Kritchevsky for the determination of the R_f values and to Dr. Keith Freeman for the infrared spectra.

Experimental[13]

Geranyl Chloride.—This preparation was carried out as described by Ruzicka[8] using phosphorus pentachloride and either hexane or ligroin (60–70°) as a diluent. From 90 g. (0.6 mole) of geraniol, 62 g. (60%) of the chloride was obtained, b.p. 93–104° (15 mm.). The thionyl chloride method of Barnard and Bateman[14] was found to be less satisfactory.

Carbonyl-Labeled Ethyl Acetoacetate.—A mixture of 2.87 g. of magnesium, 100 ml. of absolute ethanol, 20 ml. of dry xylene and 2 ml. of carbon tetrachloride was refluxed for 12 hours and then concentrated to dryness under reduced pressure on a steam-bath. Benzene was added twice and removed *in vacuo* to ensure the absence of any alcohol.[15] The residue was heated at 100° under reduced pressure for three hours, cooled, 40 ml. of anhydrous ether added and the mixture stirred vigorously to break up the solid. Ethyl *t*-butyl malonate (22.4 g.) then was added dropwise with stirring and the mixture refluxed until complete solution was obtained. Carboxyl-labeled acetyl chloride, prepared from 10.8 g. of sodium acetate containing 9.9 mc. of C^{14} by distillation from benzoyl chloride, was dissolved in 25 ml. of dry ether and the solution was added dropwise with stirring to the magnesium derivative of the malonic ester. After refluxing for 30 minutes, the reaction mixture was cooled, diluted with water and acidified with dilute sulfuric acid. The aqueous phase was separated and extracted with ether and combined with the above ether. The solvents were distilled, the residue dissolved in 100 ml. of benzene, a small amount of the solvent distilled, 0.75 g. of *p*-toluenesulfonic acid added and the solution refluxed for 90 minutes. The cooled benzene solution was extracted with saturated sodium bicarbonate, saturated sodium chloride and the benzene removed through an 18″ column. The residue was distilled, b.p. 180°, yield 12.2 g. (71.3% based upon sodium acetate).

Geranylacetone.—The carbonyl-labeled ethyl acetoacetate (11.53 g.) was diluted with 11.0 g. of non-radioactive ester and the mixture added dropwise with stirring to a cooled solution of 3.46 g. of sodium in 100 ml. of absolute ethanol. After 15 minutes, 26.4 g. of freshly distilled geranyl chloride was added slowly and the resulting mixture heated under reflux for 25 hours. The solution was diluted with 500 ml. of water, extracted with ether and the ether distilled.

The crude product was dissolved in a solution containing 8 g. of sodium hydroxide, 350 ml. of ethanol and 230 ml. of water and refluxed for 48 hours. The reaction was then diluted with 500 ml. of water, extracted twice with 250-ml. portions of ether, the ethereal solution washed with water until neutral, then with saturated sodium chloride solution and dried over sodium sulfate. The product was distilled, b.p. 115° (1 mm.), yield 15 g. (52.5%).

Labeled "Squalene."—A mixture of 15.0 g. of geranylacetone, 10 g. of tetramethylene bromide, 2.3 g. of magnesium and a crystal of iodine was heated on a steam-bath for 30 minutes under an atmosphere of nitrogen. The mixture was then diluted with 45 ml. of dry ether and refluxed for 45 minutes by which time most of the magnesium had dissolved. Another crystal of iodine was added and the solution refluxed overnight. The reaction was decomposed with water and dilute hydrochloric acid, the ethereal layer separated and dried.

The ether was distilled, 30 ml. of dry benzene and 15 ml. of phosphorus tribromide were added and the solution heated on a steam-bath for 12 hours. The cooled mixture was poured into dilute hydrochloric acid and ice-water, the layers separated and the aqueous layer re-extracted with ether. The combined extracts were washed with dilute alkali, water and saturated sodium chloride solution. The solvents were distilled, the residue dissolved in 60 ml. of collidine and heated under reflux for 3 hours. After dilution with aqueous hydrochloric acid, the mixture was extracted four times with ether, the ethereal solution processed in the usual manner and the product distilled, b.p. 210–212° (1.5 mm.), yield 3.1 g. (19.7%).

CHEMICAL LABORATORY
UNIVERSITY OF CALIFORNIA
BERKELEY 4, CALIFORNIA

(13) All boiling points are uncorrected.

(14) D. Barnard and L. Bateman, *J. Chem. Soc.*, 926 (1950).

(15) B. Riegel and W. M. Lilienfeld, THIS JOURNAL, **67**, 1273 (1945).

The Infrared Spectra of the Isomeric 2-Decalols and their Acetates. The Effects of Stereochemical Configuration

BY WILLIAM G. DAUBEN, EARL HOERGER AND N. K. FREEMAN

RECEIVED MAY 12, 1952

To date, various methods have been employed in the decalols to assign the configuration of the hydroxyl group and the nearest ring juncture hydrogen atom. Originally, Hückel[1] allocated such configurations on the basis of the von Auwer–Skita rule and many of these assignments have been confirmed by the application of the stereospecific elimination reactions[2] in the 1-decalol series and the utilization of conformational analysis[3] in the *trans*-decalols. There is no direct method which is applicable to the *cis*-2-decalols.[4] Last year, Jones, Humphries, Herling and Dobriner[5] reported that in the sterols, a study of the 1200–1260 cm.$^{-1}$ region of the infrared spectrum could aid in the determination of the stereochemical relationship between the C_3-hydroxyl group and the C_5-hydrogen atom. They found that when the acetoxyl group at C_3 and the hydrogen at C_5 were *trans* to each other, only a single strong band occurred in this region. When such a relationship was *cis*, two or three strong bands were observed. Absorption at these frequencies is characteristic of the acetate group in general and these workers suggested that the multiple bands could be due to an equilibrium mixture of unstable rotational isomers. If such were the case, this type of analysis should be applicable to the corresponding acetates of the 2-decalols. These esters have been prepared and their spectra are shown in Fig. 1. Each spectrum was obtained several times with varying operating conditions to verify the spectral details.

The stereochemical configuration assigned to the four 2-decalols at the present time is shown below.[6] If these assigned configurations are correct and if this spectral analysis is applicable, it would be expected that the acetates of I and III should show a single strong band whereas the acetates of II and IV should exhibit a multiple band structure. An examination of the curves shows that the spectrum of the acetates of *cis*-105° decalol (I) does have a single symmetrical band in this region but it was

(1) W. Hückel, *Ann.*, **441**, 1 (1925); **451**, 109 (1926); **533**, 1 (1938); W. Hückel and C. Kuhn, *Ber.*, **70**, 2479 (1937).

(2) W. Hückel, W. Tappe and G. Legutke, *Ann.*, **543**, 191 (1940); D. H. R. Barton, *J. Chem. Soc.*, 2175 (1949).

(3) D. H. R. Barton, *Experientia*, **6**, 316 (1950).

(4) W. G. Dauben and E. Hoerger (THIS JOURNAL, **73**, 1504 (1951)) have employed an indirect method in which they assign the configuration of a *cis*-decahydro-2-naphthoic acid on the basis of the *cis*-hydrogenation concept of Linstead. The acids obtained were then related to the decalols by the use of stereospecific reactions.

(5) R. N. Jones, P. Humphries, F. Herling and K. Dobriner, *ibid.*, **73**, 3215 (1951).

(6) The positions of the hydrogen atoms are represented in the formulas by black dots, a dot indicating that a hydrogen atom is above the plane of the molecule.

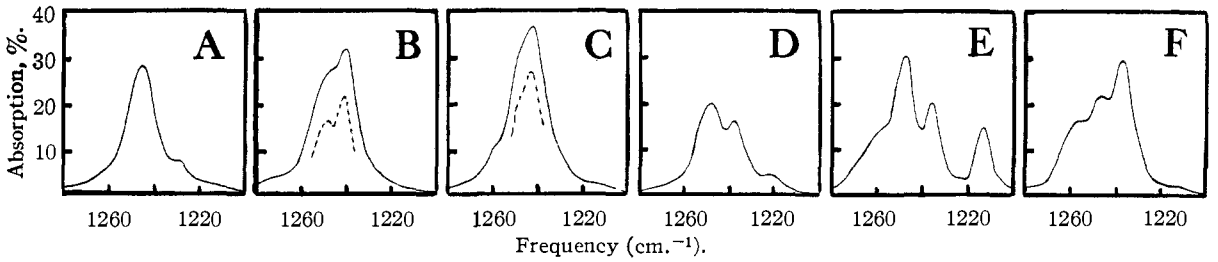
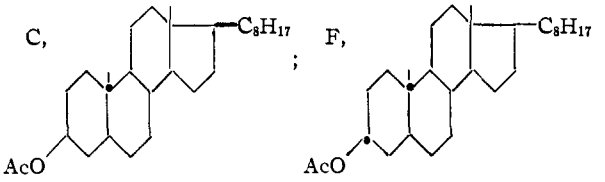
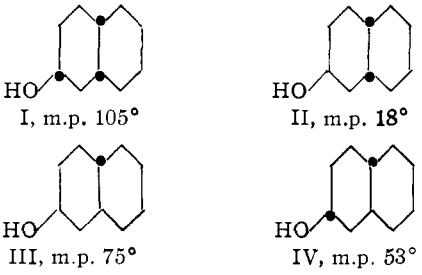


Fig. 1.—Characteristic acetate bands of stereoisomeric 2-decalyl acetates (CS_2 solution) (dotted lines indicate high resolution): acetates of A, cmpd. I; B, cmpd. III; D, cmpd. II; E, cmpd. IV;



found that, under high resolution, the ester of *trans*-75° decalol (III) is split into two bands. The acetates of *cis*-18° (II) and *trans*-53° (IV) decalols do have clear multiplicity. For comparison, the



acetates of cholestanol and epicholestanol were prepared and their spectra are also given in Fig. 1. As reported by Jones and his collaborators,[5] it was found that epicholestanyl acetate (a *cis* relationship) showed a multiplicity of bands but cholestanyl acetate (a *trans* compound) displayed a single band which was unsymmetrical. Such a band shape suggests that it might be a doublet when run under high resolution but it was found that under the running conditions where the band of the acetate of II split, the cholestanyl ester did not. Thus, in those structures in which the acetoxyl group and the ring juncture hydrogen atom have been allocated a *cis* relationship, a clear and definite multiplicity has been found whereas in the *trans* structures a splitting occurred in one isomer. In general, the relationship established by Jones, *et al.*, appears to hold quite well in this series of simpler compounds.

Another interesting correlation involving the C_3-hydroxyl group and the C_5-hydrogen atom of a sterol has recently been observed in the 1000–1100 cm.$^{-1}$ (9–10 μ) region by Rosenkrantz, Milhorat and Farber.[7] They found that compounds having the hydroxyl group *cis* to the ring juncture hydrogen atom, *i.e.*, coprostanol and epicholestanol, had bands at 1041 and 1004 cm.$^{-1}$ (9.61 and 9.96 μ) and 1038 and 1004 cm.$^{-1}$ (9.63 and 9.96 μ), respectively, while compounds with a *trans* relationship, *i.e.*, epicoprostanol and cholestanol, had bands at 1062 and 1040 cm.$^{-1}$ (9.42 and 9.62 μ) and 1075 and 1041 cm.$^{-1}$ (9.3 and 9.61 μ), respectively. Thus a shift of band pairs from 1075–1042 cm.$^{-1}$ (9.3–9.6 μ) to 1042–1000 cm.$^{-1}$ (9.6–10.0 μ) region occurs when the steric relationship changes from *trans* to *cis*. For comparison, the infrared spectra in this region for the 2-decalols are shown in Fig. 2. Assuming the assigned configurations (*vide supra*), decalols I and III should belong to the 1075–1042 cm.$^{-1}$ (9.3–9.6 μ) group and compounds II and IV to the 1042–1000 cm.$^{-1}$ (9.6–10.0 μ) group. It is seen that such a general shift of bands does occur but the difference is not as clearly defined as in the ester work described above. Nevertheless, such a generalization may be of aid, as a first approximation, in suggesting stereochemical configurations in this bicyclic series. It is not certain from this work whether such effects in the infrared discussed above bear any relationship to the C_5-hydrogen atom as suggested by the authors of the sterol papers[5,7] and used in the discussion of the decalol results or whether it is a result of an over-all steric nature of the molecule imposed by the ring juncture configurations or substitutions.

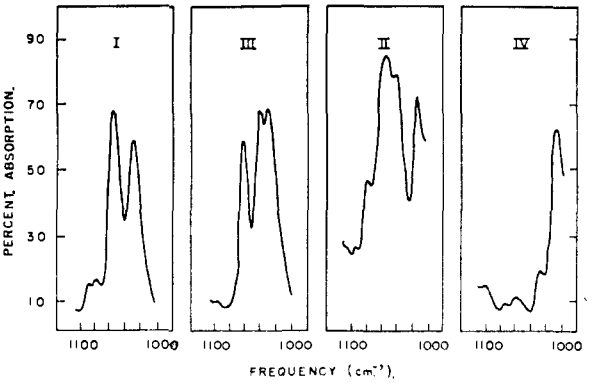


Fig. 2.—1000–1100 cm.$^{-1}$ region absorption of stereoisomeric 2-decalols (CS_2 solution).

The decalols employed in this study were prepared and purified in the most part in the standard fashion reported by Hückel.[8] The preparation of

(7) H. Rosenkrantz, A. T. Milhorat and M. Farber, *J. Biol. Chem.*, **195**, 509 (1952).

(8) W. Hückel, R. Danneel, A. Grosz and H. Naab, *Ann.*, **502**, 99 (1933), and earlier references given in this paper.

the *trans*-75°-decalol was attempted by the method of Baker and Schuetz[9] who reported that when 2-naphthol was hydrogenated at room temperature and high pressure over platinum oxide in acetic acid, the crude product obtained was essentially the pure *trans*-75°-decalol. When this work was repeated, it was found that such was not the case but the *cis*-105° isomer was the main product. Thus starting with 550 g. of 2-naphthol, the hydrogenation product was first separated into neutral and acidic components and the acidic *ar*-tetralol amounted to at least 14%. The neutral fraction was crystallized from hexane to remove the large majority of the *cis*-105°-decalol and this isomer was obtained in 36% yield. The mother liquor residues were distilled in vacuum and yielded 28% mixed decalins, 20% of mixed decalols which consisted mostly of the *cis*-18° isomer and a small amount (0.1%) of the *trans*-53° alcohol. Although no pure *trans*-75° material was isolated it cannot be stated that none is present, but if so, it is a very minor reaction product. When this hydrogenation was run on a smaller scale, the amount of hydrogenolysis was less and the crude product was a solid. It would thus appear that the conclusion of Baker and Schuetz[9] that their crude reaction product was pure *trans*-75°-decalol is erroneous and was due to the fact that their mixture was not purified.

Experimental

Hydrogenation of 2-Naphthol.—A total of 550 g. of 2-naphthol was hydrogenated at high pressure and room temperature using platinum oxide catalyst. For example, in one of the runs, 150 g. of 2-naphthol, 160 ml. of ether, 160 ml. of glacial acetic acid and 3.0 g. of platinum oxide were placed in a glass lined hydrogenation bomb and shaken at room temperature under an initial pressure of 3200 p.s.i. After 15 hours, 6.5 mole equivalents had been absorbed and the shaking was stopped. The runs were combined at this stage.

After removal of the catalyst by filtration, the catalyst was washed with ether and the washings combined with the filtrate. The ether–acetic acid solution was then made alkaline by the cautious addition of concentrated sodium hydroxide solution. Ether was added from time to time to replace that lost by evaporation during the neutralization. The ether layer was separated from the aqueous alkaline layer containing the *ar*-tetralol, washed with water and saturated sodium chloride solution. After drying, the ether was evaporated and the residue dissolved in the minimum quantity of hexane. Several crops of crystals, all melting from 100–104°, were collected and then combined and recrystallized from hexane, yield 210 g. of *cis*-105°-decalol, m.p. 104.0–104.5°.

All hexane mother liquors were evaporated and the residues combined and distilled through a 35-plate tantalum wire-packed column[10] at a reflux ratio of 10 to 1. The following fractions were collected: (1) decalin, 148 g., b.p. 81–84° (20 mm.); (2) *trans*-53°-decalol (impure), 1.5 g., b.p. 125–131° (20 mm.); (3) mixed decalols, 120 g., b.p. 131–138 (20 mm.); (4) *ar*-2-tetralol (impure), 20 g., b.p. 155–158° (20 mm.).

From fraction 2, m.p. 44–46°, 0.4 g. of pure *trans*-53°-decalol, m.p. 53.4–54.7°, was obtained by repeated recrystallization from pentane. Portions of fraction 3, liquid at room temperature, were converted to half phthalates[11] and yielded mainly the solid half phthalate of *cis*-18°-decalol, m.p. 145–148° (lit.[1] 153°). Fraction 4, m.p. 36–38°, was recrystallized from hexane and after one crystallization melts at 51–52°. Further crystallizations yielded material melting 61–62°; this result is in agreement with the known dimorphic character of *ar*-2-tetralol whose two forms melt 53–54° and 62°.[11] Apparently the tetralol was not completely extracted from ether by the alkali in the first step in the separation.

Preparation of Acetates.—The decalyl acetates were prepared by the method of Leroux.[12] The properties reported by Hückel[8] are in agreement with those found in the present research except we were unable to obtain the acetate of *cis*-105°-decalol as a solid; Hückel reports m.p. 32°. The boiling points of the esters of the decalols are as follows with the literature value given in parentheses: I, 136° (19 mm.) (122° (9 mm.)); II, 135° (15 mm.) (not reported[13]); III, 132° (20 mm.) (110° (9 mm.)) and IV, 133° (18 mm.) (118° (9 mm.)).

Infrared Spectra.—The spectra of the decalyl acetates and steroidal acetates were determined with a Model 21 Perkin–Elmer infrared spectrophotometer equipped with a NaCl prism. The carbon disulfide solutions were studied in a KBr cell of 0.1 mm. thickness with no comparison cell in the reference beam. The frequency measurements are estimated to have an uncertainty of less than ± 2 cm.$^{-1}$ and the slit width used in the high resolution studies was 5 cm.$^{-1}$. The exact maxima for the acetates are as follows: I, 1244 cm.$^{-1}$; II, 1248, 1238 cm.$^{-1}$; III, 1248, 1240 cm.$^{-1}$; IV, 1247, 1235, 1213 cm.$^{-1}$. The value for cholestanyl acetate is 1243 cm.$^{-1}$ and for epicholestanyl acetate are 1257, 1247, 1237 cm.$^{-1}$. The spectra of the decalols were determined n carbon disulfide solution at a concentration of 10 g. per liter and at a cell thickness of 0.9 mm. on a Baird infrared spectrophotometer equipped with a NaCl prism. The exact maxima for the decalols are as follows: I, 1057, 1029 cm.$^{-1}$; II, 1050, 1012 cm.$^{-1}$; III, 1062, 1022 cm.$^{-1}$; IV, 1007 cm.$^{-1}$.

Acknowledgment.—The authors wish to express their appreciation to Professor George C. Pimentel for his kind coöperation during the progress of this work.

(9) R. H. Baker and R. D. Schuetz, THIS JOURNAL, **69**, 1250 (1947).

(10) F. W. Mitchell and J. M. O'Gorman, *Anal. Chem.*, **20**, 315 (1948).

(11) R. T. Arnold, H. Klug, J. Sprung and H. Zaugg, THIS JOURNAL, **63**, 1161 (1941).

(12) H. Leroux, *Compt. rend.*, **140**, 590 (1905).

(13) *Anal.* Calcd. for $C_{12}H_{20}O_2$: C, 73.43; H, 10.27. Found: C, 72.98; H, 10.93.

CHEMICAL LABORATORY
UNIVERSITY OF CALIFORNIA
RADIATION LABORATORY
UNIVERSITY OF CALIFORNIA
BERKELEY 4, CALIFORNIA

Effect of Xanthylation on the Recovery of DNP-Amins Acids from Acid Protein Hydrolysates[1]

BY SHERMAN R. DICKMAN AND R. OWEN ASPLUND

RECEIVED JULY 21, 1952

Thompson[2] observed the destruction of DNP-amino acids after acid hydrolysis in the presence of tryptophan or the protein lysozyme which contains 10.6% tryptophan.[3] The acid stability of dixanthyltryptophan[4] prompted us to determine recoveries of N^5-DNP-L-lysine and of di-DNP-L-lysine from DNP-lysozyme and xanthyl-DNP-lysozyme. As shown in Table I recovery of N^5-DNP-L-lysine was 56% of theoretical from DNP-lysozyme and 91% from xanthyl-DNP-lysozyme after the substituted protein was refluxed for twenty-four hours in 6 *N* HCl. Similarly, recovery of di-DNP-L-lysine was 78% of the theoretical from DNP-lysozyme and 97% from xanthyl-DNP-lysozyme. The recovery of approximately one mole

(1) This work supported in part by a research grant from Armour and Co.

(2) A. Thompson, *Nature*, **168**, 390 (1951).

(3) J. C. Lewis, N. S. Snell, D. J. Hirschmann and H. Fraenkel-Conrat, *J. Biol. Chem.*, **186**, 23 (1950).

(4) W. L. Westcott and S. R. Dickman, unpublished.