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4,6,8,10,16-Penta- and 4,6,8,10,16,18-Hexamethyldocosanes from the Cane Beetle *Antitrogus parvulus* - Cuticular Hydrocarbons with Unprecedented Structure and Stereochemistry

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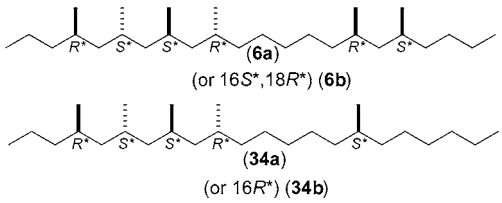
ABSTRACT



The major cuticular hydrocarbons from the cane beetle species *Antitrogus parvulus* were deduced to be 4,6,8,10,16,18-hexa- and 4,6,8,10,16-pentamethyldocosanes 2 and 3, respectively. Isomers of 2,4,6,8-tetramethylundecanal 27, 36, and 37, derived from 2,4,6-trimethylphenol, were coupled with the phosphoranes 28 and 29 to furnish alkenes and, by reduction, diastereomers of 2 and 3. Chromatographic and spectroscopic comparisons confirmed 2 as either 6a or 6b and 3 as either 34a or 34b.

Recently, we described the cuticular hydrocarbons (CHs) from several Australian melolonthine scarab beetles, the larvae of which are the chief pests of sugar-cane crops in Australia. A suite of unusual $\Delta^{9,10}$-allenic hydrocarbons was identified. In contrast, these were very low-level components in the related cane beetle, *Antitrogus parvulus*.[1] We now report that the major CHs in this species are the structurally novel 4,6,8,10,16-penta- and 4,6,8,10,16,18-hexamethyldocosanes with unprecedented relative stereochemistry within the 4,6,8,10 methyl tetrad moiety.

Cuticular hydrocarbons were extracted from intact adult female *A. parvulus* beetles with hexane. GCMS analysis of the concentrated extract showed the presence of two predominant components in a ratio of 45:38. The same components were present in similar extracts from adult male *A. parvulus*, along with a comparable amount of 9-pentacosene. The components of the female extract were separated by preparative gas chromatography, and mass spectra

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(1) McGrath, M. J.; Fletcher, M. T.; Koenig, W. A.; Moore, C. J.; Cribb, B. W.; Allsopp, P. G.; Kitching, W. *J. Org. Chem.* **2003**, *68*, 3739.

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(GCMS) (in both EI and CI mode) exhibited molecular ions at *m*/*z* 380 and 394, respectively, indicating the formulas $C_{27}H_{56}$ and $C_{28}H_{58}$. These were confirmed by accurate mass measurements (HRMS), i.e., 380.4373 (calcd for $C_{27}H_{56}$ 380.4832) and 394.4549 (calcd for $C_{28}H_{58}$ 394.4536).

The mass spectral fragmentation pattern of each component demonstrated the presence of multiple methyl branches, with a number of consecutive losses of 42 amu (or C_3H_6 units) consistent with an alternating methyl branching pattern as exhibited by **1** and **2**.

High-resolution ^{13}C NMR and DEPT spectra (187 MHz) confirmed the presence of five and six methyl branches in the C27 and C28 hydrocarbons, respectively, with the requisite number of methine and methylene signals consistent with the proposed saturated hydrocarbons. The location of methyl branches along the C22 carbon chain was based on mass spectral and NMR interpretation and shift calculations. On this basis, at least three methyls were located on alternate carbons on a C5 carbon unit (Figure 1). ^{13}C NMR shifts were

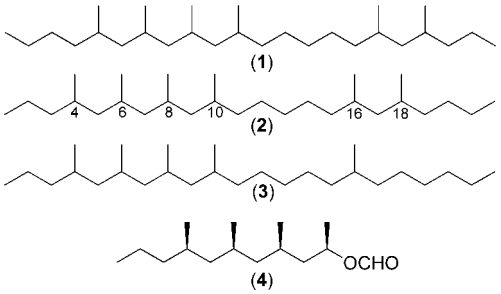

Figure 1.

then calculated for a number of alternative structures, using an equation derived from the Lindeman−Adams rule.[2]

For the C28 hydrocarbon, it was deduced that the first methyl branch at each end was four carbons from one end and five carbons from the other end. Structures **1** and **2** had calculated ^{13}C NMR shifts in best agreement with the experimental data and differ only in the number of methylenes at each end of the molecule. Two-dimensional NMR experiments (COSY, HMBC, and HSQC) provided a clear distinction in favor of **2**.

The constitution of the C27 hydrocarbon was similarly deduced to be 4,6,8,10,16-pentamethyldocosane **3**, with the calculated ^{13}C NMR resonances being in very good agreement with those observed. Mass spectral fragmentation data are consistent with the structures **2** and **3** (see Supporting Information).

The 4,6,8,10-tetramethyl pattern present in **2** and **3** was previously identified in lardolure **4**,[3] the aggregation pheromone of an acarid mite, and demonstrated to have syn, (*R*)-, stereochemistry (**4**). This unit is also present in the preen gland wax of the graylag goose.[4] To explore the possibility that hydrocarbons **2** and **3** may possess the all-syn stereochemistry as well, we fully assigned the NMR spectra of lardolure **4** (see Supporting Information). These and other data for polymethylated alkanes[5] strongly indicated that the all-syn arrangement was not present in **2** or **3**. However, the 16,18-dimethyl fragment in **2** was syn. This was established by comparisons of the methyl ^{13}C chemical shifts for syn (unresolved at δ 20.3) and anti (unresolved at δ 19.6) 7,9-dimethylhexadecane (acquired from *cis*- and *trans*-3,5-dimethylcyclohexanol; see Supporting Information) with those for the C_{16} and C_{18} methyl groups of the natural compound, **2**, at δ_C 20.3.

The foregoing analyses, when applied to the tetrad unit, suggested that the most favored relative stereochemistries for it were *anti-syn-anti*-**5** or *anti-anti-anti*-**6**, with **7**−**12** being unlikely. The various possibilities are shown in Figure 2, with all structures indicating relative stereochemistry only.

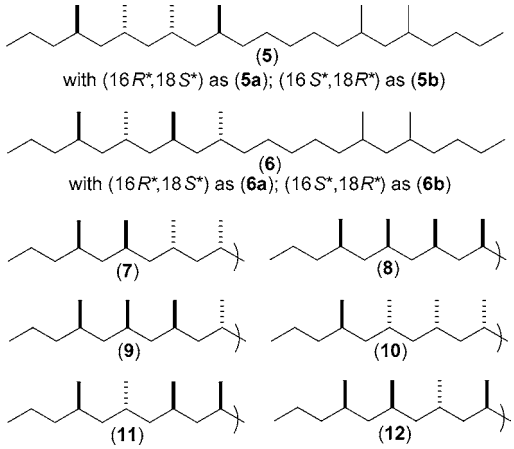

Figure 2.

Syntheses of these unusual hydrocarbons were undertaken to confirm constitutional and stereochemical features. The general approach to isomers of **2** and **3** is illustrated by the acquisition of a mixture of the anti-anti-anti system **6** and the anti-anti-syn system **11**. 2,4,6-Trimethylphenol **13** was transformed to the alcohol **26** by the sequence summarized in Scheme 1. Two one-carbon chain extensions by inverting cyanide displacement of mesylate to form **19** and **22** are the important elements. A similar undertaking, but now with *cis*-3,5-dimethylcyclohexanol, afforded syn Wittig salt and then the ylide **28**.

Aldehyde **27** was generated as needed and immediately coupled with the ylides **28** and **29** to afford the alkenes **30** and **31**, respectively, which were reduced to the hydrocarbons **32** and **33**, respectively, each as a mixture of four major

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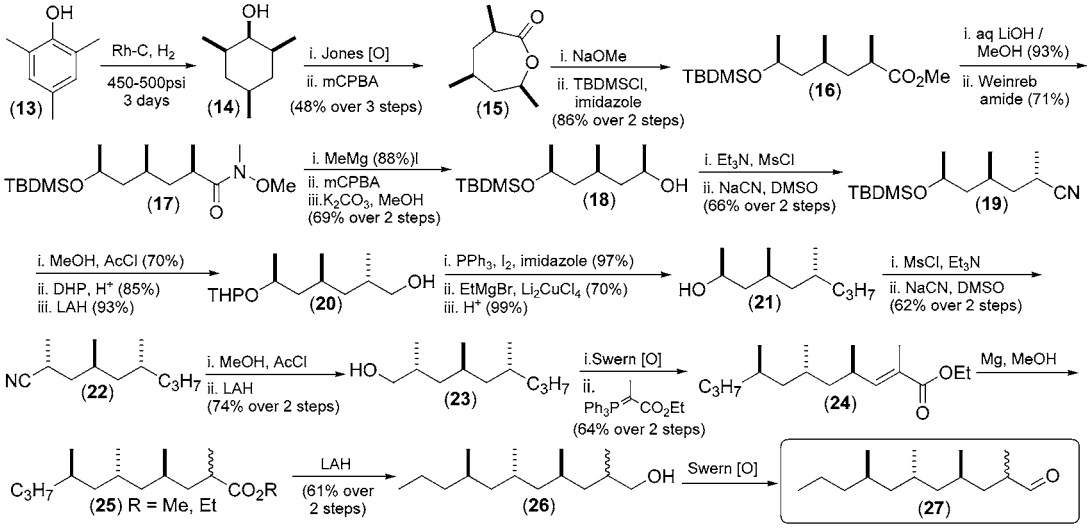
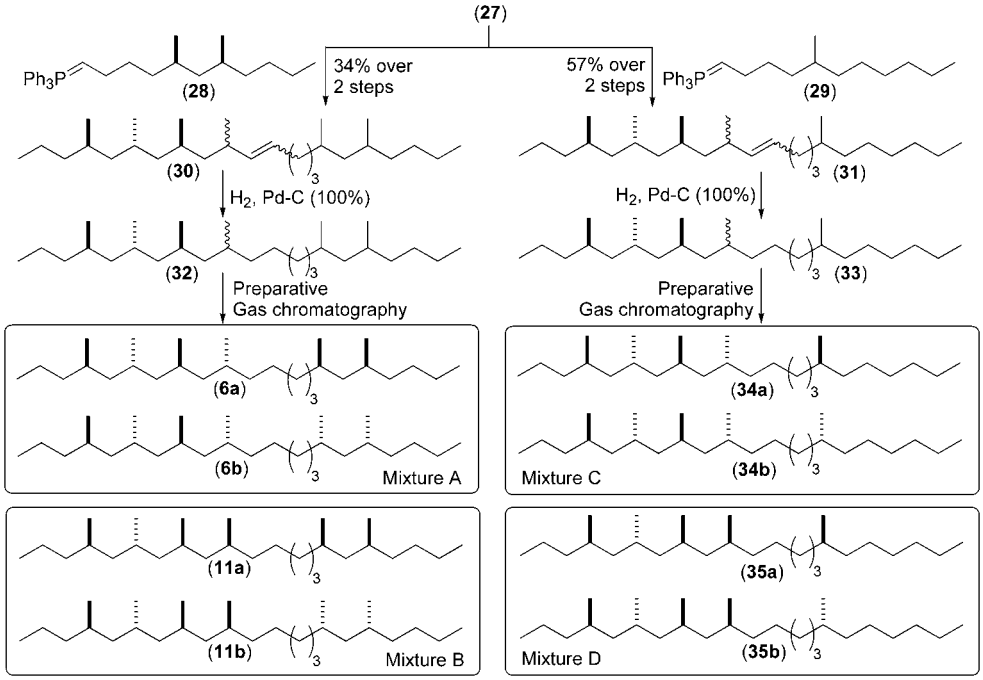
Scheme 1



diastereomers. Comparisons of NMR and mass spectra and capillary GC-MS behavior confirmed the conclusion above that the natural hydrocarbons possessed the constitutions of **2** and **3**. Very careful preparative gas chromatography of **32** and **33** permitted significant separation of the isomers, leading to mixtures A−D containing predominantly the diastereomers indicated in Scheme 2 (vide infra). Under no conditions, even capillary gas chromatography, were isomers such as **6a** and **6b** partially separated.

^{1}H and ^{13}C NMR spectra (750 MHz for ^{1}H) were then acquired for these mixtures, and consideration of the trends in ^{13}C shifts of methyl groups in 1,3-syn and anti arrangements (see Supporting Information) enabled us to assign the relative stereochemistry of the predominant diastereomers. In mixture A, **6a** and **6b** provided identical spectra except that some ^{13}C signals, for the C_{10-16} region in particular, were duplicated (with $\Delta\delta \approx 0.01$ ppm), reflecting the two possible arrangements for the $C_{16,18}$-syn dimethyl unit. A similar observation was made in the case of the pentamethyl hydrocarbons in mixtures C and D. Co-injection studies established that natural **2** coeluted with **6**, while natural **3** coeluted with **34**. There was also precise correspondence of the ^{13}C NMR shifts for natural **2** with those for **6** and of those of natural **3** with those of **34**. Not surprisingly,

Scheme 2



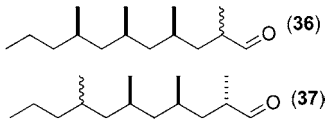


Figure 3.

however, there was no duplication of signals in the spectra of **2** and **3**, consistent with these natural compounds being single stereoisomers, but of presently unknown absolute stereochemistry. Therefore, **2** is either **6a** or **6b** and **3** is either **34a** or **34b**. Other stereoisomers of these hydrocarbons were also acquired from the aldehydes **36** and **37** (Figure 3), in turn obtained from 2,4,6-trimethylphenol with variants of the procedure summarized in Scheme 1. These provided additional comparison data that were in harmony with our stereochemical conclusions for natural **2** and **3**. Major stereoisomers acquired were always accompanied by low levels of other diastereomers (this is clear in sensitive GC-MS examinations) that result from stereoleakage during the predominant syn reduction of 2,4,6-trimethylphenol.

The novel structures and stereochemistries of the lipids **2** and **3** raise important biosynthetic questions. With **2**, four acetate and seven propionate units appear to be involved, but the absence of a terminal carboxylate group masks the direction of assembly, as made clear in Figure 4.[6]

The stereochemical alternation within the "methyl tetrad" contrasts with the situation for polyketides from other insects,[3] the graylag goose,[4] and some molluscs.[7] For this reason, **2** and **3** are of particular interest with respect to the details of the fatty acid synthase-like elongation steps in the

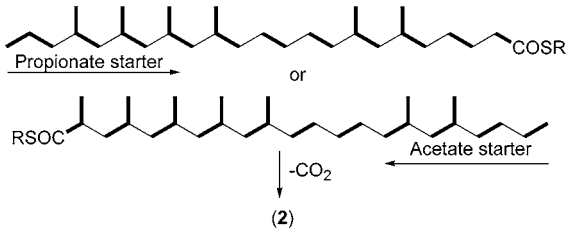


Figure 4.

polyketide chain assembly.[8] Further work will be directed toward the determination of the absolute stereochemistry of **2** and **3**.

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Supporting Information Available: Mass and NMR spectral data for natural **2** and **3**; NMR spectral assignments for **4**; NMR spectral data for **6**, **11**, **34**, and **35**; mass spectral data for **6** and **34**; synthetic scheme for *syn*- and *anti*-7,9-dimethylhexadecanes, together with NMR data for these compounds; summary of methyl ^{13}C NMR shifts for **5**, **6**, **8**, **9**, and **11**, illustrating trends for *syn*- and *anti*-1,3-dimethyl moieties; and synthetic procedures for the conversion of **19** to **21** and of **24** to **26**, with compound characterizations. This material is available free of charge via the Internet at http://pubs.acs.org.

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