxypyrazines have the lowest threshold in parts of compound per milliliter of air. 2,3,6-Trichloroanisole and *tert*-amylmercaptan have extremely low solubilities in water and hence the air/water partition coefficient would tend to favor the air medium to a much greater extent than the completely water miscible alkylmethoxypyrazines [cf. Buttery et al. (1971)]. The thiamin odor compound (I) would tend to have a water solubility, and hence air to water partition equilibrium, somewhat in between that of the alkylmethoxypyrazines and 2,3,6-trichloranisole and *tert*-amylmercaptan.

Odor Description. Panel studies were also carried out on the odor description of I with 16 judges. A solution of 1 part of I per 10^9 parts of water was described most often as having an odor like vitamin tablets. Other odor descriptions noted less frequently were "rubbery", "boiled milk", "sulfury", and "cooked beef or chicken fat". The descriptions relating to food items hints at the possible importance of this compound in the flavor (and off flavors) of foods, perhaps resulting from the natural thiamin content. Because of its very low threshold, minute amounts which could still effect the total aroma would be very difficult to detect in foods by instrumental methods.

Possible Mechanism of Formation of I. In the earlier paper (Seifert et al., 1978) no suggestion was given on a possible mechanism of how I might be derived from thiamin. On looking at the structure of thiamine, it is not obvious at first how it can be converted to I. However, the steps shown in Figure 2 illustrate a possible pathway. It is assumed that hydrogen sulfide is first formed from thiamin breakdown. This then could possibly add to the –C=N– bond in thiamin hydrochloride as shown. Cyclization of the $-CH_2-CH_2-OH$ group under the conditions of the reaction could give the tetrahydrofuran ring. Attack of free radicals at the nitrogen might give a type of Hoffman degradation, followed by cyclization of the sulfur-containing ring. The actual detailed mechanism between each step could be considerably more complex but the general pathway outlined in Figure 2 does seem reasonable on the basis of known reactions. It is known that H_2S adds to double bonds in the presence of UV light [cf. Noller (1957)]. Ethers can be made by the addition of alcohols to olefins under certain conditions (Wagner and Zook, 1957).

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Hydrocarbon Constituents from White Strains of the Mushroom *Agaricus bisporus* (Lange) Singer

The study of the chemical composition of the cultivated mushroom *Agaricus bisporus* (Lange) Singer has been limited. Two white strains of the *A. bisporus* mushroom were analyzed for their hydrocarbon content. Hexane extractable lipids were 1.81% and 1.37% (dry weight) for The Pennsylvania State University strains 310 and 342, respectively. Hydrocarbons were isolated and identified by TLC, GC, and GC–MS. *n*-Alkanes from pentadecane to hentriacontane were identified and quantitated in each mushroom strain. Strains 310 and 342 contained 6.1 and 5.1 ppm of total *n*-alkanes/dry weight of mushrooms, respectively. Several possible functions of alkanes present in the fruiting body are discussed.

Agaricus bisporus (Lange) Singer is a cultivated mushroom that is commonly grown by the mushroom industry in the United States. Almost 400 million pounds of mushrooms are produced each year with Pennsylvania contributing the largest amount (~60%).

The literature concerning the chemical composition ($M_r \leq 450$) of the *A. bisporus* mushroom has been limited mainly to the isolation and identification of L-glutamyl derivatives (Daniels et al., 1961; Levenberg, 1961; Vogel et al., 1977), volatile components (Cronin and Ward, 1971), and free fatty acids (Holtz and Schisler, 1971).

Supplementation of compost with specific polar lipids (e.g., fatty acid esters) or various plant oils (e.g., corn or soybean oils) has been shown to produce increased mycelium growth and mushroom yields (Wardle and Schisler, 1969; Schisler, 1976). Since fatty acids and/or their derivatives have been found in *A. bisporus*, the possible presence and function of nonpolar lipids, specifically hydrocarbons, have been of interest to us. Therefore, this report examines the hydrocarbon components of the sporophore of two white strains of the cultivated mushroom *A. bisporus*.

MATERIALS AND METHODS

Growing Procedure. *A. bisporus* mushrooms (strains 310 and 342) were grown from mushroom spawn, obtained from the Pennsylvania State University spawn bank, on standard compost under temperature- and humidity-controlled conditions (Schisler and Patton, 1972). Mushrooms were harvested at first fruiting, and extraction procedures were initiated within 1 h. The growing procedures were performed in duplicate.

Sample Preparation. Hydrocarbons were isolated from each strain by homogenizing mushrooms with redistilled pesticide-grade hexane. The resulting suspension was filtered under vacuum, and the remaining solid was rinsed with spectrograde methanol and hexane. The hexane layer was separated and dried over anhydrous $MgSO_4$. After filtration, hexane was removed by rotary evaporation, resulting in a yellow oil. The yellow oil was subjected to analytical separation and identification techniques. In addition, a solvent blank was prepared by using the above extraction procedure.

Analytical Techniques. The mushroom extracts were subjected to thin-layer chromatography (TLC) as described by Kostelc et al. (1980) in order to isolate the hydrocarbon fraction. Quantitative gas chromatography (GC) of the hydrocarbon fraction was carried out on a Hewlett-Packard 7610A instrument equipped with a flame ionization detector. Glass columns (1.8 m × 2 mm i.d.) packed with 3% OV-1 on 100–120-mesh Gas-Chrom Q (Applied Science Division, State College, PA 16801) were temperature programmed from 100 to 280 °C at 6 °C/min with a 2-min delay at 100 °C.

Gas chromatography–mass spectrometry (GC–MS) was performed on a Finnigan 3200 gas chromatograph–mass spectrometer with an interactive 6000 data system. Glass columns similar to those described above were used for GC–MS analyses. A temperature program from 140 to 260 °C at 15 °C/min with a 1-min delay at 140 °C was employed. Electron impact spectra were recorded from 35 to 450 amu at 70 eV. Identification of unknowns resulted from a comparison of chromatographic retention times and mass spectra with that of authentic *n*-alkanes. Quanti-

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