

Aroma Compounds Formed from 3-Methyl-2,4-nonanedione under Photooxidative Conditions

ISABELLE A. SIGRIST, GIUSEPPE G. G. MANZARDO, AND RENATO AMADÒ*

Institute of Food Science and Nutrition, Swiss Federal Institute of Technology, Schmelzbergstrasse 9,
 ETH-Zentrum, CH-8092 Zurich, Switzerland

The behavior of the prominent aroma compound 3-methyl-2,4-nonanedione under photooxidative conditions was investigated in a model experiment. The four well-known aroma compounds 2,3-butanedione, 2,3-octanedione, acetic acid, and caproic acid were identified. The main oxidation product was 3-hydroxy-3-methyl-2,4-nonanedione, an aroma compound with the odor description of rubbery, earthy, and plasticlike (GC-O). Its structure has been tentatively assigned based on mass (GC-MS) and vapor phase infrared spectra (GC-IR). The formal formation pathways are discussed for these compounds, and other origins described in the literature are presented.

KEYWORDS: Aroma compounds; 3-methyl-2,4-nonanedione; 2,3-butanedione; 2,3-octanedione; 3-hydroxy-3-methyl-2,4-nonanedione; gas chromatography–mass spectroscopy; gas chromatography–infrared spectroscopy

INTRODUCTION

3-Methyl-2,4-nonanedione (**1**) (Figure 1) is the main contributor to the off-flavor of reversed soybean oil (1–3), to the light-induced off-flavor of butter and butter oil (4), and to the haylike off-flavor that develops in dry parsley (5) and dry spinach (6). In addition, diketone **1** is reported to contribute significantly to the flavor of green tea (7, 8) and anchovy (9, 10). Recently, this compound was also detected in dry tarragon, dry chervil, dry chive, and dry leek after light exposure (11). Compound **1** is derived from the photosensitized oxidation of the furan fatty acids 10,13-epoxy-11,12-dimethyloctadeca-10,12-dienoic acid (**2**) and 12,15-epoxy-13,14-dimethyleicosa-12,14-dienoic acid (**3**) (12) (Figure 1). The present investigation was undertaken in the context of studies on the development of **1** in food during light exposure (13) with the aim of investigating the behavior of pure **1** under photooxidative conditions.

MATERIALS AND METHODS

Chemicals. Oxygen was from Pan Gas (Lucerne, Switzerland), and 2,6-di-*tert*-butyl-*p*-toluene (BHT), ethyl decanoate, hexane, and *meso*-tetraphenyl porphyrine were from Fluka (Buchs, Switzerland). Compound **1** was synthesized according to reference 1 and purified according to reference 14. The reference substances 2,3-butanedione (**4**) and hexanal were from Fluka, acetic acid was from Merck (Dietikon, Switzerland), and 2,3-octanedione (**5**) was synthesized according to reference 15.

Oxidation of MND. The reaction was carried out according to reference 16 at 4 °C in a special three neck reaction flask ($d_1 = 4$ cm, $d_2 = 10$ cm, $h = 14$ cm) with a fritted glass bottom (DEMA 13/21, Hans Mangels, Bornheim, Germany). A solution of 0.17 mmol of **1**,

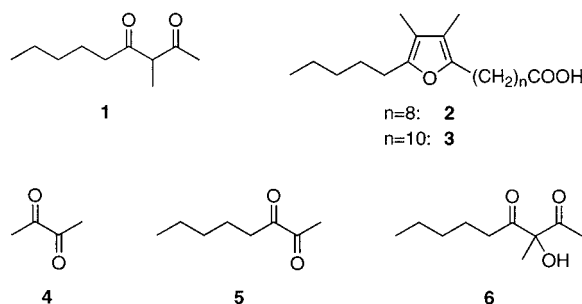


Figure 1. Structures of compounds 1–6.

20 mg of *meso*-tetraphenyl porphyrine, and 144 μ g of ethyl decanoate as an internal standard in 200 mL of hexane was exposed to light (20 000 lx) for 24 h under slightly bubbling oxygen. The light source consisted of two 150 W sodium vapor high-pressure lamps with a spectrum similar to sunlight (VIALOX NAV E 150 de Luxe, Osram AG, Winterthur, Switzerland) each placed at a distance of 0.45 m from the reaction vessel. After 24 h, 0.6 mg of BHT was added and the solution was concentrated to 3 mL at reduced pressure by means of a Vigreux column and ice cooling. Oxidation in the absence of light was carried out using the same procedure. The samples were stored at -80 °C before gas chromatography/mass spectrometry (GC-MS) analysis.

Capillary GC-MS. GC-MS was performed using the on-column injection technique on a GC 8065 gas chromatograph (Fisons Instrument, Milano, Italy) directly coupled to a SSQ 710 mass spectrometer (Finnigan, San Jose, CA). A polar-fused silica SW-10 column (60 m, 320 μ m i.d., 0.25 μ m film thickness, Supelco, Bellefonte, U.S.A.) with a deactivated fused silica precolumn (2.8 m, 530 μ m i.d., J & W Scientific, Folsom, U.S.A.) was used with helium as the carrier gas (100 kPa). The temperature program was as follows: 90 °C for 5 min, 90–240 °C at 10 °C/min, 240 °C for 10 min. EI mass spectra were recorded with an ionization energy of 70 eV in a mass range of 40–420 amu. Data processing was achieved with ICIS 2, version 8 software

* To whom correspondence should be addressed. Tel: ++41 1 632 32 91. Fax: ++41 1 632 11 23. E-mail: renato.amado@ilw.agrl.ethz.ch.

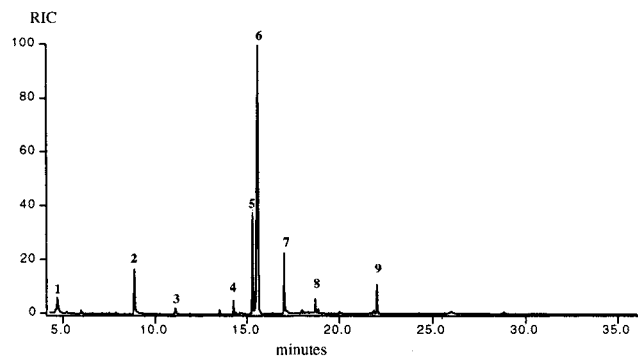


Figure 2. GC-MS chromatogram of products formed from **1** under photooxidative conditions in hexane. For peak identification, see Table 1.

Table 1. Main Products Derived from **1**^a under Photooxidative Conditions in Hexane

peak no. ^b	retention index	compd	identification ^c	photooxidative conditions	control experiment ^d
1	996	4	RS	2	nd ^e
2	1301	5	RS	5	tr/
3	1449	acetic acid	RS	1	nd
4	1640	ethyl decanoate	RS	1	1
5	1727	6	MS, IR	11	tr
6	1751	1	RS	46	103
7	1861	caproic acid	RS	5	nd
9	2270	unknown	(99, 71, 43)	2	nd

^a Data expressed as peak area ratio to the internal standard ethyl decanoate.

^b Peak numbers corresponding to those in Figure 2. ^c RS, retention index and mass spectrum consistent with reference substance; numbers in parentheses, principal ions observed in decreasing order of intensity. ^d Without light. ^e Not detected. / Traces.

(Finnigan). Identification of the products is based either on comparison of retention indices (according to van den Dool and Kratz (17)) and of the mass spectra by spiking with reference substances or on mass spectral and infrared characteristics. The products were determined in relation to the peak area of the internal standard ethyl decanoate.

Capillary GC/Infrared Spectroscopy (GC-IR). GC-IR was performed using the on-column injection technique on a HP 5890 gas chromatograph (Hewlett-Packard Co.) with a HP 5965 IR detector (Hewlett-Packard Co.). A DB 1701 column (10 m, 180 μ m i.d., 0.4 μ m film thickness, J & W Scientific) with helium as the carrier gas (100 kPa) was used. The temperature program was as follows: 40 °C for 1 min, 40–300 °C at 10 °C/min, 300 °C for 10 min. Data processing was achieved with GRAMS/32 software (Portmann Instruments, Biel-Benken, Switzerland).

Capillary GC/Olfactometry (GC-O). For sniffing experiments, a GC flame ionization detection system of the type HP GC 5890 series II (Hewlett-Packard Co.) was equipped with a column end split, leading to a sniffing port for olfactometry. A polar-fused silica SW-10 column (60 m, 320 μ m i.d., 0.25 μ m film thickness, Supelco) with helium as the carrier gas (100 kPa) was used. The temperature program was as follows: 90 °C for 7 min, 90–150 °C at 20 °C/min, 150 °C for 10 min, 150–240 °C at 20 °C/min, 240 °C for 10 min. Samples were injected in the split injection mode (split 1:12). Data processing was achieved with a HP 3365 Series II integrator (Hewlett-Packard Co.) and Chem Station Version A.03.34 software (Hewlett-Packard Co.).

RESULTS AND DISCUSSION

The chromatogram presented in Figure 2 illustrates the volatile profile of products formed from **1** (Figure 1) under photooxidative conditions (24 h in hexane). The main products are reported in Table 1. Only products with a peak area ratio to the internal standard ethyl decanoate of 1 or more were

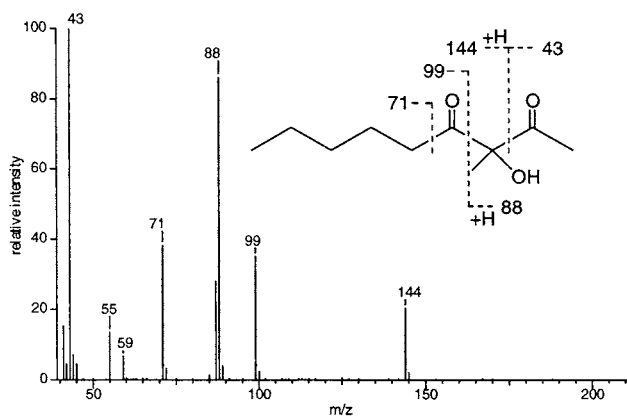


Figure 3. GC-MS spectrum of **6**.

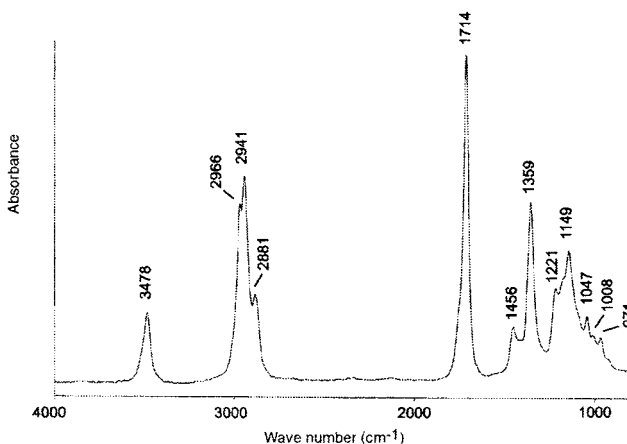


Figure 4. GC-IR spectrum of **6**.

considered. Among these products, the four well-known aroma compounds **4**, **5** (Figure 1), acetic acid, and caproic acid were identified. The main oxidation product of **1** was 3-hydroxy-3-methyl-2,4-nonanedione (**6**) (Figure 1), an aroma compound with the odor description of rubbery, earthy, and plasticlike (GC-O). Its structure has been tentatively assigned based on mass (GC-MS) and vapor phase infrared spectra (GC-IR). In the mass spectrum of **6** (Figure 3), the molecular ion was not observed. The fragment ions at m/z 43, 71, and 99 are formed by α -cleavage whereas the fragment ions at m/z 88 and 144 are attributed to McLafferty rearrangements. In the infrared spectrum of **6** (Figure 4), the OH stretching band of the tertiary hydroxy group of **6** is exhibited at 3478 cm^{-1} . According to van der Maas (18), the OH stretching band of a free (tertiary) OH group can be expected at about 3615 cm^{-1} for **6**. Evidently, the OH group is hydrogen-bonded to a C=O group; consequently, its stretching frequency is lowered and the peak is broadened. The formation of a hydroxylated dione as an oxidation product of **1** is also supported from the literature. House and Gannon (19) have shown that the oxidation of enolizable β -diketones with peracids leads to α -hydroxy- β -diketones. Furthermore, the formation of α -hydroxy- β -diketones has been reported by Yoshioka et al. (20) in the reaction of enolizable β -diketones with singlet oxygen.

In a control experiment, it was shown that the diketones **4–6**, as well as acetic and caproic acid, are only formed under photooxidative conditions. Without light exposure (Table 1, control experiment), these compounds were either absent or present in minute amounts only.

The occurrence of a compound with the same mass spectral characteristics as **6** has already been reported for green tea and

animal fat. Horita and Hara (21) described an unknown compound in an investigation on aroma concentrates of green tea after exposure to light. Braggins (22) reported on an unknown odor compound of rendered sheep fat, which elutes in the same region as **6** under comparable chromatographic conditions. The odor of the compound was described by this author as plasticlike.

Following Yoshioka et al. (20), the formation of **6** can be explained through an ene reaction of the enol form with singlet oxygen to give the α -hydroperoxy- β -diketone. This compound is subsequently cleaved to the corresponding α -hydroxy- β -diketone. The formation of **4**, **5**, acetic acid, and caproic acid can formally be explained by the oxidation of the enolic double bonds of the two most favorable enol forms of **1**: oxidation of the 2,3-enol leading to acetic acid and **5** and oxidation of the 3,4-enol to caproic acid and **4**.

Diketone **5** was reported to be an autoxidative degradation product of methyl arachidonate (23, 24) and an auto- and photooxidative degradation product of furan fatty acid **2** (16, 25), respectively. Artz et al. (24) identified diketone **4** as an oxidation product of methyl arachidonate, whereas Hollnagel and Kroh (26) have shown this compound to be a reaction product from nonenzymatic browning of D-glucose, D-fructose, maltose, and maltulose, respectively, in model experiments. Finally, **4** was identified as a photooxidative degradation product of **2** (16, 25). Compounds **1**, **4**, **5**, and **6** have been shown in green tea to increase during light exposure (13).

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