reactive peak that had a retention time identical to that of authentic makisterone A was evident. When the immunoreactive area from the reversed-phase fractionation (fig. a; fraction 14) was injected on silica, a single immunoreactive peak was also revealed that likewise matched the retention time of authentic makisterone A, although no UV-absorbance was observed. After correcting for cross-reactivity, the concentration was calculated to be 10.6 (silica) to 12.6 (reversed-phase) ng of makisterone A per gram of ovaries on a fresh weight basis. No immunoreactivity was observed in fractions co-eluting with the C_{27} ecdysteroids, 20-hydroxyecdysone or ecdysone, which were well-separated from the makisterone A fraction under both reversed-phase and silica chromatographic conditions (fig. a and b).

Since ovarian ecdysteroids are frequently present as conjugates[15–17], we enzymatically hydrolyzed the contents of the aqueous phase resulting from the butanol/water partition[10,18]. Subsequent HPLC/RIA analysis revealed that only 2.25 ng of makisterone A per gram fresh weight of ovaries were liberated by hydrolysis.

These data represent the first report of a 28-carbon molting hormone from an adult holometabolous insect and along with previous work involving honey bee pupae[3], strongly suggest that the C_{28} ecdysteroid, makisterone A, is the major molting hormone in *A. mellifera*. We have been unable to verify the presence of C_{27} ecdysteroids as previously suggested[6]. The nature of the molting hormone in other Hymenoptera, however, remains unclear. Ecdysone and 20-hydroxyecdysone have been reported from two species of ants, *Pheidole pallidula* queens[7] and *Plagiolepsis pygmaea* larvae[8]. The separation and identification of these ecdysteroids, however, was based upon thin-layer chromatography (TLC), a procedure that would not necessarily detect makisterone A if this compound were present, due to the relatively poor resolution by TLC. The nature of the neutral sterols in these species would be of interest as it might give an indication as to whether C_{27} or C_{28} ecdysteroids are synthesized. For example, analysis of brain and whole body extracts of another ant species, the leaf-cutting ant, *Atta cephalotes isthmicola*, revealed the presence of only 28-carbon sterols[19] (C-24 alkyl), an indication that this particular species cannot convert plant sterols to cholesterol and therefore may produce a 28-carbon molting hormone.

The sterol composition and correlation to ecdysteroid content in the Hemiptera (true bugs)[20,21] may have an interesting analogy in the Hymenoptera. Previous studies have shown that blood-sucking hemipterans, which have a preponderance of cholesterol in their diets, produce the C_{27} ecdysteroids, ecdysone and 20-hydroxyecdysone, while other phytophagous hemipterans, whose diets are rich in C_{28} and C_{29} phytosterols and cannot convert these sterols to cholesterol[20], utilize makisterone A as their molting hormone[21,22]. Analysis of sterols in six species of Hymenoptera reveal a similar pattern, at least with regard to sterol composition[23]. Phytophagous species contain relatively high levels of 24-methylenecholesterol, whereas omnivorous or predatory species contain mostly cholesterol. Future investigations aimed at elucidating the ecdysteroid content of insects containing predominantly 28-carbon sterols may reveal that C_{28} molting hormones, like makisterone A, are more prevalent than originally thought.

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0014-4754/86/020200-02$1.50 + 0.20/0

Hydroxylation of menthols and cineoles with *m*-chloroperbenzoic acid

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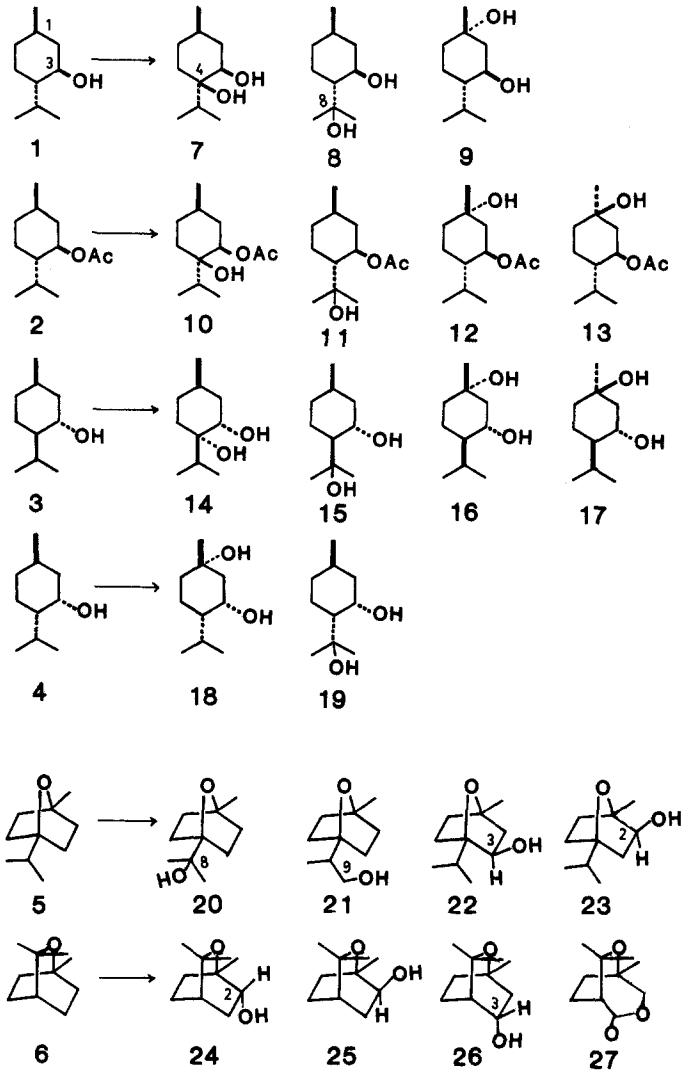
Summary. Reaction of menthols and cineoles with *m*-chloroperbenzoic acid afforded tertiary, secondary, and primary alcohols, some of which were natural products having potent plant growth regulatory activity or were mammalian metabolites.
Key words. 1-Menthol; *iso*-menthol; *neo*-menthol; 1,4-cineole; 1,8-cineole; *m*-chloroperbenzoic acid; hydroxylation.

We have been studying the hydroxylation of terpenoids in mammals[1]. Such reactions have some synthetic utility in the preparation of insect pheromones and perfume protecting agents. Although several methods for the introduction of a hy-



droxyl group are known (e.g. dry ozonization[2], microbial oxidation[3], remote oxidation[4], etc.), they are somewhat tedious and not necessarily easy to carry out. *m*-Chloroperbenzoic acid (*m*CPBA) has been used for the introduction of hydroxyl groups at bridgehead positions of polycyclic compounds[5]. In these cases, however, products are limited to tertiary alcohols. We have examined reactions of natural products with *m*CPBA and found for the first time that secondary and even primary alcohols can be produced. This note reports a simple method of preparing menthanediols and hydroxylated cineoles, some of which are natural products possessing plant growth regulatory activity[6] or are mammalian metabolites.

l-Menthol (**1**) was treated with *m*CPBA (2.2 equiv.) in chloroform under reflux for 24 h. After work up, chromatography on silica gel (**1** was recovered in 56% yield) afforded menthone (14.5%[7]), mentholactone (39.3%[7]), 4β-hydroxy-*l*-menthol (**7**, 6.6%[7]),[8,9] 8-hydroxy-*l*-menthol (**8**, 25.6%[7]),[6] and 1α-hydroxy-*l*-menthol (**9**, 4.2%[7])[10]. As a simple oxidation to a ketone and a subsequent Baeyer-Villiger reaction predominated, *l*-menthyl acetate (**2**) was next investigated.

l-Menthyl acetate (**2**) was treated with *m*CPBA in the same manner (82% recovery of **2**) to afford three hydroxy acetates, **10** (14.7%[7])[11], **11** (21.8%[7])[11], and **12** (12.4%[7])[11], corresponding to **7**, **8** and **9**, respectively, as well as another hydroxy acetate (**13**, 10%[7]) ($[\alpha]_D$ – 57.5° ($CHCl_3$))[11]. The ^{1}H-NMR spectrum of **13** suggested that it was a stereoisomer of **12**. This was confirmed by an NOE experiment. When the methine proton at δ 4.82 of **13** was irradiated, an NOE on the 1-methyl group (δ 1.26, s) was observed in the NOE difference spectrum. It is interesting to note that not only retention but also inversion of the stereochemistry can occur in these reactions.[2]

The reaction of *iso*-menthol (**3**) with *m*CPBA (**3** was recovered in 48% yield) produced *iso*-menthone (2.4%[7]), *iso*-mentholactone (52.4%[7]), 4α-hydroxy-*iso*-menthol (**14**, 6.4%[7]) ($[\alpha]_D$ + 20.2° ($CHCl_3$))[11], 8-hydroxy-*iso*-menthol (**15**, 14.8%[7]) ($[\alpha]_D$ + 5.3° ($CHCl_3$))[11], 1α-hydroxy-*iso*-menthol (**16**, 4.4%[7]) ($[\alpha]_D$ + 37.3° (CH_3OH))[11], and 1β-hydroxy-*iso*-menthol (**17**, 11.6%[7]) ($[\alpha]_D$ + 48.3° (CH_3OH))[11]. In this case again the 1β-hydroxy isomer was produced. An NOE between the 1-methyl group and the 3β-methine proton was observed in the case of **16**, while in the case of **17** no NOE was detected. Acetylation of **16** gave a mono-acetate ($[\alpha]_D$ + 57.6° ($CHCl_3$))[11], whose spectral data were completely identical with those of **13** except for the sign of optical rotation. *Neo*-menthol (**4**) furnished menthone (5.9%[7]), mentholactone (59.4%[7]), and two hydroxylated compounds: 1α-hydroxy-*neo*-menthol (**18**, 13.3%[8])[10] and 8-hydroxy-*neo*-menthol (**19**, 13.3%[7]),[6] as well as the starting material (**4**; recovered in 66% yield). Thus, compound **19**, having potent growth regulatory activity against lettuce seeds[6], was synthesized in one step from neomenthol.

1,4-Cineole (**5**), on treatment with *m*CPBA in the same way, gave 8-hydroxy-1,4-cineole (**20**, 31.3%[7]),[11,12] 9-hydroxy-1,4-cineole (**21**, 3.4%[7])[11,12], 3-*exo*-hydroxy-1,4-cineole (**22**, 2.9%[7])[11], and 2-*exo*-hydroxy-1,4-cineole (**23**, 8.5%[7])[11]. The structures were determined by decoupling and NOE experiments. Although, as expected, a tertiary alcohol predominated, it is interesting to note that a primary alcohol was also produced albeit in poor yield.

As the last example, 1,8-cineole (**6**) was allowed to react with *m*CPBA to afford the known compounds, 2-*endo*-hydroxy-1,8-cineole (**24**, 7.2%[7])[13], 2-*exo*-hydroxy-1,8-cineole (**25**, 18.1%[7])[14], and 3-*endo*-hydroxy-1,8-cineole (**26**, 10.9%[7])[13], as well as a lactone (**27**, 1.7%[7])[11]. As the C–2 protons of **27** appeared as a doublet (δ 4.03, J = 12 Hz, due to gem coupling) and a doublet of doublets (δ 4.18, J = 12 and 1.5 Hz, due to gem and long-range couplings), the structure was established as shown.

The present results show that the reaction of menthols and cineoles with *m*CPBA introduces a hydroxyl group at an unactivated carbon atom and/or gives a lactone via a Bayer-Villiger reaction of the corresponding ketone. These reactions are quite advantageous for the synthesis not only of compounds hydroxylated at a bridgehead position but also other types of alcohols. As the present method enables simple, one-step syntheses of biologically active natural products to be carried out, further investigations concerning sesqui-, di-, and triterpenes, steroids, and aromatic compounds are under way.

* We thank Nippon Terpene Co. Ltd for their generous gift of the compounds used in this work and Drs M. Kido and Y. Fukuyama, Otsuka Pharmaceutical Co. Ltd, for measurement of high resolution mass spectra. The present work was supported in part by a Grant-in-Aid for Cancer Research from the Ministry of Health and Welfare.

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0014-4754/86/020201-03$1.50 + 0.20/0

Premating behavior and male discrimination in *Jaera ischiosetosa* (Isopoda)

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Summary. Premating in females of *Jaera ischiosetosa* is statistically biased according to body size, to the size of males, to their physiological stage, and to their fertility irrespective of size. Most of these features are in agreement with the hypothesis of male discrimination between females.
Key words. Sexual selection; behavior; mate choice; body size; Isopods.

In developing the concepts of sexual selection and mate choice, Darwin[1] regarded females as the discriminating sex in most animal species. A genetic explanation for this rule, put forward by Bateman[2], states that females are in most cases the limiting factor for offspring production while males are in competition for access to mates, a view later generalized in Trivers' theory of 'parental investment'[3]. The adaptation of female discrimination between males for 'distinguishing the really fit from the pretended fit'[4] is thought to be widespread.

The picture of mate choice seems to be different in Isopod Crustaceans where premating behavior may discriminate between females. Although females make a large parental investment in incubating a few eggs in their ventral marsupium, large females are more likely to enter into precopula than smaller females[5-8]. Female size is positively correlated with fertility through the continuous growth of the adult stage, which suggest that the non-random distribution of precopulae results from a choice by males. The adaptive advantage for a male to choose a large mate is dubious, however, because copulation occurs one or two intermolts before the release of juveniles, depending on the species; due to aging, mortality during this period is correlated with size as well as fecundity. Furthermore, ecological studies carried out on the *Jaera albifrons* complex of species[9-12] have shown that the distribution of size fluctuates seasonally in natural populations, which are of a large number of small fast-reproducing females in spring and summer, and of a small number of large slow-reproducing females in autumn and winter. In studying mate choice in these species, these variations in reproductive strategies have to be taken into account. I report here seasonal variations of mate assortment in *Jaera* and a case of non-random mating which is not correlated with size.

Nine samples of *Jaera ischiosetosa*[13] were collected at intervals of 1½ to 3½ months from the large population of Le Gouinel (northwest of Plouescat, Britanny), and reared for a week under laboratory conditions[6]. The number of individuals in each sample, and the dates of collection, are shown in the table. Individuals were distributed into several buckets, with about 200 individuals in each. One bucket, the control, was kept separate while the others, the experiments, were used for recording mate choice. Mating pairs were collected every morning at 9.00 from the experiment buckets, while about a seventh of the control was sampled at random after agitating the bucket (pairs separated during this process, and therefore, no copulating pairs were recorded). The experiment and the control were therefore begun in identical conditions, and were later sampled at the same time. In both samples, the body length of individuals of the two sexes, the physiological stage of the females and the number of eggs in their marsupium (fresh eggs and rounded developing embryos corresponding to stages A and B of Stromberg)[14] were recorded. Measurements were made using a Profilprojector Carl Zeiss (magnification × 50, precision to the nearest 0.02 mm). Measured individuals were discarded; the resulting change in the composition of the experimental sample may be considered negligible in comparison to its size (table).

The reproductive cycle of females involves two intermolts: oocytes mature in the ovaries during an intermolt 'without oostegites', and embryos develop in the ventral marsupium during an intermolt 'with oostegites'. Fertilization and egg-laying occur at the molting between, and juveniles are released at the other molting. In *Jaera ischiosetosa*, the fecundation of the female may occur at any intermolt before egg-laying. Ecological data (Veuille, in preparation) indicate that there are between two and

Mean length of mating females as compared to the control (+ standard deviation and sample size), correlation in length between males and females, and fertility differential (F.D.) (number of ovigerous females more/less fertile than predicted from the fertility curve of each control) in nine successive samples of *Jaera ischiosetosa*. Body lengths are in mm. *, $p < 0.05$; **, $p < 0.01$.

Date	5.X.1979	5.XI.1979	20.II.1980	20.IV.1980	21.VI.1980	21.VIII.1980	5.XI.1980	8.I.1981	20.11.1981
N. sample	536	> 700	1885	2134	3494	1332	793	817	709
N. pairs	84	73	85	93	108	89	58	50	32
N. control	122	115	169	173	172	124	122	152	134
Length									
pairs	2.47 + 0.28	2.37 + 0.35*	2.71 + 0.52*	2.62 + 0.46*	2.40 + 0.38	2.34 + 0.32*	2.68 + 0.39	2.84 + 0.46	2.79 + 0.49
control	2.45 + 0.35	2.51 + 0.49	2.88 + 0.68	2.81 + 0.63	2.49 + 0.50	2.22 + 0.43	2.55 + 0.44	2.96 + 0.69	2.82 + 0.74
Corr. pairs	0.37**	0.50**	0.58**	0.62	0.52**	0.32**	0.34*	0.55**	0.79**
F.D.	4/6	18/10	12/7	19/4	17/11	15/8	16/3	8/3	2/5