

Synthesis of New Earthy Odorants

Barbara Finato,[†] Roberto Lorenzi,[‡] and Paolo Pelosi^{*†}

Istituto di Industrie Agrarie, Via S. Michele 4, and Dipartimento di Biologia delle Piante Agrarie, University of Pisa, 56100 Pisa, Italy

Several derivatives of cyclohexanol and fenchol have been synthesized, as structural models of geosmin, with the aim to provide easy to make and cheap earthy odorants. 1,2,2,6-Tetramethylcyclohexanol and 1-ethyl-2,2,6-trimethylcyclohexanol had odors very similar to that of geosmin. All of the compounds were synthesized in one step from cheap and readily available ketones, by reaction with Grignard or lithium derivatives. In some cases diastereoisomers were separated and the odors found to be markedly different. The products obtained from the two enantiomeric forms of fenchone also exhibited different odors.

INTRODUCTION

Geosmin was first discovered as a product of *Actinomyces* and isolated from this source (Gerber and Lechevalier, 1965; Gerber, 1968). It is responsible for the earthy odor in water (Rosen et al., 1970), soil (Buttery and Garibaldi, 1976), and some vegetables (Acree et al., 1976; Buttery et al., 1976).

The odor of geosmin is one of the most potent, its threshold in water being only 0.02 ppb in aqueous solution (Buttery et al., 1976). This indicates a very strong binding to olfactory receptors, making this molecule a very interesting probe for biochemical investigation of the olfactory code. Its pleasant earthy smell has recently found application in perfumery, as a secondary note in many formulations.

Unfortunately, the geosmin molecule is rather complex and its synthesis very long and expensive (Marshall and Hochstetter, 1978). Most of the problems are related to the number of possible stereoisomers, whose odors are quite different from that of natural geosmin. Natural geosmin is synthesized by *Actinomyces* only in tiny amounts, and its extraction for preparation purposes is not feasible.

Besides the above-mentioned reasons, we were interested in a radiolabeled earthy odorant, as well as in several structural analogues, to use as ligands for detecting odorant-binding proteins, similar to those so far isolated from the nasal tissue of several mammals (Pelosi et al., 1982; Bignetti et al., 1985; Pelosi and Maida, 1990).

Therefore, we synthesized several derivatives of cyclohexanol and fenchol that would bear structural similarity to the geosmin molecule. Early work of Polak et al. (1978) had shown that 2-ethylfenchol and, only to some extent, 2,6-dimethylcyclohexanol can reproduce the earthy note of geosmin. Derivatives of polymethylated cyclohexanol and cycloheptanol, as well as other compounds containing a cyclohexene ring, have been reported to exhibit the earthy note associated with other odors (Matsumoto et al., 1986).

MATERIALS AND METHODS

Synthesis of Compounds. All of the cyclohexanol and fenchol derivatives were prepared from the corresponding ketones by reaction with the appropriate Grignard or organolithium, under the conditions listed in Table I and along classical procedures. 2,2,6-Trimethylcyclohexanone was synthesized from commercial 2,6-dimethylcyclohexanone, by methylation with methyl iodide of the sodium salt, prepared with sodamide, according to the

procedure of Bailey and Madoff (1954) and purified by rectification. Diastereoisomers were separated by column chromatography on silica gel, using benzene as the eluent.

In a typical separation, 0.5 g of crude diastereoisomeric mixture was applied to a 2.5 × 30 cm column, well packed with dry silica gel (0.063 mm) and eluted with pure benzene. The most abundant isomer was collected between 55 and 80 mL of elution volume and the second between 90 and 120 mL.

The purity of all of the compounds prepared was checked by GLC on a 25-m OV-1 capillary column and found to be better than 99%. Mass spectra were recorded on a Hewlett-Packard 5992 B GC-MS, equipped with a jet separator and using an ionization voltage of 70 eV. ¹H NMR spectra were recorded on a 300-MHz Varian instrument in CDCl₃.

Odor Evaluation. Odor quality for the synthesized compounds was determined with a panel of about 20 untrained subjects, who had to choose among a list of descriptor terms to indicate the main character and secondary notes. Olfactory thresholds were measured in water solution for selected compounds, using the method of Amoore et al. (1975).

RESULTS AND DISCUSSION

Synthesis of Compounds. Table I lists the compounds prepared together with the methods employed, yields, NMR and mass spectra, and the ratio between the two diastereoisomers, as determined by capillary GLC.

Although, as far as the authors can determine, many of the compounds of Table I have not previously been described, their syntheses do not require any special comment, using well-known and general procedures.

In particular, the known compounds include 2,2,6-trimethyl-1-ethylcyclohexanol (Matsumoto et al., 1972), which was purified in both of its diastereoisomeric forms, 2,2,6-trimethyl-1-ethynylcyclohexanol (Sevin and Chodkiewicz, 1969), 1,5-dimethyl-2-isopropylcyclohexanol (Read and Watters, 1929), and 2-ethylfenchol (Polak et al., 1978).

The yields obtained in the preparations using Grignard reagents are strongly affected by the number of methyl groups next to the reacting carbonyl, whereas, as expected, reactions with alkyllithium derivatives were found to be less sensitive to steric factors. In this respect, the preparation of 1-ethyl-2,2,6-trimethylcyclohexanol as well as of 2-ethylfenchol was more conveniently and economically accomplished by synthesis of the corresponding ethynyl derivatives, followed by hydrogenation, rather than by direct alkylation with ethylmagnesium bromide. The ethynyl derivatives themselves are interesting intermediates for the synthesis of radioactive earthy-smelling compounds, through hydrogenation of the triple bond with tritium gas. The ratio between the two possible diastereoisomers was also found, as expected, to be in favor of the exo isomer for the fenchol derivatives and of the isomer

[†] Istituto di Industrie Agrarie.

[‡] Dipartimento di Biologia delle Piante Agrarie.

Table I. Synthesis and Spectra of Earthy Odorants and Related Compounds

compound	method of synthesis ^a	yield, %	isomer ratio ^b	spectra ^c
Cyclohexanol Derivatives				
1,2,2,6-tetramethyl	A	84	65/35	(a) ¹ H NMR (300 MHz, CDCl ₃) δ 0.81 (d, 3 H), 0.87 (s, 3 H), 0.88 (s, 3 H), 1.2 (s, 3 H), 1.04–1.70 (m, 7 H); MS 156 (41), 109 (22), 86 (20), 85 (100), 82 (52), 72 (33), 71 (25), 57 (37), 55 (21), 43 (66), 41 (36) (b) ¹ H NMR (300 MHz, CDCl ₃) δ 0.82 (d, 3 H), 0.85 (s, 3 H), 0.93 (s, 3 H), 0.98 (s, 3 H), 0.99–1.82 (m, 7 H); MS (practically identical with that of the first isomer)
2,2,6-trimethyl-1-ethyl	B	26	92/8	(a) ¹ H NMR (300 MHz, CDCl ₃) δ 0.80 (d, 3 H), 0.86 (t, 3 H), 0.865 (s, 3 H), 0.90 (s, 3 H), 0.94–1.80 (m, 9 H); MS 170 (17), 141 (27), 123 (47), 99 (100), 86 (75), 83 (26), 82 (41), 81 (23), 71 (42), 59 (26), 57 (57), 55 (37), 43 (73), 41 (53) (b) MS (practically identical with that of the first isomer)
2,2,6-trimethyl-1-propyl	B	12	89/11	¹ H NMR (300 MHz, CDCl ₃) δ 0.77 (d, 3 H), 0.82 (t, 3 H), 0.86 (s, 3 H), 0.90 (s, 3 H), 1.0–1.7 (m, 11 H); MS 184 (19), 141 (30), 123 (42), 114 (39), 113 (100), 100 (52), 82 (30), 71 (50), 57 (27), 55 (32), 43 (80), 41 (50)
2,2,6-trimethyl-1-butyl	C	78	53/47	MS 198 (10), 141 (30), 127 (80), 123 (43), 114 (52), 85 (64), 82 (47), 71 (55), 57 (72), 55 (63), 43 (97), 41 (100)
2,2,6-trimethyl-1-sec-butyl	C	95	63/37	MS 198 (5), 155 (18), 141 (33), 127 (27), 123 (59), 83 (38), 81 (29), 71 (40), 57 (100), 55 (54), 43 (89), 41 (84)
2,2,6-trimethyl-1-ethynyl	D	56	83/17	MS 151 (16), 133 (16), 125 (39), 110 (53), 109 (45), 95 (86), 82 (100), 67 (46), 55 (62), 54 (35), 53 (63), 43 (44), 41 (98)
2,2-dimethyl-1-sec-butyl	C	96		MS 184 (10), 127 (76), 109 (87), 82 (53), 67 (41), 59 (40), 57 (100), 55 (45), 43 (89), 41 (80)
2,6-dimethyl-1-ethyl	A	78		MS 156 (8), 127 (46), 109 (78), 99 (100), 86 (64), 83 (27), 71 (52), 67 (33), 57 (77), 55 (50), 43 (82), 41 (66)
2,6-dimethyl-1-butyl	A	54		MS 184 (5), 127 (100), 114 (22), 109 (52), 85 (30), 83 (23), 67 (24), 57 (36), 55 (31), 43 (41), 41 (47)
3,3,5,5-tetramethyl-1-ethyl	A	98		MS 155 (100), 137 (20), 113 (20), 97 (84), 81 (27), 69 (20), 57 (43), 55 (44), 43 (71), 41 (45)
3,3,5,5-tetramethyl-1-butyl	C	98		MS 179 (4), 155 (96), 99 (18), 97 (76), 81 (27), 69 (21), 57 (41), 55 (49), 43 (100), 41 (78)
1,5-dimethyl-2-isopropyl	A	77	67/33	MS 170 (3), 95 (9), 85 (100), 71 (10), 67 (11), 58 (8), 55 (12), 43 (46), 41 (25)
5-methyl-1-ethyl-2-isopropyl	A	32	97/3	MS 155 (26), 99 (100), 95 (22), 81 (44), 57 (30), 55 (29), 43 (48), 41 (38)
1-methyl-2-tert-butyl	A	68	90/10	MS 154 (3), 98 (100), 83 (31), 70 (25), 69 (30), 57 (33), 55 (40), 41 (57)
Fenchol Derivatives				
D-2-methyl	A	94	94/6	MS 152 (9), 109 (7), 81 (100), 69 (52), 67 (14), 53 (14), 43 (19), 41 (39)
L-2-methyl	A	90	95/5	
D-2-ethyl	B	7	93/7	MS 152 (11), 109 (7), 81 (100), 69 (53), 53 (12), 41 (41)
L-2-ethyl	B	8	93/7	
D-2-butyl	C	85	99/1	MS 179 (7), 155 (100), 137 (16), 99 (13), 97 (60), 83 (12), 81 (20), 57 (25), 55 (30), 43 (45), 41 (41)
L-2-butyl	C	90	99/1	
D-2-ethynyl	D	64	88/12	MS 163 (66), 150 (35), 145 (28), 135 (84), 123 (18), 122 (23), 121 (25), 107 (51), 96 (68), 95 (58), 91 (22), 81 (100), 67 (36), 53 (40), 41 (47)
L-2-ethynyl	D	58	85/15	

^a Grignard in diethyl ether; B, Grignard in benzene; C, butyllithium in hexane; D, lithium acetylide/ethylenediamine complex in hexane.

^b Cyclohexanols: cis/trans (referred to hydroxy and methyl 6). Fenchols: exo/endo. ^c (a) and (b) indicate the two diastereoisomers, (a) being the more abundant.

with the methyl in position 6 and the hydroxy group cis for the cyclohexanol derivative. Such ratio increased with the bulkiness of the organometallic alkyl group.

The identity of the two diastereoisomers was established on the basis of their NMR spectra, reported in Table I, by comparison with the data reported in the literature for the ethyl derivatives of trimethylcyclohexanol (Matsumoto et al., 1972). The mass spectra, on the other hand, do not allow one to distinguish between them, as they were found to be identical for the two diastereoisomers, within the reproducibility of the instrument.

Odor Properties. Table II reports the results of odor evaluation performed on the compounds synthesized. Several derivatives exhibited an earthy odor, as the main character, particularly the alkyl derivatives of 2,2,6-trimethylcyclohexanol and of fenchol.

Concerning the latter, it was known that 2-ethylfenchol had a strong earthy odor (Polak et al., 1978). This odor is also present in the methyl and, to a lesser extent, in the butyl derivatives; moreover, only minor differences in odor quality were detected between compounds prepared from the two enantiomeric fenchones. The ethynyl derivatives, prepared as reaction intermediates, showed markedly different odors from the corresponding saturated compounds.

In the series of the trimethylcyclohexanol derivatives, the first three terms presented a strong and rather pure earthy odor, very similar to that of geosmin, in the cis isomers, relative to the hydroxy group and the methyl in position 6. The trans isomers, instead, were described as mainly camphoraceous, with a faint earthy note. If we assume that the preferred conformation for the compounds of the cis series has the hydroxy group axial, particularly for the ethyl and propyl derivatives, the molecular models indicate a dramatic similarity with geosmin in terms of "oriented profile" (Figure 1). Such similarity can also be verified, although to a lower degree, with the alkyl fenchols, assuming, as reported (Polak et al., 1978), that the most abundant isomer has the hydroxy group in exo position.

In addition to the description reported, the odors of the compounds with a main earthy note were compared, on an informal basis, with that of geosmin. This was done by a few selected subjects who had previous experience in sensory analysis. As a result, the methyl and the ethyl derivatives of 2,2,6-trimethylcyclohexanol as well as of the two enantiomeric fenchols were rated as the most similar to geosmin, with only weak secondary notes.

Olfactory thresholds were measured in water for only three compounds. The values were 0.08 and 1.5 ppb for

Table II. Odor Properties of the Synthesized Compounds

compound	odor	
	main character	secondary notes
Cyclohexanol Derivatives ^a		
1,2,2,6-tetramethyl (a)	earthy	camphor
(b)	camphor	earthy
2,2,6-trimethyl-1-ethyl (a)	earthy	camphor
(b)	camphor	earthy
2,2,6-trimethyl-1-propyl (a)	earthy	camphor
(b)	camphor	earthy
2,2,6-trimethyl-1-butyl	camphor	earthy/minty
2,2,6-trimethyl-1-sec-butyl	camphor	minty
2,2,6-trimethyl-1-ethynyl	moldy	earthy
2,2-dimethyl-1-sec-butyl	camphor	minty
2,6-dimethyl-1-ethyl	earthy	moldy/camphor
2,6-dimethyl-1-butyl	camphor	earthy
3,3,5,5-tetramethyl-1-ethyl	minty	earthy/moldy
3,3,5,5-tetramethyl-1-butyl	woody	musk/earthy
1,5-dimethyl-2-isopropyl	minty	camphor/moldy
5-methyl-1-ethyl-2-isopropyl	minty	camphor
1-methyl-2-tert-butyl	minty	camphor
Fenchol Derivatives		
D-2-methyl	earthy	woody
L-2-methyl	earthy	musk
D-2-ethyl	earthy	camphor
L-2-ethyl	earthy	moldy
D-2-butyl	earthy	camphor
L-2-butyl	earthy	camphor
D-2-ethynyl	camphor	earthy
L-2-ethynyl	camphor	earthy

^a (a) and (b) are diastereoisomers as indicated in Table I.

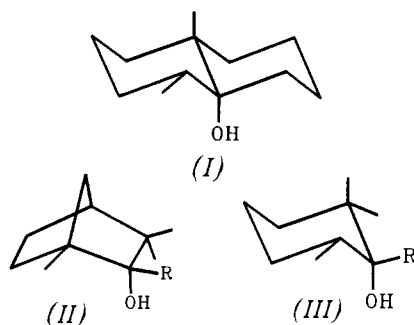


Figure 1. Comparison between the tridimensional structures of geosmin (I) and those of the earthy-smelling compounds 1-alkylfenchols (II) and 1-alkyl-2,2,6-trimethylcyclohexanols (III).

cis- and *trans*-1,2,2,6-tetramethylcyclohexanol, respectively, and 0.12 ppb for *cis*-1-ethyl-2,2,6-trimethylcyclohexanol. In the same conditions and with the method employed, the olfactory threshold of geosmin was 0.04 ppb. These values show that not only the odor character of geosmin is reproduced in the cyclohexanol derivatives but also its intensity. Moreover, the isomer with less desirable odor, which is also produced in lower yield in the synthesis, presents a higher threshold; this means that its impact on the odor of the isomer mixture, as in the crude reaction product, is not relevant, and therefore there is no need to purify the *cis* isomer from the *trans* byproduct.

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LITERATURE CITED

- Acree, T. E.; Lee, C. J.; Butts, R. M.; Barnard, J. J. Geosmin, the earthy component of table beet odor. *J. Agric. Food Chem.* **1976**, *24*, 430-431.
- Amoore, J. E.; Forrester, L. J.; Buttery, R. G. Specific anosmia to 1-pyrroline: the spermous primary odor. *J. Chem. Ecol.* **1975**, *1*, 291-310.

- Bailey, W. J.; Madoff, M. J. Separation of alkylated ketones. *J. Am. Chem. Soc.* **1954**, *76*, 2707-2708.
- Bignetti, E.; Cavaggioni, A.; Pelosi, P.; Persaud, K. C.; Sorbi, R. T.; Tirindelli, R. Purification and characterization of an odorant binding protein from cow nasal tissue. *Eur. J. Biochem.* **1985**, *149*, 227-231.
- Buttery, R. G.; Garibaldi, J. A. Geosmin and methylisoborneol in garden soil. *J. Agric. Food Chem.* **1976**, *24*, 1246-1247.
- Buttery, R. G.; Guadagni, D. G.; Ling, L. C. Geosmin, a musty off-flavor of dry beans. *J. Agric. Food Chem.* **1976**, *24*, 419-420.
- Gerber, N. N. Geosmin, from microorganisms, is *trans* 1,10-dimethyl-*trans*-9-decalol. *Tetrahedron Lett.* **1968**, *25*, 2971-2974.
- Gerber, N. N.; Lechevalier, H. A. Geosmin, an earthy-smelling substance isolated from Actinomycetes. *Appl. Microbiol.* **1965**, *13*, 935-938.
- Marshall, J. A.; Hochstetler, A. R. The synthesis of (+/-)-geosmin and the other 1,10-dimethyl-9-decalol isomers. *J. Org. Chem.* **1968**, *33*, 2593-2595.
- Matsumoto, K.; Sakata, G.; Tachibana, Y.; Fukui, K. The stereochemistry of nucleophilic addition. IV. The condensation of 2,2,6-trimethylcyclohexanone with *t*-butyl acetate in the presence of lithium amide. *Bull. Chem. Soc. Jpn.* **1972**, *45*, 1147-1152.
- Matsumoto, K.; Fujikura, Y.; Nakajima, M. Cycloalkan-1-ol derivatives and perfume compositions comprising the same. *Eur. Pat.* 0193008, 1986.
- Pelosi, P.; Maida, R. Odorant binding proteins in vertebrates and insects: similarities and possible common function. *Chem. Senses* **1990**, *15*, 205-215.
- Pelosi, P.; Baldaccini, N. E.; Pisanelli, A. M. Identification of a specific olfactory receptor for 2-isobutyl-3-methoxy-pyrazine. *Biochem. J.* **1982**, *201*, 245-248.
- Polak, E.; Trotier, D.; Baliguet, E. Odor similarities in structurally related odorants. *Chem. Senses Flavour* **1978**, *3*, 369-380.
- Read, J.; Watters, A. J. Piperitone. X. Synthesis of certain menthadienes, menthenes and menthanols. *J. Chem. Soc.* **1929**, 2165-2172.
- Rosen, A. A.; Mashni, C. T.; Safferman, R. S. Chemistry of odor in water: cause of earthy/musty odor. *Water Treat. Exam.* **1970**, *19*, 106-119.
- Sevin, A.; Chodkiewicz, W. Stereochemistry of the addition of α and β unsaturated organometallics to hindered cyclohexanones. *Bull. Soc. Chim. Fr.* **1969**, 4023-4026.

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Registry No. Fenchone, 1195-79-5; 2,2,6-trimethylcyclohexanone, 2408-37-9; 2,6-dimethylcyclohexanone, 2816-57-1; (\pm)-*cis*-1,2,2,6-tetramethylcyclohexanol, 139895-91-3; (\pm)-*trans*-1,2,2,6-tetramethylcyclohexanol, 139895-92-4; (\pm)-*cis*-2,2,6-trimethyl-1-ethylcyclohexanol, 139895-93-5; (\pm)-*trans*-2,2,6-trimethyl-1-ethylcyclohexanol, 139895-94-6; (\pm)-*cis*-2,2,6-trimethyl-1-propylcyclohexanol, 139895-95-7; (\pm)-*trans*-2,2,6-trimethyl-1-propylcyclohexanol, 139895-96-8; (\pm)-*cis*-2,2,6-trimethyl-1-butylcyclohexanol, 123735-84-2; (\pm)-*trans*-2,2,6-trimethyl-1-butylcyclohexanol, 139895-97-9; 2,2,6-trimethyl-1-sec-butylcyclohexanol, 139895-98-0; (\pm)-*cis*-2,2,6-trimethyl-1-ethynylcyclohexanol, 62861-89-6; (\pm)-*trans*-2,2,6-trimethyl-1-ethynylcyclohexanol, 62861-90-9; 2,2-dimethyl-1-sec-butylcyclohexanol, 139895-99-1; 2,6-dimethyl-1-ethylcyclohexanol, 51478-29-6; 2,6-dimethyl-1-butylcyclohexanol, 139896-00-7; 3,3,5,5-tetramethyl-1-ethylcyclohexanol, 78829-27-3; 3,3,5,5-tetramethyl-1-butylcyclohexanol, 139896-03-0; 5-methyl-1-ethyl-2-isopropylcyclohexanol, 38618-25-6; 5-methyl-1-ethyl-2-isopropylcyclohexanol, 18368-87-1; (\pm)-*cis*-1-methyl-2-tert-butylcyclohexanol, 139896-01-8; (\pm)-*trans*-1-methyl-2-tert-butylcyclohexanol, 139896-02-9; (1*S*-*exo*)-2-methylfenchol, 139974-53-1; (1*S*-*endo*)-2-methylfenchol, 20536-56-5; (1*R*-*exo*)-2-methylfenchol, 139974-54-2; (1*R*-*endo*)-2-methylfenchol, 137255-08-4; (1*S*-*exo*)-2-ethylfenchol, 137255-15-3; (1*S*-*endo*)-2-ethylfenchol, 137255-09-5; (1*R*-*exo*)-2-ethylfenchol, 137255-11-9; (1*R*-*endo*)-2-ethylfenchol, 137255-07-3; (1*S*-*exo*)-2-butylfenchol, 139974-55-3; (1*S*-*endo*)-2-butylfenchol, 139974-56-4; (1*R*-*exo*)-2-butylfenchol, 139974-57-5; (1*R*-*endo*)-2-butylfenchol, 139974-58-6; (1*S*-*exo*)-2-ethynylfenchol, 137255-14-2; (1*S*-*endo*)-2-ethynylfenchol, 21696-69-5; (1*R*-*exo*)-2-ethynylfenchol, 137255-12-0; (1*R*-*endo*)-2-ethynylfenchol, 131062-94-7.