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Separation and Determination of Isomeric Menthols

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DURING the past several years the authors have analyzed numerous samples of menthol, mixtures of menthols, and mixtures of menthol with other terpene alcohols. In some cases the usual methods for the determination of menthol have been found satisfactory, but in many instances they do not give correct results, particularly when dealing with mixtures of isomeric menthols. The purpose of this work is to study the application of various methods of analysis to the separation and determination of menthol and to indicate certain necessary modifications.

The structural formula for menthol as established by Semmler, Beckman, and others (*8*) is shown in Figure 1. It is apparent that menthol is capable of existing in eight optically active forms. Six of the eight have been isolated and identified, as shown in Table I.

TABLE I. MENTHOL STEREOISOMERS

Stereoisomer (*7*)	Melting Point	Specific Rotation	Boiling Point (760 Mm.)
	° C.	α_D^{25}	° C.
l-Menthol	43	−49.5	216
d-Menthol	43	+50.1	216
l-Neomenthol	Liquid	−19.6	212
d-Neomenthol	−17	+19.6	212
l-Isomenthol	80.5	−24.1	218
d-Isomenthol	85.0	+26.3	218

Stereoisomers containing two or more asymmetric carbon atoms may differ in their physical properties. Further, although they undergo similar reactions, the difference in rates of reaction may be marked, as shown by this study.

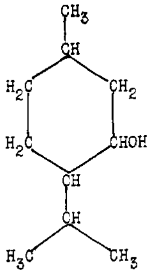

FIGURE 1

Until a few years ago the industry obtained all its "menthol U. S. P." from Japan, where it occurs naturally in oil of peppermint, *Mentha arvensis*. *l*-Menthol in the pure state is the U. S. P. material. In recent years, however, many synthetic menthols have appeared on the market. They have consisted of varied mixtures of the other stereoisomers along with some of the U. S. P. product. The number and proportion of isomers in any mixture of menthols is dependent on the method of synthesis. When *d*-citronella (*4*) obtained from citronella oil is used as the starting material, a mixture of three isomers—*d*-neomenthol, *d*-isomenthol, and *l*-menthol—is obtained. The large proportion of *l*-menthol present in this particular mixture makes possible its separation on a commercial scale. The chief interest of the authors has been in the analytical reactions of these three stereoisomers. The physical constants of the various menthols studied were determined as shown in Table II.

TABLE II. PHYSICAL CONSTANTS OF MENTHOLS

	Melting Point	Specific Rotation	Refractive Index	Boiling Point (760 Mm.)
	° C.	α_D^{25}	n_D^{20}	° C.
l-Menthol (U. S. P.)	43	−50.0	1.4600	216
d-Neomenthol	−17	+19.7	1.4600	212
d-Isomenthol	83	+25.5	1.4600	218
Mixture[a]	15	− 2.85	1.4600	214

[a] The mixture consisted of approximately 35% *l*-menthol, 40% *d*-neomenthol, and 25% *d*-isomenthol.

The *d*-neomenthol was obtained by separating it from an isomeric mixture which had been enriched in this lower-boiling material by careful fractional distillation in a 240-cm. (8-foot) laboratory fractionating unit. The final purification was effected by preparation of the *d*-neomenthol acetate (*9*) ester and subsequent hydrolysis of the recrystallized product. The *d*-isomenthol was prepared by separating it from a mixture which had been enriched in this higher-boiling material by careful fractionating as in the previous case. Final purification was achieved through repeated alcohol recrystallizations. Although it is evident that the *d*-isomenthol was slightly impure, the *l*-menthol and *d*-neomenthol were pure, as shown by comparison with Table I.

Discussion of Methods

Most methods for the determination of total menthol (*1*, *5*) involve acetylation followed by hydrolysis of the acetylated product with an excess of standard alcoholic potassium hydroxide. However, saponification by the use of potassium hydroxide in diethylene glycol (*6*) is an improvement for the reaction and can be carried out in one fourth to one fifth of the time. Preliminary studies indicated that the rates of both the acetylation and hydrolysis varied for different isomers of menthol. However, an acetylation time of 2 hours was found to be sufficient for complete reaction in all instances—in fact, 1 hour sufficed for all except the *d*-neomenthol. The low results obtained for some of the isomers were found without exception to be due to a too short hydrolysis time. Therefore,

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in the authors' study the acetylations were done in a 2-hour period, and only the time of hydrolysis was varied.

Apparatus and Reagents. The pieces of equipment necessary are 125-ml. conical Pyrex flasks, burets, pipets, and reflux condensers. The reagents are alcoholic potassium hydroxide (approximately 0.5 *N*) and 0.5 *N* aqueous hydrochloric acid. The acid is standardized against the potassium hydroxide which in turn is standardized against benzoic acid (Primary Standard 39c, National Bureau of Standards). In applying the methods of Redemann and Lucas (*6*) conical Pyrex flasks fitted with 90-cm. (3-foot) air condensers instead of glass stoppers were used, since it was found that 15 minutes or more of gentle refluxing were required for complete hydrolysis of some of the samples.

Procedures. For the acetylation, 20 grams of the menthol were introduced into an acetylation flask along with 25 ml. of acetic anhydride (c. p. grade), and 5 grams of c. p. anhydrous sodium acetate. The mixture was refluxed gently for 2 hours. One hundred milliliters of distilled water were added, the mixture was heated on a steam bath for 30 minutes to destroy excess acetic anhydride, and the aqueous layer was drawn off. The oil layer was washed twice with 10 per cent sodium bicarbonate solution and three times with distilled water until the last washings were neutral. In the past high results have been reported (*2*) because of failure completely to remove the excess acetic anhydride. The use of the bicarbonate washes very effectively eliminates this source of error. Finally the material was dried over anhydrous sodium sulfate and filtered.

For the hydrolyses, 1.0 gram of the ester was weighed and transferred to conical Pyrex flasks equipped with water-cooled condensers. Twenty-five milliliters of 0.5 *N* alcoholic potassium hydroxide were added and the solutions were brought to refluxing temperature. Determinations were made in triplicate and a blank was run with each set.

Table III. Saponification with Alcoholic Potassium Hydroxide

Sample	Ester Hydrolysis Time *Hours*	Total Menthol %	Average Menthol %	Corresponding Ester Value	Mean Ester Value
l-Menthol	0.5	99.7 99.7 99.7	99.7	282.4 282.5 282.4	282.4
l-Menthol	1.0	100.5 100.0 100.0	100.2	284.2 283.1 283.2	283.5
d-Isomenthol	0.5	98.4 98.7 98.4	98.5	279.5 279.5 280.2	279.8
d-Isomenthol	1.0	99.8 99.9 100.4	100.0	282.5 282.7 284.0	283.0
d-Neomenthol	1.0	68.9 69.2 68.4	68.8	209.0 209.7 207.8	208.8
d-Neomenthol	2.0	88.9 88.6 88.5	88.7	257.7 257.1 256.8	257.2
d-Neomenthol	3.5[a]	99.9 100.2 100.4	100.2	282.9 283.6 284.0	283.5
Mixture	2.0	93.0 92.9 93.4	93.1	267.3 267.1 267.8	267.4
Mixture	3.5[a]	100.2 100.4 100.2	100.3	283.6 284.0 283.6	283.7

[a] Not complete at end of 3 hours.

The results are shown in Table III. Values are given in terms of total menthol calculated according to the usual formula for ester-free materials (*3*) and also in terms of the corresponding ester value. The results obtained when the diethylene glycol reagent (*6*) was used are shown in Table IV.

Inspection of Table III indicates that the hydrolysis of the esters of *l*-menthol and *d*-isomenthol is practically complete within 30 minutes and is definitely finished at the end of 1 hour, but the *d*-neoester requires 3.5 hours for complete hydrolysis. At the end of 1 hour only 68 per cent "total menthol" is obtained for material that is really pure *d*-neomenthol, as is shown by the result obtained on the same material when hydrolysis time was 3.5 hours. Low results were obtained also on the mixture at the end of 1- and 2-hour periods. Although the error is less here than in the case of *d*-neomenthol, the time required for complete saponification is essentially the same.

Table IV. Saponification with Potassium Hydroxide in Diethylene Glycol

Sample	Ester Hydrolysis Time *Min.*	Total Menthol %	Average Menthol %	Corresponding Ester Value	Mean Ester Value
l-Menthol	5	98.5 99.0 99.5	99.0	279.5 280.9 282.1	280.6
l-Menthol	15	99.8 100.1 100.1	99.9	282.5 283.3 283.3	283.0
d-Isomenthol	5	99.5 99.1 99.2	99.2	282.2 281.0 281.3	281.5
d-Isomenthol	15	100.2 99.5 100.2	99.9	283.6 281.8 283.6	283.0
d-Neomenthol	15	99.5 99.3 99.4	99.4	282.1 281.6 281.9	281.9
d-Neomenthol	30	99.7 99.7 100.3	99.8	282.5 282.5 283.7	282.9
Isomeric mixture	15	98.4 98.5 98.5	98.5	279.7 279.8 279.8	279.8
Isomeric mixture	30	100.4 100.3 *100.0*	100.2	284.0 283.7 283.1	283.6

By the use of the diethylene glycol solution of potassium hydroxide, all the hydrolysis times are greatly reduced, and the precision of the results is equally as good. Again, the *d*-neomenthol and the isomeric mixture require the longer time. However, all the results indicate the definite advantage of using the diethylene glycol as a hydrolyzing medium instead of the usual alcoholic solution.

Summary

A comparison has been made of the value of alcoholic potassium hydroxide and of potassium hydroxide in diethylene glycol for the saponification of isomeric menthyl acetates. In the case of the acetate of *d*-neomenthol and of mixtures containing it, it has been found necessary greatly to increase the time usually specified for hydrolysis, since the *d*-neoester hydrolyzes so much more slowly than other menthol esters.

A method for separating *d*-neo- and *d*-isomenthol from an isomeric mixture of menthols has also been described. The method of Redemann and Lucas (*6*) for rapid hydrolysis of esters has been satisfactorily applied to the menthol esters.

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Literature Cited

(1) Assoc. Official Agr. Chem., "Official and Tentative Methods of Analysis", 4th ed., p. 571 (1935).
(2) Elliot, F. L., *J. Assoc. Official Agr. Chem.*, 13, 333 (1930).
(3) Gildemeister and Hoffman, "The Volatile Oils", 2nd ed., New York, John Wiley & Sons, 1933.
(4) Glass, H. B., and Bliss, A. R., *Drug Cosmetic Industry*, 44, 289–91 (March, 1939).
(5) Griffin, R. C., "Technical Methods of Analysis", New York, McGraw-Hill Book Co., 1927.
(6) Redemann, C. E., and Lucas, H. J., Ind. Eng. Chem., Anal. Ed., 9, 521 (1937).
(7) Schimmel & Co., "Annual Report on Essential Oils and Synthetic Perfumes", 1933.
(8) Simonsen, "The Terpenes", Cambridge University Press, 1931.
(9) Zeitschel, O., and Schmidt, H., *Ber.*, 59, 2301 (1926).