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Note

Structural Elucidation and Synthesis of 3-Hydroxybenzene-1,2-dicarbaldehyde from Astigmatid Mites[†]

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The structure of a novel aromatic compound contained in the unidentified *Rhizoglyhus* mite (Acaridae: Astigmata) was elucidated, without its isolation, to be 3-hydroxybenzene-1,2-dicarbaldehyde (tentatively named γ-acaridial) by a combination of GC/MS and GC/FT-IR together with knowledge of related mite compounds. The structure was confirmed by a 5-step synthesis, starting from methyl 3-hydroxybenzoate, in a 15% overall yield. The compound is widely distributed not only among Astigmata but also among Oribatida, although its biological function is obscure at present.

Key words: aromatic dialdehyde; Astigmata; astigmatic mite; 3-hydroxybenzene-1,2-dicarbaldehyde

Astigmata are slow-moving and weakly sclerotized mites. However, such characteristics as their prolific reproductive rate, their rapid development, and, potentially, their use of chemicals from opisthonotal glands have enabled the Astigmata to lead a cosmopolitan distribution and a wide diversity of life styles.[1] Those gland components consist of various kind of monoterpenes, hydrocarbons and aromatics.[2] Although there are several species-specific compounds, most are shared among astigmatid mites in different combinations. The present compound (whose M^+ appeared at m/z 150, and is therefore temporarily named M150) has been characterized as one of such widely distributed compounds among 28 species of Astigmata within the 52 species examined.[3] In this paper, we report the structural elucidation of M150 as a novel compound and its identification by synthesis.

The GC/MS analysis of the hexane extract from an unidentified *Rhizoglyphus* sp. (tentatively named sapporo-nedani in Japanese) showed a peak for M150 at 10.231 min (the 4th largest peak) along with 2(*E*)-(4-methyl-3-pentenyl)-butenedial (α-acaridial)[4] at 10.093 min (the largest), tridecane at 10.301 min (the second largest) and 2(*E*)-(4-methyl-3-pentenylidene)-butanedial (β-acaridial)[5] at 11.044 min (the third largest), in an HP-5 column (0.25 mm i.d. × 25 m) at a temperature programmed from 60°C to 290°C at 10°C/min with an initial 2-min hold. 3-Methyl-2-(3-methyl-2-butenyl)-furan (rosefuran),[6] neral, 4-isopropyl-3-oxo-1-cyclohexene-1-carboxyaldehyde (isorobinal),[7] and hexyl 2-formyl-3-hydroxybenzoate (hexyl rhizoglyphinate)[8] were also detected as minor components.

All attempts to isolate M150 from extracts of mite bodies (ca. 6.4 g) with an SiO_2 (Wako Gel C-200) column were unsuccessful, possibly because of chelate formation with some metallic cations present in SiO_2 as impurities. Therefore, the structure of M150 was established without its isolation, based only on information from the GC/MS and GC/FT-IR spectra. However, no difficulty was encountered with the SiO_2 purification of synthetic 3-hydroxybenzene-1,2-dicarbaldehyde (M150), as described. The limited amount (less than 1 μg) such as that present in the mite extract might have been lost during chromatography. Since the present study was conducted, we obtained purified SiO_2 (Cosmosil 75SL-II-Prep, Nacalai Tesque) and confirmed that M150 at the ng–μg level present in mites was recoverable by this purified SiO_2 without any difficulty.

M150 showed the M^+ ion at m/z 150 (66%) and the base peak at m/z 121, together with the following diagnostic ions at m/z 122 (62%), 93 (43%) and 65 (33%). The spectrum indicates losses of m/z 28 or 29 twice, suggesting the presence of a CHO or ethyl group in the molecule. The GC/FT-IR spectrum indicated two carbonyl bands at 1667 and 1716 cm^{-1}; the former corresponds to an intramolecular hydrogen-bonded aromatic aldehyde like cases of 2-hydroxy-6-methyl-benzaldehyde (1665 cm^{-1})[9] and hexyl 2-formyl-3-hydroxybenzoate (1670 cm^{-1}),[10] and the latter together with H–C–O bands at 2740 cm^{-1} and 2835 cm^{-1} indicated the presence of a free aromatic aldehyde. The structure was elucidated to be that of a salicylaldehyde with an additional formyl substitution at 2, based on related mite aromatics such as 2-hydroxy-6-methylbenzaldehyde and hexyl 2-formyl-

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[†] Chemical Ecology of Astigmatid Mites, Part LXII

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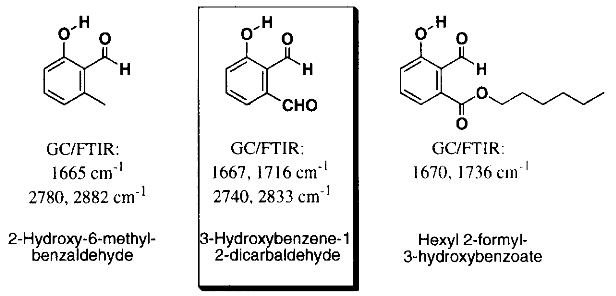

Fig. 1. Comparison of the GC/FT-IR Spectrum of 3-Hydroxybenzene-1,2-dicarbaldehyde (M150) with Those of Related Compounds.

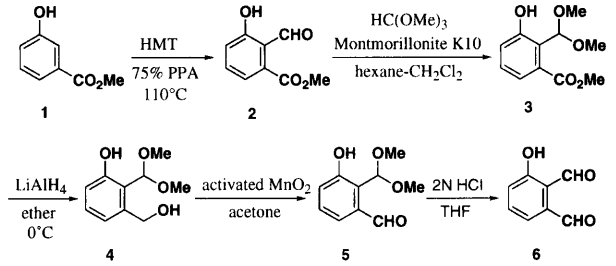

Fig. 2. Synthesis of 3-Hydroxybenzene-1,2-dicarbaldehyde (M150).

3-hydroxybenzoate (Fig. 1). As a result, the chemical structure of M150 is proposed as 3-hydroxybenzene-1,2-dicarbaldehyde. This structure was synthesized from methyl 3-hydroxybenzoate **1** in 5 steps with a total 15% yield, as summarized in Fig. 2.

The ^{1}H-NMR and ^{13}C-NMR spectra corroborate this chemical structure. GLC retention times and both GC/MS and GC/FT-IR spectra were identical to those of M150; thus the novel aromatic M150 was identified as 3-hydroxybenzene-1,2-dicarbaldehyde **6** (tentatively named γ-acaridial, based on the partial structure of butenedial).

α- and β-Acaridial present in *Rhizoglyphus* sp. are known as powerful antifungal agents.[11] The former is also known as a potent sensitizer of atopic dermatitis patients by a patch test,[12] and the latter is known as the sex[13] and aggregation pheromone[14] of *Caloglyphus polyphyllae*. Based on these biological functions of related α- and β-acaridials, compound **6** can obviously be expected to possess some biological activities such as a pheromones and (or) antifungal agent, although its actual function is obscure at present.

3-Hydroxybenzene-1,2-dicarbaldehyde is widely distributed not only among astigmatid mites[3] but also among several oribatid mites (unpublished observation). The distribution of this compound among oribatid mites will be discussed elsewhere. Astigmatid mites are assumed to have evolved from a common ancestor with oribatid mites, possessing the lateral opisthonotal glands.[15] It has recently been revealed that all of the secretory components present in the oribatid mite, *Hydronothrus crispus* Aoki, were the same as those identified among astigmatid mites.[16] Further investigation of the oribatid mite components may also provide better systematic information by which to understand the evolutional relationship between the two mite groups, Oribatida and Astigmata.

Experimental

Analytical methods. GC was performed by a Hewlett Packard 5890 Plus gas chromatograph equipped with an HP-5 column (30 m × 0.25 mm; 0.25 μm) operated in the splitless mode at 60°C for 2 min, then programmed at 10°C/min to 290°C and finally held at this temperature for 5 min. Mass spectra were measured by a Hewlett Packard HP 5989B mass spectrometer operated at 75 eV, using the same capillary column and conditions as those just described. IR spectra were recorded by a Bio-Rad FT-IRD instrument coupled with the Hewlett Packard 5890 Plus gas chromatograph. Both the ^{1}H-NMR and ^{13}C-NMR spectra were measured by a Bruker AC 300 MHz spectrometer in a CD_3OD solution. HRMS data were obtained by a Jeol JMS HX/HX 110A spectrometer.

Preparation of 2-dimethoxymethyl-3-hydroxymethylphenol (4). Methyl 3-hydroxybenzoate (**1**) was reacted at 110°C with hexamethylenetetramine (HMT) and polyphosphoric acid (PPA) to give methyl 2-formyl-3-hydroxybenzoate (**2**) in a 29% yield as previously reported.[10] After protecting the formyl group as dimethylacetal **3** by orthomethylformate and Montmorillonite K-10 as reported,[10] the methoxycarbonyl group of **3** was reduced by $LiAlH_4$ to 2-dimethoxymethyl-3-hydroxymethylphenol (**4**) in a 74% yield (two steps); mp 45–47°C; ^{1}H-NMR δ_H ($CDCl_3$): 8.72 (1H, s, Ar-OH), 7.22 (1H, dd, J=7.8, 7.8 Hz, Ar-H), 6.88 (1H, d, J=7.8 Hz, Ar-H), 6.86 (1H, dt, J=7.8, 1.5 Hz, Ar-H), 5.95 (1H, s, $-CH=(OMe)_2$), 4.63 (2H, dd, J=6.3, 1.5 Hz, Ar-$\underline{CH_2}$-OH), 3.44 (6H, s, $-OCH_3 \times 2$) and 2.51 (1H, t, J=6.3 Hz, Ar-CH_2-O$\underline{H}$); ^{13}C-NMR δ_C ($CDCl_3$): 157.1 (C(1)), 139.8 (C(3)), 130.5 (C(5)), 121.1 (C(4)), 118.3 (C(2)), 117.8 (C(6)), 103.3 [$-\underline{C}H=(OMe)_2$], 63.5 ($-\underline{C}H_2$-OH), and 53.3 ($-O\underline{C}H_3 \times 2$).

2-Dimethoxymethyl-3-hydroxybenzenecarbaldehyde (5). Phenol **4** (509 mg, 2.57 mmol) in acetone (40 ml) was oxidized at r.t. by activated MnO_2 (910 mg total in 4 portions) to 2-dimethoxymethyl-3-hydroxy-benzenecarbaldehyde (**5**) (412 mg, a rod-like colorless crystal from ether-hexane, 82%); mp 75–76°C; IR ν cm^{-1}: 3300 (-OH), 2740 (aldehyde C-H), 2830 (aldehyde C-H) and 1690 cm^{-1} (C=O); ^{1}H-NMR δ_H ($CDCl_3$): 10.09 (1H, s, -CHO), 8.95 (1H, s, Ar-OH), 7.43 (1H, dd, $J=8.0$, 7.5 Hz, Ar-H), 7.36 (1H, dd, $J=7.5$, 1.5 Hz, Ar-H), 7.15 (1H, dd, $J=8.0$, 1.5 Hz, Ar-H), 6.51 (1H, s, -CH=(OMe)$_2$) and 3.49 (6H, s, -O$\underline{C}$H$_3$×2); MS *m/z* (rel. int.) 196 (M$^+$-31, 2), 136 (10), 135 (100), 134 (30), 105 (4), 78 (6) and 210 (5).

3-Hydroxybenzene-1,2-dicarbaldehyde (6). To a THF (10 ml) solution of carbaldehyde **5** (472 mg, 2.41 mmol), 2 N HCl (1 ml) was added, and the mixture was stirred for 1 hr at r.t. The product was extracted with ether and purified in an SiO$_2$ column to give the target 3-hydroxybenzene-1,2-dicarbaldehyde (**6**); 304 mg, a light yellow needle from hexane, 84%; mp 89.5–90.5°C; GC/FT-IR ν cm^{-1}: 1667 (C=O, hydrogen bonded), 1716 (C=O), 2740 (aldehyde C-H) and 2833 cm^{-1} (aldehyde C-H); ^{1}H-NMR δ_H (CDCl$_3$): 12.35 (1H, s, C(3)-OH), 10.94 (1H, s, C(2)-CHO), 10.12 (1H, s, C(1)-CHO), 7.72 (1H, dd, $J=8.5$, 7.3 Hz, C(5)-H), 7.42 (1H, dd, $J=7.3$, 1.1 Hz, C(6)-H) and 7.27 (1H, dd, $J=8.5$, 1.1 Hz, C(4)-H); ^{13}C-NMR δ_C (CDCl$_3$): 197.3 (C(2)-$\underline{C}$HO), 192.7 (C(1)-$\underline{C}$HO), 163.6 (C(3)), 136.9 (C(1)), 136.2 (C(5)), 127.8 (C(6)), 124.5 (C(4)) and 117.3 (C(2)); MS *m/z* (rel. int.): 150 (M$^+$, 66), 122 (62), 121 (100), 104 (5), 93 (43), 76 (10) and 65 (33); HRMS *m/z* (M$^+$): calcd. for $C_8H_6O_3$, 150.0317; Found, 150.0309.

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