CHROMATOSTRIP ISOMERIZATION OF TERPENES*

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INTRODUCTION AND LITERATURE REVIEW

The possibility of the isomerization of terpene hydrocarbons on silicic acid has interested chemists for several years, and the literature is sometimes contradictory on this subject. ARBUZOV AND ISAEVA[1] reported that α-pinene, 3-carene and dipentene undergo isomerization when passed through a silica gel column at room temperature. RUTOVSKII AND LYUBOMILOV[2] found that neither silica gel nor aluminum sulfate alone had any appreciable effect on the isomerization of pinene to camphene. The determining factor for catalytic activity was reported to be the deposition of $Al_2(SO_4)_3$ on the SiO_2 carrier surface. RUDAKOV AND SHESTAEVA[3] used the isomerization of α-pinene as a system in studying the characteristics of a variety of silica gels. The industrial silica gels which the Russian workers examined catalyzed the isomerization of α-pinene even at 20°. These gels contained 1–10 % Al_2O_3. Silica gel freed from Al_2O_3 by the hydrolysis of $SiCl_4$ did not catalyze the isomerization at room temperature or at 100°. Their studies indicated that the activity of silica gels containing aluminum was due to the formation of alumino-silicate on the silica gel surface. HUNTER AND BROGDEN[4] made a detailed study of the isomerization and disproportionation of limonene with silica gel at 100° and 150°. Limonene initially isomerized to α-terpinene, γ-terpinene, terpinolene and isoterpinolene which subsequently disproportionated into 1-p-menthene, trans-2-p-menthene, 3-p-menthene, trans-8(9)-p-menthene and p-cymene. Polymerization occurred, as well as isomerization and disproportionation.

In studies of the volatiles of black pepper in the authors' laboratory, the monoterpene hydrocarbons were isolated by vacuum distillation and by thin-layer chromatography. The chromatostrip technique of STANLEY, IKEDA AND COOK[5] was used. It has advantages in that it is rapid, and that analyses can be run on minute quantities of material. With pepper oil, however, the material recovered by thin-layer chromatography possessed α-terpinene besides the compounds present in the vacuum distillate, and greater amounts of γ-terpinene, terpinolene and p-cymene. The compounds present in pepper oil were studied individually in an effort to determine what compounds were responsible for the observed changes. In an effort to elucidate the mechanism of sabinene isomerization with silicic acid the reaction was examined in more detail.

EXPERIMENTAL METHODS AND PROCEDURES

Preparation of chromatostrips

The chromatostrips were prepared according to the procedure of KIRCHNER, MILLER AND KELLER[6]. "Neutral" chromatostrips were prepared by using 0.1 N NaOH in place of distilled water. The chromatostrips used for detecting oxygenated compounds or polymers were prepared by the same procedure except that the starch binder was omitted. Organic compounds were detected by spraying the strips with concentrated sulfuric acid and heating on a hot plate.

Analytical reagent-grade 100-mesh silicic acid was obtained from Mallinckrodt Chemical Works, N.Y. Reagent-grade silicic acid powder was purchased from Allied Chemical and Dye Corporation, N.Y. Silica gel for thin-layer chromatography was purchased from Research Specialties Co., Richmond, Calif. The silica gel was in some instances treated by a method described by MANGOLD[7] for removal of ferric ion.

Procedure for the chromatostrip isomerization experiment

The procedure described by STANLEY, IKEDA AND COOK[5] for isolating the monoterpene hydrocarbons by downward elution of chromatostrips was used. Samples of α-pinene, α-thujene, β-pinene, sabinene, 3-carene, myrcene, α-phellandrene, α-terpinene, limonene, β-phellandrene, γ-terpinene, p-cymene and terpinolene were analyzed. 1–10 μl of the terpene hydrocarbon was diluted with 50 μl of heptane solvent, and 25 μl of this solution was applied to the chromatostrip. The remaining solution was used as a control, and exposed to the same conditions of temperature and light as the sample on the thin-layer strip. 1 μl of the eluant was examined by gas chromatography and compared to the control. Tailing of the solvent sometimes made it difficult to calculate the percentage composition. In such instances, the majority of the solvent was removed from the 2-ml eluant by evaporation, aided by passing nitrogen gas over the surface of the sample.

Sources of terpene hydrocarbons

α-Pinene, β-pinene, d-limonene, α-phellandrene and p-cymene were purchased from commercial companies and purified by gas chromatography. The sources for the other terpenes studied are reported in another publication[8].

Gas chromatography

The starting materials, reaction products, and control samples were analyzed by gas chromatography, using an Aerograph Hy-fi with a flame ionization detector. It was fitted with a 10-ft. × 1/8 in. stainless-steel column packed with 40–60-mesh HMDS-treated Chromosorb P coated with 15 % Carbowax 20M. Operating conditions were 95° and N_2 and H_2 flow rates of 20 ml/min. A Beckman Thermotrac was also used, in conjunction with a Carle thermistor detector. It utilized a 23-ft. × 1/8-in. stainless-steel column packed with 20 % Apiezon L on Gas-Pack F.

Percent composition was estimated from the chromatograms by calculating the percentage of the total peak area.

Procedure for the sabinene–silica gel isomerization experiment

A slurry of sabinene and silica gel in a 2-ml conical test tube was stoppered and

held in a 25° water bath. The starting material was 98 % sabinene, with impurities of
1 % myrcene and less than 1 % quantities of α- and γ-terpinene. The silica gel used was
Fisher Cat. No. S-157, 28–200 mesh, suitable for chromatography. Samples were
withdrawn with a syringe and injected on the gas chromatograph every 2 h for the
first 10 h. Analyses were also made at 14, 22, 26, 48 and 74 h.

At the end of the experiment, the presence of oxidized and/or polymerized
compounds was checked by ascending thin-layer chromatography in heptane solvent.

RESULTS AND DISCUSSION

Chromatostrip isomerization experiment

α-Thujene, α-pinene, β-pinene, myrcene, 3-carene, limonene, γ-terpinene and
p-cymene were found to chromatograph without change. The β-phellandrene sample
contained small amounts of 1,8-cineole, α-phellandrene, γ-terpinene and p-cymene.
1,8-Cineole would not be expected to elute from the chromatostrip, because it is
oxygenated. The chromatostrip eluant contained no 1,8-cineole, and 1 % increases of
α-phellandrene, γ-terpinene and p-cymene. These increased concentrations probably
resulted from abstraction of the 1,8-cineole, rather than isomerization of β-phellan-
drene.

The terpinolene sample contained 7 % p-cymene and 1 % limonene. If isomeriza-
tion of terpinolene to either of those compounds occurred, it did so in quantities of
less than 1 %.

The products arising from the chromatostrip isomerization of sabinene are

TABLE I

CHROMATOSTRIP ISOMERIZATION OF SABINENE

Experiment		Starting material	Product	Per-centage
A	Normal chromatostrip	Sabinene (99 %) Myrcene (1 %) α-Thujene 1 %)	Sabinene Myrcene α-Thujene α-Terpinene Limonene γ-Terpinene Terpinolene	47 1 7 13 2 26 6
B	Normal chromatostrip	Sabinene (98 %) Myrcene (1 %) α-Thujene (1 %)	Sabinene Myrcene α-Thujene α-Terpinene Limonene β-Phellandrene γ-Terpinene Terpinolene	40 2 3 14 1 2 31 7
C	"Neutral" chromatostrip	Sabinene (98 %) Myrcene (1 %) α-Thujene (1 %)	Sabinene Myrcene α-Thujene α-Terpinene γ-Terpinene Terpinolene	93 2 1 1 2 1

shown in Table I. Experiments A and B were run on different days. The differences in product composition could possibly be attributed to differences in the activity of the chromatostrips, since the strips were not of the same lot, or to differences in room temperature. The most striking difference is the presence of β-phellandrene in experiment B. Experiments B and C were run at the same time on identical samples. The chromatostrips were prepared the same day from identical materials except that 0.1 N NaOH was employed in place of distilled water in making the "neutral" chromatostrip. The greatly reduced isomerization of sabinene on the "neutral" chromatostrip supported the idea that the mechanism observed in experiments A and B is an acid-catalyzed isomerization of sabinene. The isomerization in experiment C could be ascribed to either acid or base catalysis. The unreactive impurity myrcene served as an internal standard and indicated the possibility that polymerization and/or oxidation reactions occurred, as well as isomerization. There was no indication of polymerization or oxidation when chromatostrips were sprayed with sulfuric acid and charred on a hot plate.

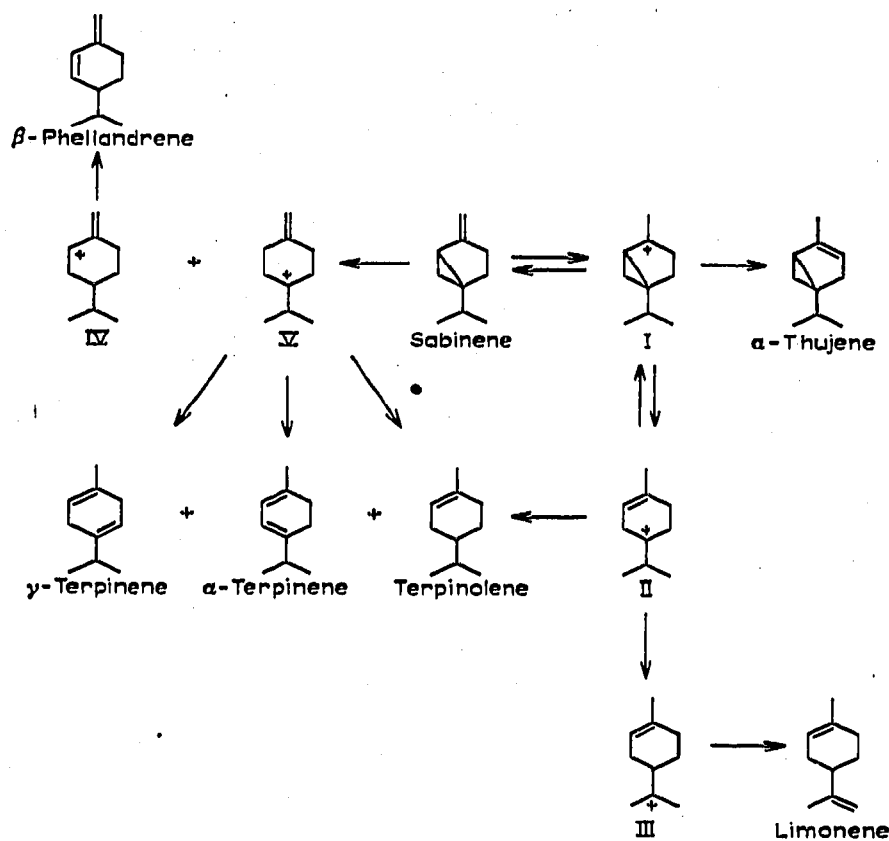Fig. 1 is a scheme to account for the products obtained in the chromatostrip

Fig. 1. Mechanism for chromatostrip isomerization of sabinene.

isomerization of sabinene. Attack of a hydrogen ion on the exocyclic double bond could result in the formation of a cyclopropylcarbonyl system (I). Loss of a hydrogen ion would lead to α-thujene, while breakage of the 3-membered ring could result in another tertiary carbonium ion (II). Dehydrogenation of II would result in the formation of terpinolene, γ-terpinene and α-terpinene. Hydrogen transfer from the

number 8 to the number 4 carbon of carbonium ion II results in the formation of carbonium ion III, from which limonene and terpinolene can be formed. β-Phellandrene was formed either in small amounts or not at all. Its presence could possibly result from breakage of the 3-membered ring of sabinene to form two carbonium ions (IV and V). Loss of hydrogen from IV would lead to β-phellandrene, while dehydrogenation of V accompanied by isomerization of the 1(7) double bond would lead to α-terpinene, γ-terpinene and terpinolene. β-Phellandrene probably does not isomerize further under these conditions, because of its more stable conjugated configuration.

Both the sample of α-phellandrene eluted from the chromatostrip and the control contained p-cymene. This oxidation product, however, was not detected in the starting material. Its formation amounted to 1 % of the chromatostrip eluant and less than 1 % of the control. α-Terpinene, which is also a conjugated endocyclic diene, reacted similarly. The sample of α-terpinene was only 60 % pure, 40 % of the mixture being limonene. p-Cymene was present in the eluant from the chromatostrip in quantities of 1 %, and was also detected in the control in quantities of less than 1 %.

The isomerization of black pepper oil observed in isolation of the monoterpene hydrocarbons by thin-layer chromatography can be accounted for by the presence of sabinene and α-phellandrene in pepper oil. α-Terpinene and increased amounts of α-terpinene and terpinolene would result from the isomerization of sabinene, and increased amounts of p-cymene from the isomerization of α-phellandrene.

Sabinene–silica gel isomerization experiment

The reaction scheme given for the isomerization of sabinene on the chromatostrip (Fig. 1) can be used to account for the products of silica gel isomerization. In addition, α-phellandrene could arise from isomerization of β-phellandrene, and p-cymene could be formed from the oxidation of α-terpinene and α-phellandrene.

Fig. 2 plots the percentage of monoterpene hydrocarbons present in the reaction mixture during the experiment. The decrease of sabinene is not shown.
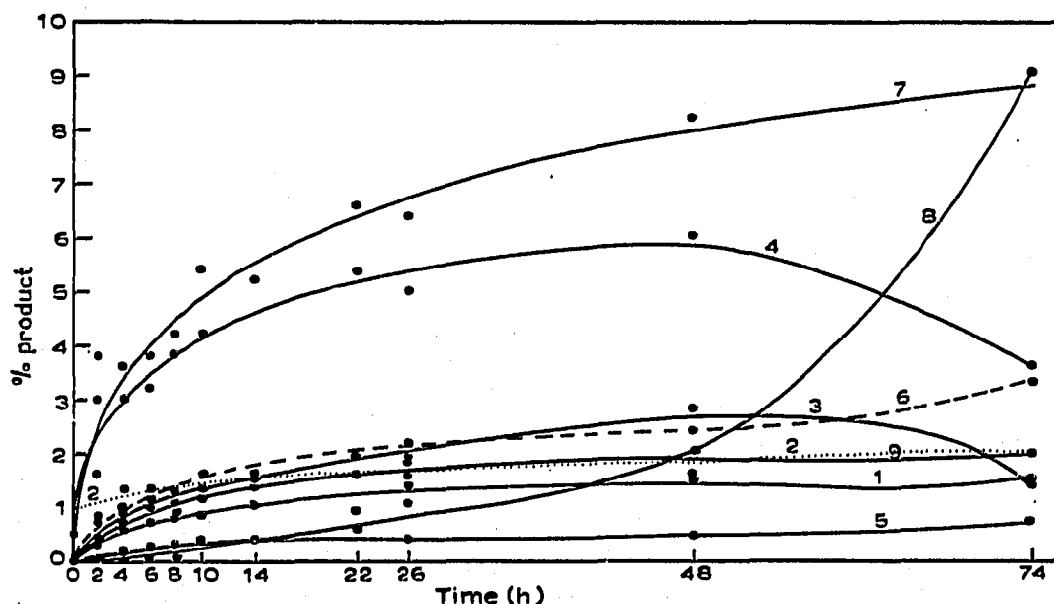

Fig. 2. Silica gel isomerization of sabinene. 1 = α-thujene; 2 = myrcene; 3 = α-phellandrene; 4 = α-terpinene; 5 = limonene; 6 = β-phellandrene; 7 = γ-terpinene; 8 = p-cymene; 9 = terpinolene.

The quantity of myrcene increased slightly. It was thought that myrcene might result not from isomerization, but from oxidation or polymerization of sabinene. Thin-layer chromatography of the reaction product substantiated this idea. The relative intensity of the charred spots on the chromatostrip indicated that as much as half of the reaction product could be higher-boiling material.

α-Thujene, limonene, β-phellandrene, γ-terpinene and terpinolene exhibit similarly shaped concentration curves. Initially, these products are formed quite rapidly; their rate of formation declines but their concentrations continue to increase throughout the experiment.

α-Phellandrene and α-terpinene, both conjugated endocyclic dienes, show marked decreases in concentration at the end of the experiment. p-Cymene is present in small quantities during the first part of the experiment and increases rapidly during the latter stages. This large increase of p-cymene can be accounted for by the decrease of α-phellandrene and α-terpinene. The oxidation of both these compounds to p-cymene observed in the chromatostrip experiment would support the possibility of this occurrence.

It is interesting that α-terpinene and not γ-terpinene appears to undergo dehydrogenation to p-cymene. If the oxidation mechanism shown in Fig. 3 is correct,

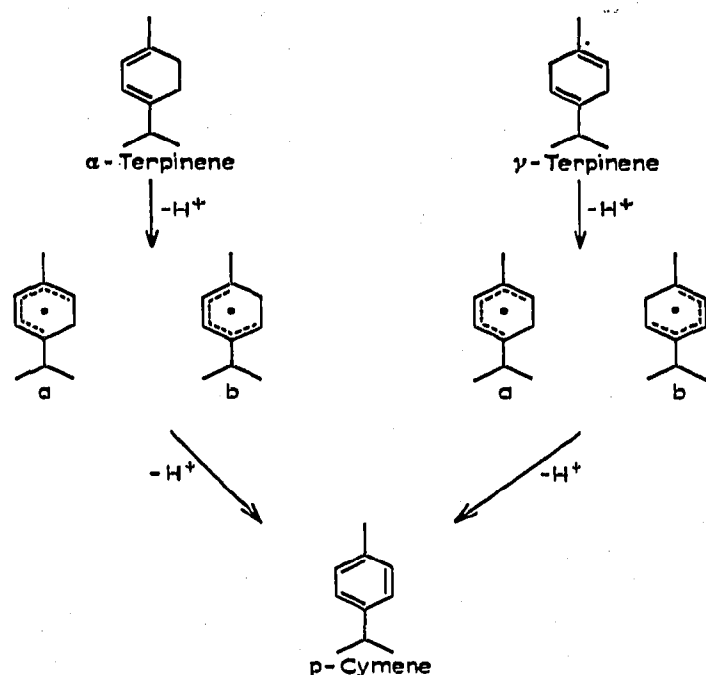Fig. 3. Oxidation of α- and γ-terpinene.

both compounds would lead to identical intermediates. The conformation of α-terpinene must be such that the transition state a, b is more easily reached. One would not expect γ-terpinene to be more stable than the conjugated isomer α-terpinene. BATES, CARNIGHAN AND STAPLES[9] studied the relative stabilities of two analogous compounds 1,3- and 1,4-cyclohexadiene. They reported that the relative stability of the two compounds was essentially the same, the conjugated isomer being only 0.07 kcal/mole more stable than the unconjugated isomer. It was proposed that an interaction occurred between the double bonds of 1,4-cyclohexadiene, resulting in resonance

stabilization. In the most stable configuration of 1,4-cyclohexadiene as suggested by HERBSTEIN[10], the π-electrons of the two double bonds are quite close on one side of the ring, allowing for such an interaction.

The formation of larger amounts of γ-terpinene than of α-terpinene does not indicate that γ-terpinene is thermodynamically more stable. The two compounds are not existing under equilibrium conditions. It must be remembered that unidentified oxygenated and/or polymerization products were formed. The possibility cannot be overlooked that α-terpinene takes part in such reactions.

ACKNOWLEDGEMENT

SUMMARY

Isomerization of the monoterpene hydrocarbons of black pepper oil occurred during isolation by thin-layer chromatography. α-Thujene, α-pinene, β-pinene, myrcene, 3-carene, limonene, β-phellandrene, γ-terpinene, terpinolene and p-cymene were each eluted from chromatostrips and were all found to chromatograph without change. Sabinene isomerized to α-thujene, α-terpinene, γ-terpinene, limonene, β-phellandrene and terpinolene when so treated. A possible mechanism for this reaction is discussed. Oxidation of α-terpinene and α-phellandrene to p-cymene occurred both in the control samples and on chromatostrips.

In the silica-gel-induced isomerization of sabinene, α-phellandrene and p-cymene were formed in addition to the chromatostrip isomerization products. The results indicated that p-cymene was formed from α-terpinene and α-phellandrene.

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