was extracted with methylene chloride; the extract was washed with water, dilute hydrochloric acid, 5% sodium bicarbonate solution, and dried over sodium sulfate. Evaporation of the solvent gave 20.20 g of solid product which was recrystallized from aqueous methanol, mp 91–93°.

Anal. Calcd for $C_{18}H_{29}NO$: C, 78.40; H, 10.61; N, 5.09. Found: C, 78.29; H, 10.90; N, 5.04.

N-Cyclohexylcarbonyl-N-methyl-1-adamantanamine-4α,6α-diol (25).—The methylene chloride extract residue from a 2.0-g fermentation of **24** was chromatographed over 100 g of Florisil. The column was eluted by the gradient method with 4 l. of solvent, SSB containing increasing amounts of acetone from 0 to 40%. Fractions of 110 ml each were collected, and the residues were examined by tlc.[8] Fractions 25–29 were pooled and recrystallized from acetone to yield 0.174 g of **25**: mp 190–191°; ν_{OH} 3400, $\nu_{N-C=O}$ 1620 cm^{-1} in Nujol.

Anal. Calcd for $C_{18}H_{29}NO_3$: C, 70.32; H, 9.51; N, 4.56. Found: C, 70.31; H, 9.51; N, 5.35.

Fractions 33–37 gave another material which also analyzed for a diol. This structure has not been determined.

N-(1-Adamantyl)phthalimide (26).—A mixture of 7.5 g of 1-adamantanamine, 10.0 g of phthalic anhydride, and 100 ml of pyridine was heated at 90° for 15 min; 100 ml of acetic anhydride was added; and the mixture was again heated at 90° for 1 hr. After cooling and stirring with 500 ml of water for 1 hr the product was recovered by filtration, washed with water, and crystallized from methanol: yield, 2.13 g; mp 140–143°.

Anal. Calcd for $C_{18}H_{19}NO_2$: C, 76.84; H, 6.81; N, 4.98. Found: C, 76.75; H, 7.02; N, 5.01.

N-(4α,6α-Dihydroxy-1-adamantyl)phthalimide (27).—The filtered beer (10 l.) from the conversion of 2.0 g of **26** was poured over a column of 300 g of CAL carbon.[9c] The column was eluted first with 10 l. of methanol, followed with 5 l. of ethyl acetate, and finally 5 l. of chloroform. Thin layer chromatography[8] showed that the chloroform eluate contained the product. The residue therefrom was chromatographed over 100 g of Florisil. Elution was by the linear gradient method with 4 l. of solvent SSB containing increasing amounts of acetone from 0 to 40%; cuts were *ca.* 110 ml each. The product eluted in fractions 23–28 was recrystallized from acetone–hexane: mp 218–220°.

Anal. Calcd for $C_{18}H_{19}NO_4$: C, 68.99; H, 6.11; N, 4.47. Found: C, 68.81; H, 6.39; N, 4.21.

Registry No.—**2**, 16790-57-1; **3**, 778-10-9; **4**, 16790-59-3; **5**, 16790-60-6; **5′**, 16790-61-7; **6′**, 16790-62-8; **7**, 16790-63-9; 1-adamantanamine formic acid salt, 16790-64-0; N-formyl-1-adamantanamine, 3405-48-9; N-methyl-1-adamantanamine hydrochloride, 3717-39-3; **8**, 16790-67-3; free base of **9**, 16790-68-4; **10**, 16790-69-5; **11**, 16790-70-8; **11′**, 16790-71-9; **12**, 16790-72-0; **13**, 16790-73-1; **14**, 16790-74-2; **14′**, 16790-75-3; **15**, 16790-76-4; **16**, 16790-77-5; **17′**, 16790-78-6; **19**, 3717-37-1; **20**, 16790-80-0; **21**, 16790-81-1; **22**, 16790-82-2; **23**, 16790-83-3; **24**, 16790-84-4; **25**, 16790-85-5; **26**, 16808-41-6; **27**, 16790-86-6.

Acknowledgment.—We thank J. R. Heald, I. N. Pratt, S. L. Towne, H. M. Woltersom, J. M. Noteboom, and M. J. Sutton for technical assistance; N. H. Knight and associates for microanalyses; and G. Slomp and associates for some of the nmr spectra.

The Microbiological Hydroxylation of 1-Benzoyl-*trans*-decahydroquinoline. Determination of Structure, Stereochemistry, and Absolute Configuration of the Products

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Received February 7, 1968

Microbiological hydroxylation of (±)-1-benzoyl-*trans*-decahydroquinoline [(±)-**2**] with *Sporotrichum sulfurescens* has been shown to give (4a*S*,5*S*,8a*R*)-1-benzoyl-*trans*-decahydroquinolin-5-ol (**3**), (±)-1-benzoyl-*trans*-decahydroquinolin-6-ol (**4**), and (4a*S*,7*S*,8a*S*)-1-benzoyl-*trans*-decahydroquinolin-7-ol (**5**) in a total yield of 80–90%. Under the same conditions hydroxylation of (+)-**2** gave optically pure (+)-**5** and (4a*S*,6*S*,8a*S*)-1-benzoyl-*trans*-decahydroquinolin-6-ol [(+)-**4**] in a ratio of 35:65. Hydroxylation of (−)-**2** gave optically pure (−)-**3** and (4a*R*,6*R*,8a*R*)-1-benzoyl-*trans*-decahydroquinolin-6-ol [(−)-**4**] in a ratio of 87:13. Various chemical modifications of these products were carried out in order to determine their structures and stereochemistry and included the conversions of **3**, (+)-**5**, and (+)-**4** into (4a*S*,8a*R*)-*trans*-decahydroquinolin-5-one (**24**), (4a*S*,8a*S*)-*trans*-decahydroquinolin-7-one (**26**), and (4a*S*,8a*S*)-*trans*-decahydroquinolin-6-one (**25**), respectively. Application of the octant rule to the optical rotatory dispersion curves of the latter compounds allowed assignment of absolute configurations to the hydroxylation products. The absolute configurations of the parent molecules, (−)-*trans*-decahydroquinoline [(−)-**1**] and (+)-*trans*-decahydroquinoline [(+)-**1**], can be assigned as (4a*R*,8a*S*)-*trans*-decahydroquinoline and (4a*S*,8a*R*)-*trans*-decahydroquinoline, respectively.

The increasing number of substrates which are hydroxylated by the microorganism *Sporotrichum sulfurescens* provides an opportunity to explore further the relationships of the substrate molecules to the enzymic hydroxylation site. A recent proposal[1] has suggested that an electron-rich center of the substrate molecule provides an attachment site for the hydroxylating enzyme and thus facilitates oxygenation at some point in a saturated portion of the molecule. The approximate distance of this point from the attachment site was suggested to be 5.5 Å. Among the electron-rich centers which have been found useful are the alcohol[1] and the amide[2–4] functional groups. It seemed possible that additional information concerning the stereochemical relationship of the substrate molecule to the site of oxygenation could be obtained from examination of the oxygenated products. Some information concerning the stereochemistry of hydroxylation of the steroid nucleus has been gathered.[5] It is known,

(1) G. S. Fonken, M. E. Herr, H. C. Murray, and L. M. Reineke, *J. Amer. Chem. Soc.*, **89**, 672 (1967).

(2) G. S. Fonken, M. E. Herr, H. C. Murray, and L. M. Reineke, *J. Org. Chem.*, **33**, 3182 (1968).

(3) R. A. Johnson, M. E. Herr, H. C. Murray, and G. S. Fonken, *ibid.*, **33**, 3187 (1968); R. A. Johnson, M. E. Herr, H. C. Murray, L. M. Reineke, and G. S. Fonken, *ibid.*, **33**, 3195 (1968)

(4) M. E. Herr, R. A. Johnson, H. C. Murray, L. M. Reineke, and G. S. Fonken, *ibid.*, **33**, 3201 (1968).

(5) C. Tamm, *Angew. Chem. Intern. Ed. Engl.*, **1**, 178 (1962).

for example, that both 11α hydroxylation of steroids by *Rhizopus nigricans*[6] and 11β hydroxylation by the adrenal glands[7] proceed by substitution of the hydrogen by oxygen without changing the configuration of the remaining 11-hydrogen atom.

In studies of the oxygenation of various amides, we have found that hydroxylation of (±)-1-benzoyl-*trans*-decahydroquinoline [(±)-**2**] with *S. sulfurescens* gave a mixture of monohydroxy products, some of which were optically active, in excess of 80% yield. Formation of optically active products in yields greater than 50% suggested that the hydroxylating enzyme of the organism has a specificity for oxidative attack at different methylene groups of the enantiomers of the substrate. Thus, both enantiomers are hydroxylated, but necessarily at different positions in order to produce optically active products. This bioconversion reaction has therefore been studied in depth with the hope of obtaining stereochemical information about the hydroxylation process. The characterization, the determination of structure and stereochemistry, and the determination of the absolute configuration of the products are included in the following discussion.

Characterization of Products.[8]—The products from bioconversion of (±)-1-benzoyl-*trans*-decahydroquinoline [(±)-**2**] with *S. sulfurescens* were extracted from the filtered beer with methylene chloride. Initial attempts at separation of the products by column chromatography were only partially successful. Two products, **3** (mp 121–123°, [α]D −94°) and **4** (mp 149–151°, [α]D +3°), were obtained from the chromatography fractions. The optical rotation of **3** indicated that it had been obtained with some degree of optical purity; however, no conclusion about the optical purity of **4** could be drawn from its rotation. In addition to these two compounds, a third (**5**, mp 185–187°, [α]D +132°) was shown to be present in the later column fractions by paper chromatography. It appeared that both the problems of characterization of the bioconversion products and of the determination of their optical purity could be attacked more easily if the racemic substrate [(±)-**2**] could be resolved into the (+) and (−) enantiomers.

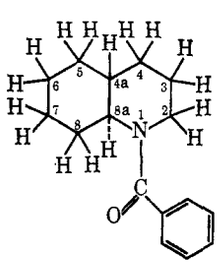
Resolution of racemic *trans*-decahydroquinoline [(±)-**1**] with *d*-tartaric acid[9] gave (−)-**1**, which was converted into (+)-**2** (+139°). Oxygenation of (+)-**2** with *S. sulfurescens* gave a mixture of two products, which were partially separated by column chromatography over silica gel. The two optically pure products, (+)-**5** (mp 185–186°, [α]D +137°) and (+)-**4** (mp 138–139°, [α]D +115°), had the same R_f values on paper chromatograms as did the optically impure compounds **5** and **4**, respectively.

Resolution of (±)-**1** with *d*-(α)-bromocamphor-π-sulfonic acid[10] gave (+)-**1** from which the benzamide (−)-**2** ([α]D −145°) was prepared. Hydroxylation of (−)-**2** with *S. sulfurescens* gave two products, which were separated by chromatography on a silica gel column. These optically pure products, (−)-**3** (mp 125–127°, [α]D −109°) and (−)-**4** (mp 136–138°, [α]D −112°), had the same R_f values on paper chromatograms as did the optically impure compounds **3** and **4**, respectively.

The interrelationships between the products, suggested by paper chromatography, were confirmed by infrared and nmr spectra and are outlined in Scheme I. The pairs of compounds **3** and (−)-**3**, **5** and (+)-**5**, (+)-**4** and (−)-**4** each have identical infrared spectra. It was necessary to compare compounds (+)-**4** and **4** by the means of a solution spectra since, in the solid phase, their infrared spectra differed. Nmr spectra of (+)-**4** and **4** also are identical.

The ratios of products in this bioconversion are of interest if enzyme specificity for the enantiomeric forms of the substrate is to be considered. Estimates of product ratios can best be made from the yields obtained when the resolved forms of the substrate were oxygenated. The ratio of (+)-**5** to (+)-**4** is 35:65 as estimated from paper and thin layer chromatograms of the product mixtures. The ratio of (−)-**3** to (−)-**4** is 87:13 as determined from the yields of the two products after separation on a silica gel column. These ratios between products also should exist when racemic substrate is used. From this the ratio of (+)-**4** to (−)-**4** can be estimated as 83:17, *part* of which is represented by the racemate **4**.

Structure and Stereochemistry.—The substrate molecule, (±)-1-benzoyl-*trans*-decahydroquinoline (**2**), contains 16 geometrically different hydrogen atoms. When the enantiomers of **2** are considered, there are 32 stereoisomeric possibilities to choose from in assigning structures to the four products obtained from bioconversion of **2**. These four products are known to consist



(±)-**2**

of an enantiomeric pair of alcohols, (+)-**4** and (−)-**4** (which also form **4**), and two optically active products, **3** and **5**. The problem of structural determination is therefore essentially that of determining three unknowns. The 32 possible structures were quickly reduced to 16 by oxidation of each alcohol to a ketone. Formation of a ketone, in itself, eliminates placing the hydroxy groups at carbon atoms 4a, 8a, and 2 of the decahydroquinoline nucleus since the first two posi-

(6) M. Hayano, M. Gut, R. I. Dorfman, O. K. Sebek, and D. H. Peterson, *J. Amer. Chem. Soc.*, **80**, 2336 (1958).

(7) E. J. Corey, G. A. Gregoriou, and D. H. Peterson, *ibid.*, **80**, 2338 (1958).

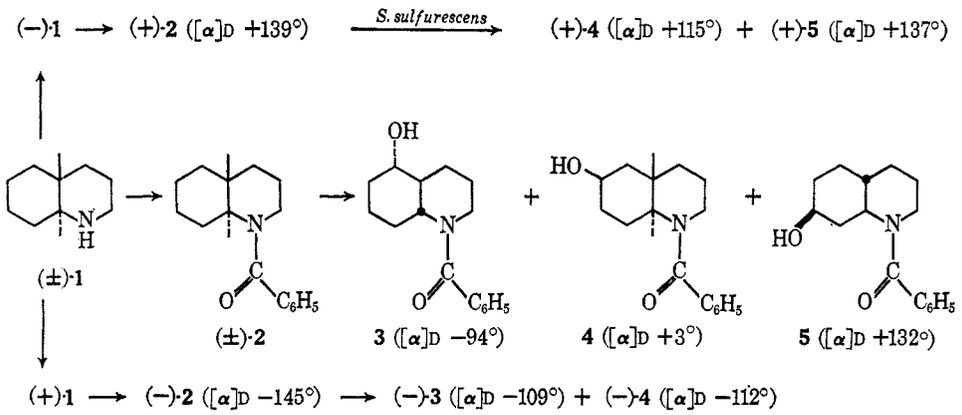
(8) Several of the compounds encountered in this work have been obtained in varying degrees of optical purity. We have adopted the following system for numbering these compounds in this paper. For compounds **1** through **5**, placement of (±)- before the number indicates that the compound is a racemate. When (+)- or (−)- is placed before the number, the optically pure enantiomer is indicated. When no sign is placed before the number, an optically impure (but still optically active) compound is indicated. Compounds **6–26** involve chemical transformations and in most cases the optical nature of the compound is apparent from the discussion. Signs, in accord with the above system, are placed before these numbers only when they add to the clarity of the discussion.

In the structural formula, the use of solid or dotted lines to depict the ring junction hydrogen atoms (4a and 8a) indicates that the compound is a racemate. Use of heavy dots to depict ring junction stereochemistry indicates that the compound is optically active and in all cases is indicative of the correct absolute configuration of the molecule.

(9) A. Popovici, C. F. Geschickter, E. L. May, and E. Mosettig, *J. Org. Chem.*, **21**, 1283 (1956).

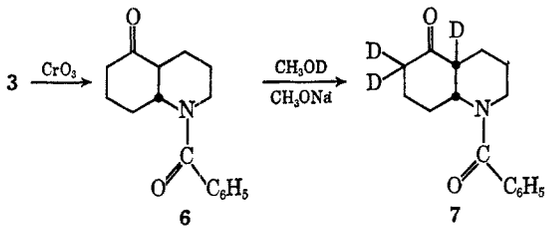
(10) L. Mascarelli and F. Nigrisoli, *Gazz. Chim. Ital.*, **45**, 106 (1915).

SCHEME I



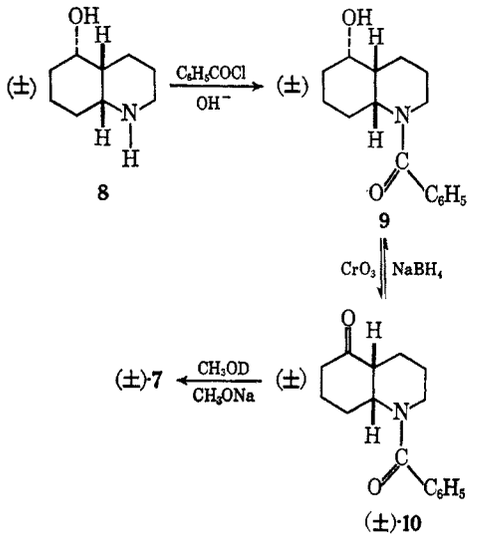
tions are tertiary and the latter would be an imide. In addition, nmr spectra of the ketones clearly eliminated carbon atoms 3 and 8 as positions of oxygenation, since the very characteristic signal expected for protons (at C-2 and C-8a) adjacent to both nitrogen and carbonyl were not observed.

The remaining possible positions of oxygenation were further differentiated by deuterium-exchange experiments with the ketones. The ketone (**6**), obtained upon oxidation of **3** with Jones reagent,[11] undergoes ex-



change of three hydrogen atoms by deuterium atoms in methyl alcohol-*d* with sodium methoxide catalyst. The incorporation of deuterium was demonstrated by an increase in the molecular ion peak from 257 to 260 in the mass spectra of the two compounds. It will be shown below that, in addition to isotopic exchange, the deuterated product (**7**) has an inverted configuration at the 4a position. To undergo exchange of only three hydrogens by deuterium, the carbonyl group must be at either C-4 or C-5 in **6**, since C-8 has already been eliminated as a possible site for oxygen. Under the same conditions, the other two ketones both underwent exchange of four hydrogens by deuterium. Consequently one ketone must be a C-6 ketone and the other a C-7 ketone, since the only other position having four α hydrogens (C-3) has been eliminated by the nmr spectra. These two ketones will be discussed more fully later.

The position of the carbonyl in ketone **6**, now known to be adjacent to C-4a, could be established by the synthesis of either the C-4 or C-5 ketone.[12] A simple route to the 5 ketone seemed feasible using (±)-5α-hydroxy-*cis*-decahydroquinoline (**8**), a compound readily available by the catalytic reduction of 5-hydroxyquinoline,[13] as a starting point. The presence of a *cis*-ring junction in **8** was not serious since the ring system of the 5 ketone could be equilibrated to the more stable ring juncture. Reaction of **8** with benzoyl chloride in the presence of an excess of sodium hydroxide gave the hydroxy amide **9** directly. Oxidation of **9** with Jones reagent[11] gave racemic keto amide (±)-**10**. The *cis*-ring junction was



shown to remain in (±)-**10** by reduction of the ketone with sodium borohydride, which gave hydroxy amide **9** starting material as the only product isolated (70%). Deuterium exchange with (±)-**10** was carried out, and the product had a mass spectrum identical with that of **7**. The position of the carbonyl in **6** must be at C-5 and likewise, the hydroxyl group of compound **3** must be at C-5.

Several additional points of interest concerning the ketone **6** may be noted. Reaction of **6** with sodium methoxide in methanol resulted in isomerization to the *cis* ketone **10**, confirming that the *cis*-ring system is the more stable for this molecule. A small amount of **10** also was separated from **6** when the products of the Jones oxidation of **3** were chromatographed on Florisil. Reduction of ketone **6** with sodium borohydride gave two alcohols, one of which was identical with **3**. The second alcohol (**11**) is assumed to be the C-5 epimer of alcohol **3**. It had been hoped that this reduction would be stereospecific and would indicate the configuration of the hydroxyl group in **3**.

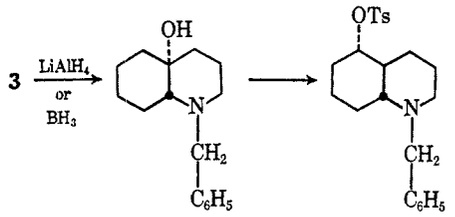
A second method for distinguishing between an axial and an equatorial hydroxyl group is that of measuring the half-band width of the nmr signal of the proton of

(11) K. Bowden, I. M. Heilbron, E. R. H. Jones, and B. C. L. Weedon, *J. Chem. Soc.*, 39 (1946).

(12) When this synthesis was carried out, the deuterium-exchange experiments had not been completed and we felt that on the basis of previous experience[1] oxygenation at C-4 and C-5 had probably occurred and that the synthesis of either would identify one compound.

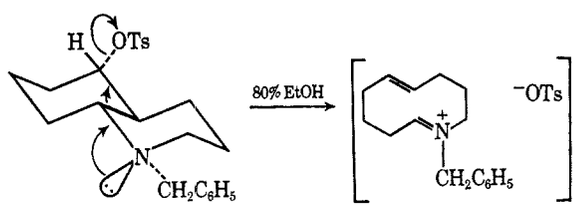
(13) C. A. Grob and H. R. Kiefer, *Helv. Chim. Acta*, **48**, 799 (1965).

the alcohol carbon.[14] This method takes advantage of the greater coupling constants between axial protons and assigns the axial configuration to protons having a half-band width of 20 cps or greater and an equatorial configuration to those having a half-band width of about 8 cps.[14] The signal of the C-5 proton in the nmr spectra of **3** and several of its derivatives is very broad, suggesting an axial configuration and therefore an equatorial configuration for the hydroxyl group. This stereochemical assignment was confirmed by carrying out a fragmentation reaction analogous to those described by Grob and coworkers.[15] To accomplish this, alcohol–amide **3** was reduced with either lithium aluminum hydride or diborane to the benzylamine (**12**). We have found that diborane is an excellent agent for the reduction of benzamides to benzyl amines, a reaction which often results in additional hydrogenolysis of the benzyl group when done with lithium aluminum hydride.



12 **13**

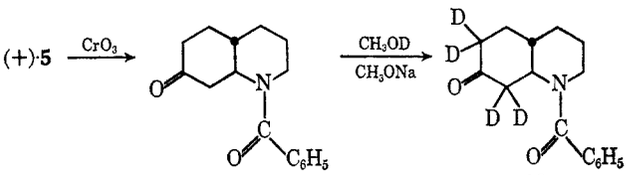
The preparation of the tosylate (**13**) of **12** gave a compound, which, if the tosyl group is equatorial, differs from a compound described by Grob and coworkers[15] only in having a N-benzyl instead of a N-methyl group. Fragmentation of this molecule would then be expected when warmed in 80% ethanol.[15] The fragmentation product (**14**) may be expected to hydrolyze to



13 **14**

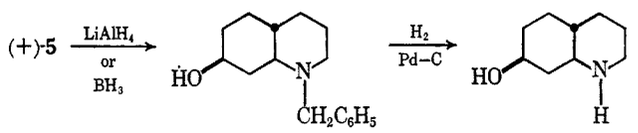
an amino aldehyde under the conditions of the reaction.[15] When **13** was heated to 75–78° in 80% ethanol, the partially insoluble compound slowly went into solution. The oil isolated from the reaction had a multiplet (integrating for 1.7 hydrogens) at 5.28 (δ) ppm in the nmr spectrum characteristic of olefinic protons, which would result in large amounts only from the fragmentation reaction. This firmly establishes that the C-5 hydroxyl group in the bioconversion product **3** has an equatorial configuration.

As outlined previously, the other hydroxylic bioconversion products also were converted to ketones and were submitted to deuterium-exchange reactions. Thus, oxidation of alcohol (+)-**5** with Jones reagent[11] gave ketone **15** of molecular weight 257 by mass spectrometry. Deuterium exchange with ketone **15** gave ketone **16** of molecular weight 261 by mass spectrometry, showing an exchange of four hydrogens by deuterium. A polymorphic crystalline form of ketone **15** was obtained from an early experiment in which the ketone was treated with a dioxane–ether solution of hydrogen chloride.



15 **16**

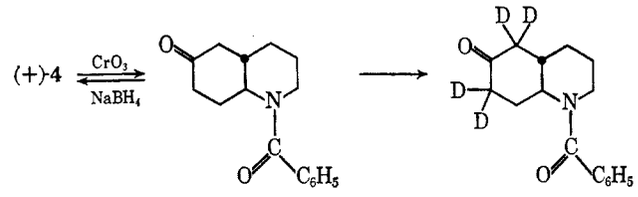
Reduction of bioconversion alcohol (+)-**5** with lithium aluminum hydride gave a mixture of benzyl amine **17** and amine **18**. When diborane was used



17 **18**

as the reducing agent, the benzyl amine **17** was the only product isolated. Compound **17** had a wide melting point range which could not be improved by varying the crystallization procedure. Elemental analysis showed an additional mole of water present in the product, presumably as a hydrate. Hydrogenolysis of the hydrate of **17** over palladium on carbon gave the amine **18**. This reaction sequence enabled us to compare compound **18** with the 7α-hydroxy-*trans*-decahydroquinoline [(±)-**18**] prepared and described by Grob and Wilkens.[16] It was necessary to compare the two samples by solution spectra since the biconversion product (**18**) is optically active while the synthetic sample of Grob and Wilkens[16] is not. The two compounds have identical infrared spectra in chloroform solution. This identity establishes the structure of **18** as being the 7-hydroxy-*trans*-decahydroquinoline having an equatorial hydroxyl configuration.

By elimination, the remaining bioconversion alcohols (**4**, obtained in both enantiomeric forms and as a racemate) must have the hydroxyl group at the 6 position. Oxidation of (+)-**4** with Jones reagent[11] gave ketone **19**, which incorporated four deuterium atoms dur-



19 **20**

ing exchange with methyl alcohol-*d*. The deuterated ketone (**20**) had a molecular ion peak at 261 mass units in the mass spectrum.

To determine the configuration of the C-6 hydroxyl group in (+)-**4**, we have relied on the stereoselective reduction of sterically unhindered ketones to equatorial alcohols by sodium borohydride.[17] Reduction of ketone **19** with sodium borohydride in ethanol gave a single alcohol in 74% yield, which was identical

(14) R. U. Lemieux, R. K. Kullnig, H. J. Bernstein, and W. G. Schneider, *J. Amer. Chem. Soc.*, **80**, 6098 (1958).

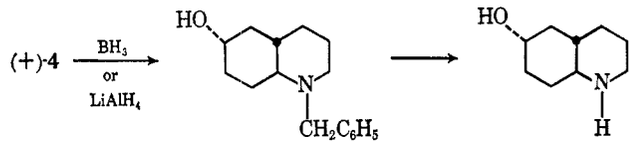
(15) C. A. Grob, H. R. Kiefer, H. J. Lutz, and H. J. Wilkens, *Helv. Chim. Acta*, **50**, 416 (1967).

(16) C. A. Grob and H. J. Wilkens, *ibid.*, **48**, 808 (1965). We thank Professor Grob for a sample of 7α-hydroxy-*trans*-decahydroquinoline.

(17) D. H. R. Barton, *J. Chem. Soc.*, 1027 (1953).

with bioconversion product (+)-**4**. We consider this proof that the hydroxyl group of (+)-**4** has the equatorial configuration.

For the purpose of determining the optical rotatory dispersion curve of the simple unsubstituted 6-ketodecahydroquinoline molecule, several modifications of alcohol–amide (+)-**4** were necessary. First, (+)-**4** was reduced to the benzyl amine (**21**) with diborane.

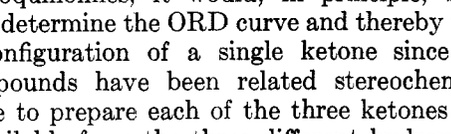


21 **22**

As before a crystalline hydrate of the benzyl amine (**21**) was obtained, as shown by elemental analyses and the infrared spectrum (see Experimental Section). Hydrogenolysis of the benzyl group of **21** was rapid over 5% palladium on carbon, giving the optically active amino alcohol **22**. Reduction of (+)-**4** with lithium aluminum hydride gave a mixture of benzyl amine **21** and amine **22**. A similar reduction of the racemic **4** gave (±)-**22**.

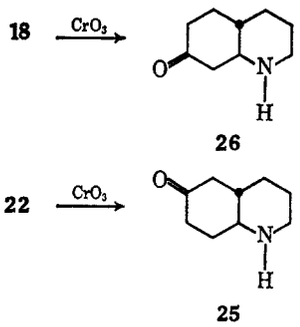
Absolute Configuration.—The application of the octant rule[18] to the optical rotatory dispersion (ORD) curves of many substances has been used extensively[19] in the determination of the absolute configuration of optically active molecules. In the present study optically active oxygenated derivatives in the *trans*-decahydroquinoline series have been obtained, which require only a few additional modifications in order to give keto-*trans*-decahydroquinoline molecules. Such molecules are closely analogous to the *trans*-decalones and similarly should give ORD curves which would be useful in the determination of their absolute configurations. This would in turn determine the absolute configuration of all the optically active compounds encountered in this study. Such information is of value when considering the relationships of the substrate molecules to the enzyme hydroxylation sites.

In considering the preparation of the keto-*trans*-decahydroquinolines, it would, in principle, be sufficient to determine the ORD curve and thereby the absolute configuration of a single ketone since all of the compounds have been related stereochemically. We chose to prepare each of the three ketones potentially available from the three different hydroxy compounds obtained in the bioconversion because little has been reported on the ORD curves of ketonic compounds containing amines. The 5 ketone (**24**) was prepared by repeated Jones oxidation of the amino alcohol **23**, which



23 **24**

in turn had been prepared by the hydrogenolysis of benzyl amino alcohol **12**. We believe that the *trans*-ring juncture in **24** is the more stable even though it gives the keto amide (−)-**10** of the *cis*-decahydroquinoline series when treated with benzoyl chloride in pyridine. In the closely related 4-ketodecahydroquinoline series, the *trans*-ring juncture is the more stable.[20] Correlation of our ORD results also are consistent only if **24** is assigned the *trans*-ring junction. Similar oxidations of the amino alcohols **18** and **22** gave the 7 ketone (**26**) and the 6-ketone (**25**), respectively.

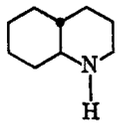


26

25

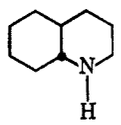
The ORD curves of the three ketones were determined in methanol solution and are illustrated in Figure 1. The curves obtained for ketones **24** and **25** appear normal and the amplitudes of +4720 and +4640°, respectively, are similar to those observed for the *trans*-decalones. *trans*-1-Decalone has an ORD amplitude of ±4000°[21] while *trans*-2-decalone has an amplitude of ±5400°.[22] However, ketone **26**, which was expected to have an ORD curve similar to that of ketone **25** shows a very weak, positive Cotton effect of +28° amplitude. The reason for this unusual curve is not apparent at the present time. The ultraviolet spectrum of ketone **26** is normal for an isolated carbonyl group, in that it has an absorption maximum at 281 mμ (ε 18).

Application of the octant rule[18] to the ORD curves of the three ketones leads to the following assignments of absolute configuration: **24** becomes (4a*S*,8a*R*)-*trans*-decahydroquinolin-5-one,[23] **25** becomes (4a*S*,8a*S*)-*trans*-decahydroquinolin-6-one, and **26** becomes (4a*S*,8a*S*)-*trans*-decahydroquinolin-7-one. These configurations are shown by the drawings used above. By employing the structural and stereochemical relationships used throughout the preceding discussion, the two epimers of *trans*-decahydroquinoline may be assigned the absolute configurations as shown. Similarly, ab-



(−)-**1**

(−)-*trans*-decahydroquinoline ([α]D −4.6°)[9] or (4a*R*,8a*S*)-*trans*-decahydroquinoline



(+)-**1**

(+)-*trans*-decahydroquinoline ([α]D +4.8°)[10] or (4a*S*,8a*R*)-*trans*-decahydroquinoline

(18) W. Moffitt, R. B. Woodward, A. Moscowitz, W. Klyne, and C. Djerassi, *J. Amer. Chem. Soc.*, **83**, 4013 (1961).

(19) P. Crabbe, "Optical Rotatory Dispersion and Circular Dichroism in Organic Chemistry," Holden-Day, San Francisco, Calif., 1965.

(20) E. Mistryukov, *Izv. Akad. Nauk SSR, Otd. Khim. Nauk*, 929 (1963)

(21) Depending on which enantiomer is used, the amplitude will be + or −4000°.

(22) W. Klyne, *Experientia*, **20**, 349 (1964).

(23) Although use of the *R* and *S* nomenclature is sufficient to indicate the nature of the ring juncture in these compounds, inclusion of the terms *cis* and *trans* in the compound name eliminates the task of determining this feature for every name by use of the rules. For this reason we suggest that *cis* and *trans* be retained in the naming of these compounds.

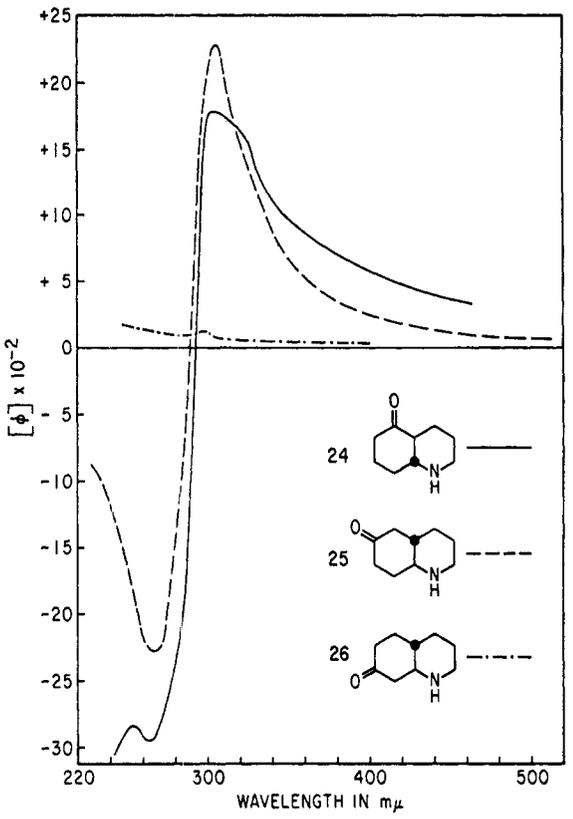


Figure 1.

solute configurations may be assigned to all other related optically active molecules in this study, and these assignments are given by the nomenclature used throughout the Experimental Section.

Experimental Section[24]

Biotransformation Process.—The culture used in these experiments was *Sporotrichum sulfurescens* V. Beyma (ATCC 7159). The biotransformation procedure has been described previously.[3]

Isolation of the Products from Bioconversion of (±)-1-Benzoyl-*trans*-decahydroquinoline [(±)-2] with *S. sulfurescens*. A. (4a*S*,5*S*,8a*R*)-1-Benzoyl-*trans*-decahydroquinolin-5-ol (3) and (±)-1-Benzoyl-*trans*-decahydroquinolin-6-ol (4).—The oily extracts from the 125-l. bioconversion of (±)-2 (25 g, 0.103 mol) were chromatographed on a column of Florisil (2.5 kg) packed with Skellysolve B. The following fractions (2.0-l. vol) were collected by elution with 25% (v/v) acetone in Skellysolve B. Fractions 19 and 20 were eluted with acetone (Table I). On the basis of infrared spectra, fractions 13 and 14 were combined in acetone, decolorized with activated charcoal, and crystallized as colorless crystals (3.925 g, first and second crops), mp 122–124°. A third crop gave 1.703 g (total 5.628 g, 0.0219 mol, 21%), mp 121–123°. Two recrystallizations from acetone–Skellysolve B gave colorless crystals of **3**: mp 121–123°; $[\alpha]_D$ −94° (*c* 0.648, chloroform); ν_{OH} 3350, $\nu_{C=O}$ 1610, $\nu_{C=C}$ 1600, 1575, 1525, 1500, ν_{C-O} 1210, 1125, 1065, 1010, $\nu_{C_6H_5}$ 785, 730, 700 cm^{-1} in Nujol.

Anal. Calcd for $C_{16}H_{21}NO_2$: C, 74.10; H, 8.16; N, 5.40. Found: C, 74.11; H, 8.27; N, 5.67.

This alcohol was shown not to be a polymorph of alcohol **4**, mp 149–151°, by comparison of infrared spectra prepared from addition of a chloroform solution to KBr with subsequent pellet preparation.

(24) Melting points were determined on a calibrated Fisher-Johns hot stage and are corrected. Magnesium sulfate was used as the drying agent unless indicated otherwise. Infrared spectra were determined with either a Perkin-Elmer Infracord or Model 421 spectrophotometer. The nmr spectra were determined at 60 Mc with a Varian Model A-60 spectrometer, using tetramethylsilane as an internal standard. Mass spectra were determined on an Atlas CH4 instrument. The optical rotatory dispersion curves were obtained on a Cary Model 60 spectrophotometer.

TABLE I

Fraction.	Wt, g
12	1.02
13	4.52
14	3.33
15	1.93
16	2.69
17	3.53
18	2.25
19	1.54
20	2.10
Total	22.91

Fraction 17 (mp 131–134°) was recrystallized from acetone–Skellysolve B, giving colorless crystals (2.553 g), mp 128–135°. A portion of recrystallized fraction 17 was again recrystallized from acetone–Skellysolve B. Initially colorless crystals formed, but after standing for 5 days at room temperature the crystalline mass was composed of shiny, colorless transparent flat needles (mp 175–180°) covering a mass of opaque, chunky crystals (mp 143–148°). The two forms have different infrared spectra in Nujol. It was then discovered that the shiny crystals were more soluble in acetone. Fractions 16, 18, and 19 were recrystallized from acetone–Skellysolve B, giving a mixture of crystals (3.102 g). From this mixture a sample (0.364 g) of the product less soluble in acetone (100 ml) was obtained by decanting the acetone solution. Recrystallization of this sample from acetone gave colorless crystals, mp 148–150°. A final recrystallization from acetone gave crystals of **4** (0.212 g): mp 149–151°; $[\alpha]_D$ +3° (*c* 0.801, chloroform); ν_{OH} 3360, $\nu_{C=O,C=C}$ 1595, 1570, 1525, 1490, ν_{C-O} 1055, 1045, $\nu_{C_6H_5}$ 790, 735, 700 cm^{-1} in Nujol.

Anal. Calcd for $C_{16}H_{21}NO_2$: C, 74.10; H, 8.16; N, 5.40. Found: C, 74.22; H, 8.47; N, 5.82, 5.73.

B. Chromatography of a Large-Scale Bioconversion of Racemic 1-Benzoyl-*trans*-decahydroquinoline.—The extracts from three 250-l. bioconversions of (±)-**2** (total 150.0 g, 0.618 mol) was chromatographed on a Florisil column (6 kg) packed with Skellysolve B. A fraction (3 l.) of Skellysolve B was taken followed by four fractions of 10% acetone–Skellysolve B, 15 fractions of 20% acetone–Skellysolve B, five fractions of 25% acetone–Skellysolve B, and five fractions of 50% acetone–Skellysolve B. The products were found in fractions 14–29. Fractions 14–20 were pooled on the basis of their infrared spectra. The crude fraction weights are given in Table II. Fractions 14–20 were dissolved in acetone and decolorized with activated charcoal. Crystallization from acetone–Skellysolve B gave three crops of **3** as product: first crop, 22.097 g, mp 120–123°; second crop, 22.419 g, mp 123–125°; third crop, 5.230 g, mp 115–130°; total, 49.746 g (0.192 mol, 31%).

TABLE II

Fractions	Wt, g
14–20	58.01
21	8.57
22	11.35
23	11.14
24	9.70
25	19.15
26	21.06
27	8.54
28	4.47
29	1.64
Total	153.63 (0.593 mol, 96%)

Fractions 21–23 were combined in methylene chloride–benzene and rechromatographed on a silica gel column (2500 g) packed with benzene. No material eluted with 1% (ten 2-l. fractions) and 1.5% (nine fractions) methanol in benzene. With 2% methanol in benzene, the products were eluted with no sharp separation. On the basis of similar infrared spectra, fractions 25–27 (crude wt, 5.71 g) were combined and crystallized from acetone. The first crop (2.584 g) was colorless crystals, mp 179–185°. The second crop (1.571 g) was a mixture of products, mp 133–170°. Two recrystallizations of the first crop from acetone gave colorless, rectangular crystals of **5**: mp 185–187°; $[\alpha]_D$ +132°

(c 0.494, chloroform); infrared spectrum in Nujol is identical with that of (4a*S*,7*S*,8a*S*)-1-benzoyl-*trans*-decahydroquinolin-7-ol [(+)-5] from bioconversion of (+)-2.

Anal. Calcd for $C_{16}H_{21}NO_2$: C, 74.10; H, 8.16; N, 5.40. Found: C, 73.99; H, 8.19; N, 5.46.

From combined fractions 31–42 (crude wt, 12.089 g) of the silica gel column, crystals (first crop, 3.900 g, mp 146–149°; second crop, 3.192 g, mp 138–140°) were obtained from acetone–Skellysolve B. The infrared spectra in Nujol of the two crops were identical with that of **4**. The remaining fractions (28–30 8.762 g) were a mixture of **4** and **5**. Further purification of the remaining fractions from the initial chromatography has not been carried out.

(4a*R*,8a*S*)-1-Benzoyl-*trans*-decahydroquinoline [(+)-2].—(4a*R*, 8a*S*)-*trans*-Decahydroquinoline (obtained from 8.0 g of the *d*-tartrate)[9] was benzoylated by a Schotten–Baumann reaction in which the reactants were shaken with ice in a separatory funnel. The product was crystallized from Skellysolve B, giving crystals, mp 66–68°. A final crystallization from Skellysolve B gave colorless crystals of (+)-2: mp 68–69°; $[\alpha]_D$ +139° (c 0.764, chloroform).

Anal. Calcd for $C_{16}H_{21}NO$: C, 78.97; H, 8.70; N, 5.76. Found: C, 79.05; H, 8.49; N, 5.74.

Bioconversion of (4a*R*,8a*S*)-1-Benzoyl-*trans*-decahydroquinoline [(+)-2]. (4a*S*,6*S*,8a*S*)-1-Benzoyl-*trans*-decahydroquinolin-6-ol [(+)-4] and (4a*S*,7*S*,8a*S*)-1-Benzoyl-*trans*-decahydroquinolin-7-ol [(+)-5].—The dry methylene chloride extracts from two 125-l. bioconversions of (+)-2 (25.0 g each, 0.103 mol) were each chromatographed on a Florisil column (2 kg, 10.5 × 50 cm, 2-l. fractions) packed with Skellysolve B. Elution with 20–25% acetone in Skellysolve B gave 21.82 (0.0832 mol, 81%) and 25.09 g (0.0968 mol, 94%) of mostly crystalline material from the two columns. Chromatography (paper, vapor phase, thin layer) showed the product to consist of two major products distributed through all the column fractions. The two components were found to be separable, in part, by chromatography on silica gel with 1–3% methanol in benzene. Thus, for example, chromatography of 3.24 g of material chosen from the earlier fractions above (shown to be richer in less polar component) on a silica gel column (300 g, 3.8-cm diameter, 335-ml fractions) packed from a slurry in benzene gave no material in 25 fractions when eluted with 1% methanol in benzene. Elution with 2% methanol in benzene gave three fractions of less polar material (1.57 g), two fractions of a mixture (0.78 g) of the two products, and six fractions of more polar product (0.53 g). The separation of products was determined by tlc (silica gel, 20% methanol in benzene). The first three fractions of less polar product were combined and crystallized from acetone, giving a first crop (0.410 g) of crystals, mp 150° (softening), 183–188°. Two recrystallizations from acetone gave (+)-5 as colorless crystals: mp 185–186° with some sublimation; $[\alpha]_D$ +137° (c 0.678, chloroform); ν_{OH} 3360, $\nu_{C=O}$ 1605, $\nu_{C=C}$ 1595, 1570, 1555, 1495, $\nu_{C-O/other}$ 1290, 1115, 1065, $\nu_{C_6H_5}$ 775, 745, 700 cm^{-1} in Nujol.

Anal. Calcd for $C_{16}H_{21}NO_2$: C, 74.10; H, 8.16; N, 5.40. Found: C, 73.90; H, 8.21; N, 5.22.

Chromatography of 3.25 g of material from the latter fractions of the Florisil columns on silica gel (200 g, 3.8-cm diameter, 250-ml fractions) in benzene gave four fractions of a mixture of products and six fractions of pure more polar product. The latter fractions were combined in acetone, decolorized with activated charcoal, and crystallized from acetone–Skellysolve B, giving a first crop (0.811 g) of colorless crystals, mp 137–139°. Two recrystallizations from acetone–Skellysolve B gave (+)-4 as colorless feathers: mp 138–139°; $[\alpha]_D$ +115° (c 1.072, chloroform); ν_{OH} 3400, $\nu_{C=O}$ 1600, $\nu_{C=C}$ 1590, 1570, 1555, 1490, $\nu_{C-O/other}$ 1185, 1055, $\nu_{C_6H_5}$ 765, 745, 700 cm^{-1} in Nujol.

Anal. Calcd for $C_{16}H_{21}NO_2$: C, 74.10; H, 8.16; N, 5.40. Found: C, 73.99; H, 8.23; N, 5.41.

(4a*S*,8a*R*)-1-Benzoyl-*trans*-decahydroquinoline [(−)-2].—A sample of (4a*S*,8a*R*)-*trans*-decahydroquinoline (4.0 g, 0.0288 mol), prepared by basification of the *d*-[α]-bromocamphor-π-sulfonate,[10] was benzoylated under Schotten–Bauman conditions. The crude solid reaction product (7.117 g) was collected by filtration and washed with water. Crystallization from Skellysolve B gave 5.771 g (0.0238 mol, 82%) of crystals, mp 66–68°, in two crops. Recrystallization from Skellysolve B gave colorless, chunky crystals of (−)-2: mp 67–69°; $[\alpha]_D$ −145° (c 1.048, chloroform).

Anal. Calcd for $C_{16}H_{21}NO$: C, 78.97; H, 8.70; N, 5.76. Found: C, 79.01; H, 8.88; N, 5.70.

Bioconversion of (4a*S*,8a*R*)-1-Benzoyl-*trans*-decahydroquinoline [(−)-2]. (4a*S*,5*S*,8a*R*)-1-Benzoyl-*trans*-decahydroquinolin-5-ol [(−)-3] and (4a*R*,6*R*,8a*R*)-1-Benzoyl-*trans*-decahydroquinolin-6-ol [(−)-4].—The dry methylene chloride extract from the 10-l. bioconversion of (−)-2 (2.0 g, 0.00823 mol) was chromatographed on a Florisil column (3.8 × 34 cm). Elution with 20% acetone in Skellysolve B gave crude crystalline solid (1.50 g, 0.00578 mol, 70%). This material was rechromatographed on a silica gel column (120 g) packed with benzene. No material was eluted with 20 fractions (100 ml each) of 1% methanol in benzene. Elution with 2% methanol in benzene gave several fractions of crystalline solid. Recrystallization of fraction 30 from acetone–Skellysolve B gave colorless crystals (0.273 g), mp 123–125°. Two recrystallizations from acetone–Skellysolve B gave (−)-3 as colorless needles: mp 125–127°; $[\alpha]_D$ −109° (c 0.750, chloroform); infrared spectrum in Nujol is identical with that of the compound **3**, mp 121–123°, obtained from bioconversion of the racemic substrate.

Anal. Calcd for $C_{16}H_{21}NO_2$: C, 74.10; H, 8.16; N, 5.40. Found: C, 73.53; H, 8.31; N, 5.20.

The extract from a second 10-l. bioconversion of (−)-2 (2.0 g, 0.00823 mol) was chromatographed directly on a silica gel column (250 g) packed with benzene. Elution with 20 fractions (335 ml) of 1% methanol in benzene was followed by elution with 2% methanol in benzene. Thin layer chromatography showed a separation of components between fractions 26 and 29. Fractions 19–26 were combined in acetone, decolorized with activated charcoal, and crystallized from acetone–Skellysolve B, giving colorless crystals (1.260 g, 0.00487 mol, 59%): mp 124–127°; infrared spectrum identical with that of (−)-3 described above. Fractions 28–31 were combined in acetone, decolorized, and crystallized from acetone–Skellysolve B, giving colorless crystals (0.201 g, 0.000776 mol, 9%), mp 135–138°. Two recrystallizations from acetone–Skellysolve B gave (−)-4 as colorless crystals: mp 136–138°; $[\alpha]_D$ −112° (c 0.997, chloroform); infrared spectrum in Nujol is identical with that of (+)-4.

Anal. Calcd for $C_{16}H_{21}NO_2$: C, 74.10; H, 8.16; N, 5.40. Found: C, 73.90; H, 8.03; N, 5.33.

Analytical Chromatography Data. A. Paper Chromatography.—The best resolution of the bioconversion products was achieved with paper chromatography. Systems B-3[25] and FBF[2] were used with Whatman No. 2 filter paper (6 × 34 in.). The R_f values for the compounds are given in Table III. On the K-1[25] system, the 5- and 7-hydroxy compounds move with the same R_f (mobility of 1.5 with respect to the 6-hydroxy compound).

TABLE III

R_f VALUES ON PAPER CHROMATOGRAPHY

Compound	R_f in system[a] FBF	B-3
4 (6-hydroxyl)	0.29	0.076
5 (7-hydroxyl)	0.33	
3 (5-hydroxyl)	0.38	0.11
2 (substrate)	0.87	...

[a] Reference 25.

B. Thin Layer Chromatography.—The bioconversion products were partially separated on Anatech prepared silica gel GF plates which were developed with 20% (v/v) methanol in benzene. Products **4** and **3** had R_f values of 0.38 and 0.43, respectively, while the substrate had a R_f value of 0.86.

C. Vapor Phase Chromatography.—The products were not separated by vapor phase chromatography.

(4a*S*,8a*R*)-1-Benzoyl-*trans*-decahydroquinolin-5-one (6).—A solution of **3** (1.049 g, 0.00404 mol) in acetone was cooled on an ice bath and oxidized by addition of excess Jones reagent. After 15 min at room temperature, the excess oxidant was consumed by the addition of isopropyl alcohol. The mixture evaporated to dryness at room temperature. The residual solids were washed twice with methylene chloride. The methylene chloride solution was dried and allowed to evaporate, leaving an oily residue (0.986 g). The oil was dissolved in acetone. Skellysolve B was added to the solution, which then was concentrated on the steam bath until it became cloudy. Cooling in the freezer caused an oily phase to separate. Crystals slowly formed in this phase and were collected after 2 days (0.261 g), mp 81–82°. Recrystal-

(25) L. M. Reineke, *Anal. Chem.*, **28**, 1853 (1956).

lization from acetone–Skellysolve B gave **6** as colorless crystals: mp 81–83°; $\nu_{C=O}$ 1700, 1635, $\nu_{C=C}$ 1595, 1580, 1490, $\nu_{C_6H_5}$ 750, 720, 700 cm^{-1} in Nujol.

Anal. Calcd for $C_{16}H_{19}NO_2$: C, 74.68; H, 7.44; N, 5.44. Found: C, 74.72; H, 7.87; N, 5.22.

This ketone was not polymorphic with ketone **19** as shown by infrared spectra prepared in potassium bromide pellets from chloroform solution.

Additional alcohol (**3**) (3.208 g) was oxidized in a manner the same as described above. The oily product (3.069 g) of this oxidation, and the oily residue (0.675 g) remaining from separation of the crystalline ketone above were combined and chromatographed on a Florisil column (300 g) packed with Skellysolve B. The combinations of fractions given in Table IV were made on the basis of infrared spectra. Recrystallization of fractions 13–19 from acetone–Skellysolve B gave colorless crystals of **6** (0.889 g): mp 79–80°; $[\alpha]_D$ −76° (*c* 0.700, chloroform).

TABLE IV

Fractions	Eluent	Wt, g
13–19	10% acetone–Skellysolve B	1.232
20–27	10% acetone–Skellysolve B	0.219
28–30	10% acetone–Skellysolve B	0.385
31 32	25% acetone–Skellysolve B	0.604
33–36	25% acetone–Skellysolve B	1.079
	Total	3.519

Recrystallization of fractions 28–30 from acetone–Skellysolve B gave part colorless crystals together with a gummy material, which solidified (0.272 g). The crystals had mp 92–94°. The infrared spectrum of this material in Nujol is identical with the spectrum of ketone **10**, mp 97–98°. Recrystallization from acetone–Skellysolve B gave crystals, mp 104–107°.

(4a*R*,8a*R*)-1-Benzoyl-*cis*-decahydroquinolin-5-one-$d_{4a}d_6d_6$ (7).—Sodium (0.005 g) was added to a solution of **6** (0.036 g) in methyl alcohol-*d* (4 ml). The solution was kept at room temperature for 22 hr and then concentrated to half-volume on the steam bath. Aqueous acetic acid-*d* (0.5 ml, prepared from 20 drops of acetic anhydride and 25 drops of deuterium oxide) and then deuterium oxide (2 ml) were added to the solution, which was concentrated under reduced pressure. Water was added to the oily aqueous mixture, which was extracted with methylene chloride. The organic phase was dried and then concentrated to an oil, which crystallized, giving 0.017 g of crystals: mp 92–96°; *m/e* 260 (M^+).

(±)-5α-Hydroxy-*cis*-decahydroquinoline (8).—The procedure of Grob and Kiefer[13] was followed. 5-Hydroxyquinoline (Aldrich Chemical Co., 3.851 g, 0.0266 mol) was hydrogenated over prereduced platinum (0.75 g of platinum oxide) in glacial acetic acid (200 ml), giving crystals of **8** (2.604 g in two crops, 0.0168 mol, 63%), mp 150–153° (lit.[13] mp 149–150°).

(±)-1-Benzoyl-*cis*-decahydroquinolin-5α-ol (9). A. From Benzoylation of (±)-5α-hydroxy-*cis*-decahydroquinoline (8).—A mixture of **8** (1.370 g, 0.00883 mol), ice, 50% aqueous sodium hydroxide solution (5 ml), and benzoyl chloride (3.2 ml, 3.84 g, 0.0273 mol) was shaken vigorously in a separatory funnel for 15 min. The mixture was transferred to a beaker and warmed on a steam bath for 10 min and then left at room temperature overnight. The mixture was extracted with three 50-ml portions of ether. The ether was dried and partially evaporated. Crystallization began and a first crop (1.494 g) of crystals was collected, mp 136–138°. The filtrate was concentrated to a gum (0.400 g, total 1.894 g, 0.00732 mol, 83%). Two recrystallizations from acetone–Skellysolve B gave **9** as colorless crystals: mp 137–139°; ν_{OH} 3390, $\nu_{C=O}$ 1600, $\nu_{C=C}$ 1590, 1570, 1490, $\nu_{C_6H_5}$ 735, 725, 700 cm^{-1} in Nujol.

Anal. Calcd for $C_{16}H_{21}NO_2$: C, 74.10; H, 8.16; N, 5.40. Found: C, 73.94; H, 8.22; N, 5.65.

B. From Reduction of (±)-1-Benzoyl-*cis*-decahydroquinolin-5-one [(±)-10] with Sodium Borohydride.—A solution of (±)-**10** (0.140 g, 0.545 mmol) in absolute ethanol (3 ml) was added dropwise to a mixture of sodium borohydride (0.109 g, 2.88 mmol) in absolute methanol (5 ml). The mixture was kept at room temperature for 3 hr after which tlc (silica gel, 20% methanol in benzene) showed complete disappearance of ketone. Aqueous 1 *M* sulfuric acid (~3 ml) was added until release of hydrogen gas stopped. The mixture was made alkaline with 1 *M* sodium hydroxide solution. Water (10 ml) was added, and the solution was extracted with methylene chloride (five 10-ml portions). An oil, which crystallized, was obtained and recrystallized from acetone–Skellysolve B, giving 0.100 g (0.386 mmol, 70%) of crystals, mp 135–137°. The infrared spectrum in Nujol is identical with that of the alcohol **9** obtained from benzoylation of **8** above.

(±)-1-Benzoyl-*cis*-decahydroquinolin-5-one [(±)-10].—A solution of **9** (1.043 g, 0.00404 mol) in acetone (100 ml) was oxidized with excess Jones reagent. The excess oxidant was destroyed with isopropyl alcohol, the organic solvent was removed under reduced pressure, water was added, and the resulting mixture was extracted with methylene chloride. Concentration of the dry methylene chloride solution gave an oil. Crystallization occurred in acetone–Skellysolve B, giving colorless crystals (0.825 g, 0.0321 mol, 79%), mp 138–140°. Two recrystallizations from acetone–Skellysolve B gave (±)-**10** as colorless crystals: mp 138–140°; $\nu_{C=O}$ 1705, 1625, $\nu_{C=C}$ 1600, 1575, 1490, $\nu_{C_6H_5}$ 725, 705 cm^{-1} in Nujol; *m/e* 257 (M^+), 229, 214, 188, 187, 105 ($OC{\equiv}O^+$), 97, 77 (C_6H_5)$^+$.

Anal. Calcd for $C_{16}H_{19}NO_2$: C, 74.68; H, 7.44; N, 5.44. Found: C, 74.46; H, 7.38; N, 5.61.

(±)-1-Benzoyl-*cis*-decahydroquinolin-5-one-$d_{4a}d_6d_6$ [(±)-7].—Sodium (0.004 g) was added to a solution of (±)-**10** (0.030 g, 0.117 mmol) in methyl alcohol-*d* (4 ml). The solution was kept at room temperature for 20 hr and the product isolated as described for **7**. Crystallization of the product occurred from methylene chloride–Skellysolve B when the solution was kept in the refrigerator, giving 0.006 g (0.0231 mmol, 20%) of crystals: mp 139–141°; *m/e* 260 (M^+). A mixture melting point with ketone (±)-**10** was undepressed, 139–141°.

(4a*R*,8a*R*)-1-Benzoyl-*cis*-decahydroquinolin-5-one (10). A. By Isomerization of 6 with Sodium Methoxide.—A solution of **6** (1.13 g) and sodium (0.080 g) in methanol was kept at room temperature for 24 hr. Glacial acetic acid (1 ml) was added to the solution, which then was concentrated under reduced pressure. Water was added to the solution, which then was concentrated under reduced pressure. Water was added to the residue, and the mixture was extracted with three 25-ml portions of methylene chloride. The dried extract was concentrated under reduced pressure. Crystallization of the residue from acetone–Skellysolve B gave only 0.054 g of sticky crystals. The filtrate from these crystals was allowed to evaporate slowly. Crystals formed in the residual oil. They were washed with ethyl acetate and collected by filtration, giving 0.308 g of product. Recrystallization from acetone–Skellysolve B gave 0.175 g of crystals, mp 94–97°. A second recrystallization from acetone-Skellysolve B gave some rectangular crystals: mp 108–109°; $[\alpha]_D$ −77° (*c* 0.778, chloroform); $\nu_{C=O}$ 1715, 1635, 1625, $\nu_{C=C}$ 1575, 1495, $\nu_{C_6H_5}$ 800, 745, 730, 705 cm^{-1} in Nujol.

Anal. Calcd for $C_{16}H_{19}NO_2$: C, 74.68; H, 7.44; N, 5.44. Found: C, 74.49; H, 7.48; N, 5.38.

The filtrate gave chunky crystals, mp 97–98°, when cooled: $\nu_{C=O}$ 1710, 1620, $\nu_{C=C}$ 1580, 1495, $\nu_{C_6H_5}$ 792, 740, 730, 705 cm^{-1} in Nujol.

Anal. Found: C, 74.66; H, 7.38; N, 5.43.

The two crystalline materials were shown to be polymorphic crystalline forms by identical infrared spectra in KBr pellets prepared from solutions of each.

B. From (4a*S*,8a*R*)-*trans*-Decahydroquinolin-5-one (24).—A solution of benzoyl chloride (0.203 g, 0.00144 mol) in pyridine (2 ml) was added to a solution of **24** (0.210 g, 0.00137 mol) in pyridine (2 ml). The solution darkened. The solution was heated on a steam bath for 5 min and then was left at room temperature several hours. The solution was stored in a freezer overnight. A thin layer chromatogram indicated that reaction was largely complete. Crystals, assumed to be pyridine hydrochloride, were removed from the solution. The solution was concentrated under reduced pressure until the pyridine odor was very faint. Water was added to the brownish yellow oily–crystalline residue, and the resulting mixture was extracted with three 20-ml portions of methylene chloride. The extract was dried and concentrated under reduced pressure, giving a viscous oil. The oil failed to crystallize from acetone–Skellysolve B. An infrared spectrum of the oil in chloroform solution was nearly identical with that of (±)-1-benzoyl-*cis*-decahydroquinolin-5-one [(±)-**10**] but was quite different from that of (4a*S*,8a*R*)-1-benzoyl-*trans*-decahydroquinolin-5-one (**6**).

C. From Oxidation of (4a*S*,5*S*,8a*R*)-1-Benzoyl-*trans*-decahydroquinolin-5-ol (3) with Jones Reagent.—See above under preparation of **6**.

Reduction of (4a*S*,8a*R*)-1-Benzoyl-*trans*-decahydroquinolin-5-one (6) with Sodium Borohydride. (4a*S*,5*S*,8a*R*)-1-Benzoyl-*trans*-decahydroquinolin-5-ol (3) and (4a*S*,5*R*,8a*R*)-1-Benzoyl-*trans*-decahydroquinolin-5-ol (11).—A solution of **6** (0.316 g, 1.23 mmol) in absolute ethanol (5 ml) was added to a mixture of sodium borohydride (0.255 g, 6.75 mmol) and absolute ethanol (10 ml). After 3 hr at room temperature, the product was isolated and crystallized from acetone–Skellysolve B. The crystals were a mixture of needles and chunky solid, which were separated manually by difference in density. The needles (0.097 g, 0.374 mmol, 30%) had mp 113–117° and, after recrystallization from acetone–Skellysolve B, mp 125–127°. The infrared spectrum in Nujol is identical with that of **3**, isolated from the bioconversion of (±)-**2**. The chunky crystals (0.128 g, 0.494 mmol, 40%) had mp 108–114°. Two recrystallizations from acetone–Skellysolve B gave **11** as colorless crystals: mp 110–113°; $[\alpha]_D$ −123° (*c* 1.058, chloroform), ν_{OH} 3460, $\nu_{C=O}$ 1635, $\nu_{C=C}$ 1605, 1490, $\nu_{C_6H_5}$ 785, 700 cm^{-1} in Nujol.

Anal. Calcd for $C_{16}H_{21}NO_2$: C, 74.10; H, 8.16; N, 5.40. Found: C, 74.02; H, 7.81; N, 5.62.

(4a*S*,5*S*,8a*R*)-1-Benzyl-*trans*-decahydroquinolin-5-ol (12). A. From Lithium Aluminum Hydride Reduction of 3.—A solution of **3** (14.67 g, 0.566 mol) in tetrahydrofuran (200 ml) was added to a mixture of lithium aluminum hydride (5.0 g, 0.131 mol) and tetrahydrofuran (200 ml). The mixture was heated at the reflux temperature of tetrahydrofuran for 5 hr. Following the addition of water to decompose the excess hydride, the reaction was worked up, giving an oil. The oil was transferred to a simple distillation apparatus. A liquid (2.013 g, 0.0187 mol, 33%) distilled, bp 68–70° (0.6 mm), and then crystals formed in the neck of the distillation head. The liquid was found to be largely benzyl alcohol by comparison of its infrared spectrum with a known spectrum. The undistilled material remained as oily crystals. A portion (3.78 g) of the oily crystals was chromatographed on aluminum oxide (Woelm neutral, activity I, 300 g, 335-ml fractions) packed with benzene. Elution with 20% chloroform in benzene gave **12** as a faintly pink viscous gum: $[\alpha]_D$ −74° (*c* 0.505, chloroform); ν_{OH} 3350, $\nu_{Na-lkyl}$ 2780, 2740, $\nu_{C=C}$ 1600, 1580, 1490, $\nu_{C_6H_5}$ 740, 700 cm^{-1} on a smear.

Anal. Calcd for $C_{16}H_{23}NO$: C, 78.32; H, 9.45; N, 5.71. Found: C, 76.38; H, 9.41; N, 5.88.

The perchlorate salt of **12** was prepared in ethanol and was crystallized three times from ethanol–ether. Colorless crystals of the product had mp 194–196°; $[\alpha]_D$ −7°; ν_{OH} 3540, ν_{NH} 3090, $\nu_{C=C}$ 1585, 1495, $\nu_{C_6H_5}$ 740, 695 cm^{-1} in Nujol.

Anal. Calcd for $C_{16}H_{24}NO_5Cl$: C, 55.57; H, 6.99; N, 4.05. Found: C, 55.40; H, 7.08; N, 3.88.

A sample of crystals, obtained by washing the above oily crystals with acetone, was shown to be (4a*S*,5*S*,8a*R*)-*trans*-decahydroquinolin-5-ol (**23**) by its infrared spectrum, which was identical with the spectrum of the compound (**23**) prepared by hydrogenolysis of **12**.

B. From Reduction with Diborane.[26]—A solution of diborane in tetrahydrofuran (*ca.* 1 *M*, 120 ml) was added cautiously to a solution of **3** (12.103 g, 0.0467 mol) in tetrahydrofuran (150 ml). The clear solution which resulted was stirred at reflux temperature for 18 hr. Methanol (20 ml) was added, the first few drops slowly. The solution was stirred 1 hr at room temperature and then was concentrated under reduced pressure, giving an oil. Ether (75 ml) and dilute hydrochloric acid (2 *M*, 25 ml) were added to the oil. The mixture was swirled occasionally while kept at room temperature for an hour. The mixture was shaken vigorously and then the layers were separated. The ether phase was washed with water and the aqueous layers were combined. Aqueous sodium hydroxide solution (25%) was added until the solution was alkaline and an oil separated. The mixture was extracted with three 60-ml portions of ether. The ether solution was dried and concentrated under reduced pressure, giving an oil. Additional product was obtained from the acid-extracted ether layer above after it had been left standing for several days. The ether layer was extracted with dilute hydrochloric acid. The aqueous acid phase was made alkaline with sodium hydroxide solution. An oil separated and was extracted with ether. The ether was dried and concentrated under reduced pressure, giving an oil. In this way, a total of 10.43 g (0.0425 mol, 91%) of colorless oily product (**12**) was obtained.

(26) *Cf.* Z. B. Papanastassiou and R. J. Bruni, *J. Org. Chem.*, **29**, 2870 (1964); H. C. Brown and P. Heim, *J. Amer. Chem. Soc.*, **86**, 3566 (1964).

(4a*S*,5*S*,8a*R*)-1-Benzyl-*trans*-decahydroquinolin-5-ol Tosylate (13).—A solution of **12** (0.943 g, 3.85 mmol) in pyridine (8 ml) was cooled on an ice bath. *p*-Toluenesulfonyl chloride (0.734 g, 3.86 mmol) was added and the resulting deep red solution was left at room temperature for 70 hr. Water (0.1 ml) was added to the solution, which was concentrated under reduced pressure to an oil. Aqueous 1 *M* sodium hydroxide was added to the oil, and the resulting mixture was extracted with two 30-ml portions of chloride. Drying and concentrating of the solution left an oil, which crystallized. Recrystallization from methylene chloride–Skellysolve B gave, in two crops, 0.808 g (2.02 mmol, 52%) of reddish tinged crystals, mp 141–143°. Three more recrystallizations, the first preceded by decolorization with activated charcoal, from methylene chloride–Skellysolve B gave faintly pink crystalline flakes: mp 148–149°; $[\alpha]_D$ −45° (*c* 0.874, chloroform); $\nu_{N-alkyl}$ 2790, $\nu_{C=C}$ 1600, 1490, ν_{SO_2O-} 1360, 1350, 1190, 1180, $\nu_{C_6H_5}$ 745, 700 cm^{-1}; $\delta_{TMS}^{CDCl_3}$ 7.80 and 7.31 (−$C_6H_4SO_3$-*p*, doublets, J = 8.0 cps, 4 H), 7.26 (C_6H_5, singlet, 5 H), 4.00 and 3.14 (>$NCH_2C_6H_5$, doublets, J = 13.5 cps, 2 H), 2.40 ppm (O–CH_3, singlet).

Anal. Calcd for $C_{23}H_{29}NO_3S$: C, 69.15; H, 7.32; N, 3.51; S, 8.01. Found: C, 69.07; H, 7.26; N, 3.66; S, 8.24.

Fragmentation of (4a*S*,5*S*,8a*R*)-1-Benzyl-*trans*-decahydroquinolin-5-ol Tosylate (13) in 80% Ethanol in Water.—The tosylate **13** (0.060 g) did not dissolve completely in 80% ethanol in deuterium oxide (1.0 ml) at 75°. The mixture was held at this temperature for 0.5 hr, during which time the undissolved crystals slowly went into solution. No crystals formed, even with seeding with starting compound, following cooling to room temperature. After remaining at room temperature overnight, the solution was concentrated to dryness under reduced pressure. A colorless oil remained, which was dissolved in deuteriochloroform and the nmr spectrum measured: $\delta_{TMS}^{CDCl_3}$ 7.45 (−$C_6H_4SO_3$-*p*, quartet, 4 H), 7.33 (−C_6H_5, 5 H), 5.28 (*trans*–HC=CH–, ~1.7 H), 4.08 ($\gtrless \overset{+}{N}$–CH_2–C_6H_5, 2 H), 2.75 ($\gtrless \overset{+}{N}$–CH_2–C, 2 H), 2.37 (–O–CH_3, singlet), 1.87 ppm (broad band with peak at this point).

(4a*S*,8a*S*)-1-Benzoyl-*trans*-decahydroquinolin-7-one (15).—A solution of (+)-**5** (0.160 g, 0.618 mmol) in cold acetone was oxidized with an excess of Jones reagent. The reaction was worked up as described for **6**. Crystallization of the oily product from acetone–Skellysolve B gave a first crop of slightly oily, pale yellow crystals (0.095 g) and a second crop of nearly colorless crystals (0.036 g, total 0.131 g, 0.510 mmol, 82%). A sample of the best crystals was chosen for analyses and had mp 69–72°; $\nu_{C=O}$ 1705, 1615, $\nu_{C=C}$ 1575, 1495 cm^{-1} in Nujol.

Anal. Calcd for $C_{16}H_{19}NO_2$: C, 74.68; H, 7.44; N, 5.44. Found: C, 74.55; H, 7.61; N, 5.47.

(4a*S*,8a*S*)-1-Benzoyl-*trans*-decahydroquinolin-7-one-$d_6d_6d_8d_8$ (16).—A solution of **15** (0.038 g) and sodium (0.009 g) in methyl alcohol-*d* (5 ml) was left at room temperature for 21 hr. The product was isolated in the manner described for isolation of **7**. The product was obtained as a colorless, viscous oil, *m/e* 261 (M^+).

(4a*S*,7*S*,8a*S*)-1-Benzyl-*trans*-decahydroquinolin-7-ol (17).—A solution of (+)-**5** (2.128 g, 0.00822 mol) in tetrahydrofuran was reduced with a solution of diborane (*ca.* 1 *M*, 25 ml) in tetrahydrofuran in the manner described above for reduction of **3** with diborane. An oil was obtained from the ether extract of the basic solution. Crystallization from acetone–Skellysolve B gave colorless crystals (0.165 g), mp 65–80°. The filtrate was concentrated and taken up in acetone. Addition of water caused crystallization of colorless needles (0.518 g), mp 60–75°. Recrystallization from methanol–water gave colorless needles, mp 65–80°. When first dissolved in chloroform and the clear solution decanted, crystals were obtained from acetone–water having mp 65–80° (Fisher-Johns); mp 107–109° (capillary); ν_{OH} 3360, 3200, $\nu_{C=C}$ 1605, 1590, 1500, ν_{C-O} 1060 s, $\nu_{C_6H_5}$ 750, 700 cm^{-1} in Nujol.

Anal. Calcd for $C_{16}H_{23}NO \cdot H_2O$: C, 72.96; H, 9.57; N, 5.32. Found: C, 73.73; H, 9.51; N, 5.18.

A further recrystallization from acetone–water gave colorless crystals: mp 77–80° (capillary); $[\alpha]_D$ +109° (*c* 0.687, chloroform); infrared spectrum in Nujol is identical with that above.

Anal. Found: C, 72.79; H, 9.73; N, 5.17.

Additional product was obtained from the dry residue remaining after the orginal acid-extracted ether solution was allowed to evaporate to dryness. Aqueous 1 *M* sodium hydroxide was added to the residue, and the resulting mixture extracted with

ether. The oily product from the dried and concentrated ether solution as crystallized from acetone–water, giving colorless needles (0.686 g, total 1.369 g, 0.00520 mol, 63%).

When reduction of (+)-**5** (2.30 g) was carried out with lithium aluminum hydride in refluxing tetrahydrofuran, oily crystals were obtained. The crystals were washed with acetone and collected by filtration (0.237 g) and had mp 181–183°. The infrared spectrum of the crystals in Nujol is identical with that of **18** taken in Nujol. Additional crystals were obtained from hydrogenolysis of the filtrate over 5% palladium on carbon.

(4a*S*,7*S*,8a*S*)-*trans*-Decahydroquinolin-7-ol (18).—A solution of **17** (hydrate) (1.036 g, 0.00394 mol) in methanol was shaken with hydrogen over 5% palladium-on-carbon catalyst. Hydrogen consumption was rapid. The catalyst was removed by filtration. The filtrate was concentrated under reduced pressure, giving a crystalline residue. The residue was washed with acetone and collected by filtration, giving 0.487 g (0.00314 mol, 80%) of crystalline product, mp 179–182°. Two recrystallizations from acetone gave **18** as colorless crystals: mp 182–183°: $\nu_{OH,NH}$ 3310, 3200 cm^{-1} in Nujol; $\nu_{NH,OH}$ 3600, 3170, ν_{C-O} 1047 s, 1031 m, 1010 w cm^{-1} in chloroform and identical with that of authentic (±)-7α-hydroxy-*trans*-decahydroquinoline[16] in chloroform.

Anal. Calcd for $C_9H_{17}NO$: C, 69.63; H, 11.04; N, 9.02. Found: C, 69.37; H, 11.10; N, 9.06.

(4a*S*,8a*S*)-1-Benzoyl-*trans*-decahydroquinolin-6-one (19).—A slight excess of Jones reagent was used to oxidize (+)-**4** (1.911 g, 7.38 mmol) in an acetone solution (30 ml). After the usual work-up, oily crystals were obtained. Crystallization from acetone Skellysolve B gave 1.59 g (6.21 mmol, 84%) of ketone as rectangular crystals, mp 129–131°. Two recrystallizations from acetone–Skellysolve B gave **19** as feathery crystals: mp 129–131°; $[\alpha]_D$ +181° (*c* 0.587, chloroform); $\nu_{C=O}$ 1705, 1610, $\nu_{C=C}$ 1595, 1575, 1515, 1490, $\nu_{C_6H_5}$ 790, 725, 700 cm^{-1} in Nujol.

Anal. Calcd for $C_{16}H_{19}NO_2$: C, 74.68; H, 7.44; N, 5.44. Found: C, 74.49; H, 7.46; N, 5.51.

A Polymorphic Form of 19.—Dioxane (5 ml) and an ethereal solution of hydrogen chloride (1 ml) were added to **19** (0.227 g) to determine if the compound might isomerize. The crystals went into solution and then other crystals formed. These were collected after 6 hr (0.147 g): mp 120–123°; $\nu_{C=O}$ 1705, 1605, $\nu_{C=C}$ 1590, 1570, 1530, 1490, $\nu_{C_6H_5}$ 795, 722, 703 cm^{-1} in Nujol. Recrystallization from acetone–Skellysolve B gave crystals having an infrared spectrum identical with that of the original ketone.

Reduction (4a*S*,8a*S*)-1-Benzoyl-*trans*-decahydroquinolin-6-one (19) with Sodium Borohydride. (4a*S*,6*S*,8a*S*)-1-Benzoyl-*trans*-decahydroquinolin-6-ol [(+)-4].—A solution of **19** (0.518 g, 2.01 mmol) in absolute ethanol (5 ml) was added to a mixture of sodium borohydride (0.4 g) and absolute ethanol (15 ml). The mixture was kept at room temperature for 22 hr. The mixture was first treated with 1 *M* sulfuric acid (3 ml), then was made alkaline with 1 *M* sodium hydroxide solution. Water (10 ml) was added and the solution was extracted with five 15-ml portions of methylene chloride. The organic extract was dried, then concentrated to oily crystals. Recrystallization from acetone–Skellysolve B gave a first crop (0.243 g) of hexagonal needles, mp 134–137°. Second and third crops (0.117 and 0.028 g, total 0.388 g, 1.50 mmol, 74%) of crystals, mp 122–130°, were obtained. All crops had infrared spectra in Nujol identical with that of (+)-**4**, obtained in the bioconversion of (+)-**2**.

(4a*S*,8a*S*)-1-Benzyl-*trans*-decahydroquinolin-6-one-$d_5d_6d_7d_7$ (20).—Sodium (0.004 g) was added to a solution of **19** (0.031 g, 0.121 mmol) in methyl alcohol-*d* (4 ml). The product was isolated exactly as described for the preparation of **7**. Crystals (0.022 g, 0.0843 mmol, 70%), mp 117–120°, were obtained, *m/e* 161 (M^+).

(4a*S*,6*S*,8a*S*)-1-Benzyl-*trans*-decahydroquinolin-6-ol (21).—A solution of diborane (*ca.* 1 *M*, 60 ml) in tetrahydrofuran was added cautiously to a stirred solution of (+)-**4** (6.043 g, 0.0233 mol) in tetrahydrofuran (75 ml). The reaction was run at reflux temperature for 18 hr in a flask fitted with a condenser and calcium chloride drying tube. The reaction was worked up in the same manner as was used for the diborane reduction of **3**. Crystals (2.216 g), mp 57–70°, were deposited slowly from the ether extract of the basic products. The ether was decanted from the crystals and was dried. Concentration under reduced pressure gave an oil, which crystallized from acetone–water as shiny, colorless plates (2.275 g), mp 55–65°. Several recrystallizations from acetone–water did not improve the melting point when taken on a Fisher-Johns block. In a sealed, evacuated capillary, the compound had mp 68–71°; $[\alpha]_D$ +100° (*c* 0.695, chloroform); ν_{OH} 3340, 3160, $\nu_{C=C}$ 1605, 1585, 1500, ν_{C-O} 1165, 1150s, 1125, 1115s, $\nu_{C_6H_5}$ 750, 700 cm^{-1} in Nujol.

Anal. Calcd for $C_{15}H_{23}NO \cdot H_2O$: C, 72.96; H, 9.57; N, 5.32. Found: C, 74.00; H, 9.42; N, 5.21.

(4a*S*,6*S*,8a*S*)-*trans*-Decahydroquinolin-6-ol (22). A. From Lithium Aluminum Hydride Reduction of (+)-4.—A solution of (+)-**4** (2.213 g, 8.53 mmol) in tetrahydrofuran (75 ml) was added slowly to a stirred mixture of lithium aluminum hydride (2.1 g) tetrahydrofuran (400 ml). The resulting mixture was heated at reflux temperature for 5 hr and was kept at room temperature for 16 hr. Following typical work-up, oily crystals were obtained from the organic phase. The crystals (0.527 g, mp 216–217°) were washed with acetone and collected by filtration. Recrystallization from methanol–acetone gave colorless crystals: mp 217–218° (sublime); $[\alpha]_D$ −10° (*c* 0.707, 95% ethanol); the infrared spectrum in Nujol is identical with that of (+)-**22**.

Anal. Calcd for $C_9H_{17}NO$: C, 69.63; H, 11.04; N, 9.02. Found: C, 69.29; H, 10.97; N, 9.13.

The oily residue obtained from the filtrate was assumed to be a mixture of the product (**22**) and the corresponding benzylamine (**21**). Accordingly, a solution of the residue in methanol was shaken with hydrogen over a 5% palladium-on-carbon catalyst. Additional crystalline product (**22**) (0.377 g, total 0.904 g, 5.83 mmol, 68%), mp 216–217°, was obtained from the hydrogenolysis.

B. From Hydrogenolysis of 21.—Hydrogenolysis of a solution of benzylamine (**21**) hydrate (3.00 g, 0.0114 mol) over 5% palladium on carbon (1.5 g) was rapid and complete after 10 min. Removal of the catalyst by filtration and concentration of the filtrate gave colorless crystals. Recrystallization from methanol–acetone gave two crops (total 1.446 g, 0.00932 mol, 81%) of colorless crystals, mp 213–215°. The infrared spectrum in Nujol is identical with that of the above product.

(±)-*trans*-Decahydroquinolin-6-ol [(±)-22].—A solution of **4** (0.546 g, 0.00211 mol) in tetrahydrofuran (100 ml) was added to a mixture of lithium aluminum hydride (0.60 g) and tetrahydrofuran (100 ml). The mixture was heated at the reflux temperature for 2.5 hr and was stirred at room temperature 16 hr. Ethyl acetate and water were added to consume the excess hydride. The solids were removed by filtration through Celite and washed with tetrahydrofuran. The organic layer was dried and concentrated under reduced pressure, giving oily crystals. Crystallization from acetone gave crystals (0.1 g), mp 180–186°. The filtrate was concentrated to an oil, which had an infrared spectrum indicating that the benzyl group was still present. The oil was dissolved in methanol and shaken with hydrogen over palladium on carbon (0.2 g) in a Parr apparatus. After removal of the catalyst and concentration of the solution, additional crystals (0.075 g, total 0.175 g, 0.00113 mol, 53%) were obtained, which had an infrared spectrum identical with the spectrum of the first crystals obtained. Three recrystallizations from acetone, the last preceded by decolorization with charcoal, gave (+)-**22** as colorless crystals: mp 189–190°; $\nu_{NH,OH}$ 3260, 3100, 2800 cm^{-1} in Nujol.

Anal. Calcd for $C_9H_{17}NO$: C, 69.63; H, 11.04; N, 9.02. Found: C, 69.46; H, 11.21; N, 9.43.

(4a*S*,5*S*,8a*R*)-*trans*-Decahydroquinolin-5-ol (23).—A solution of **12** (2.179 g, 0.00888 mol) in methanol was shaken with hydrogen over 5% palladium on carbon (0.62 g) for 30 min on a Parr apparatus. The catalyst was removed by filtration and washed with methanol. The filtrate was concentrated under reduced pressure, giving an oil which crystallized. Recrystallization from methylene chloride–Skellysolve B gave 0.824 g (0.00532 mol, 60%) of crystals, mp 141–144° (sublimes). Two recrystallizations from methylene chloride, the second preceded by decolorization with activated charcoal gave crystals, mp 148°. The crystalline material was sublimed and recrystallized from methylene chloride–Skellysolve B, giving **23** as colorless crystals: mp 147–148°; $[\alpha]_D$ +51° (*c* 0.964, chloroform); $\nu_{OH,NH}$ 3250, 3100, $\nu_{N\text{-alkyl, bonded OH,NH}}$ 2790, 2750, 2700 cm^{-1} in Nujol.

Anal. Calcd for $C_9H_{17}NO$: C, 69.63; H, 11.04; N, 9.02. Found: C, 69.38; H, 11.09; N, 9.14.

(4a*S*,8a*R*)-*trans*-Decahydroquinolin-5-one (24).—Jones reagent (2.5 ml) was added rapidly with stirring to a solution of **23** (0.984 g, 0.00633 mol) in warm (40°) acetone (100 ml). The resulting mixture was left at room temperature for 1 hr and then was treated with isopropyl alcohol (0.6 ml). Aqueous 1 *M* sodium hydroxide solution (50 ml) was added, and the mixture

was concentrated under reduced pressure until the acetone was removed. A little additional water was added to the remaining mixture, which was then extracted with four 35-ml portions of methylene chloride. The extract was dried and concentrated under reduced pressure. The oily product (0.721 g) crystallized. An infrared spectrum showed that some alcohol remained. The product was dissolved in acetone (50 ml) and oxidized at 40° with Jones reagent (1 ml). The isolation procedure used following the first oxidation was repeated, giving a crystalline product (0.660 g). The product in acetone solution was decolorized with activated charcoal. The acetone was replaced by Skellysolve B, and an initial deposit of crystals (**23**) in this solvent was removed by filtration. Crystallization of **24** from the filtrate at room temperature gave a first crop of 0.204 g of crystals, mp 75–77°. A second crop of colorless needles (0.085 g, total 0.289 g, 0.00189 mol, 30%), mp 76–78°, was collected: RD (*c* 0.535, CH_3OH) $[\phi]_{400}$ +572°, $[\phi]_{320}$ +1087°, $[\phi]_{314}$ +1573°, $[\phi]_{306}$ +1745°, $[\phi]_{294}$ 0°, $[\phi]_{266}$ −2975°, $[\phi]_{257}$ −2850, $[\phi]_{239}$ −3160°; ν_{NH} 3230, $\nu_{C=O}$ 1705 cm^{-1} in Nujol.

Anal. Calcd for $C_9H_{15}NO$: C, 70.55; H, 9.87; N, 9.14. Found: C, 70.27; H, 9.84; N, 9.11.

(**4a***S*,**8a***S*)-*trans*-**Decahydroquinolin-6-one (25).**—A solution of **22** (0.862 g, 0.00555 mol) in hot acetone (200 ml) was oxidized with Jones reagent (2.5 ml). The reaction was worked up following the procedure described above for **24**. A solid crystalline product was obtained, which was separated into starting material (0.124 g, identified by an infrared spectrum) and product by the insolubility of the starting material in ether. The product (0.214 g) crystallized from Skellysolve B, giving colorless needles: mp 82–83°; RD (*c* 0.584, CH_3OH), $[\phi]_{589}$ +63°, $[\phi]_{400}$ +281°, $[\phi]_{350}$ +592°, $[\phi]_{320}$ +1610°, $[\phi]_{316}$ +1972°, $[\phi]_{307}$ +2323°, $[\phi]_{290}$ 0°, $[\phi]_{263}$ −2320°, $[\phi]_{231}$ −894°; ν_{NH} 3230, 3220, $\nu_{C=O}$ 1710 cm^{-1} in Nujol.

Anal. Calcd for $C_9H_{15}NO$: C, 70.55; H, 9.87; N, 9.14. Found: C, 69.76, 69.86; H, 9.69, 9.77; N, 8.93.

(**4a***S*,**8a***S*)-*trans*-**Decahydroquinolin-7-one (26).**—A cold solution of **18** (0.487 g, 0.00314 mol) in 7 *M* sulfuric acid (2 ml) was oxidized with the dropwise addition of a solution of chromium trioxide (0.222 g) in 7 *M* sulfuric acid (3 ml) (method of Grob and Wilkens[16]). After 15 min at room temperature, the solution was made basic by the slow addition of aqueous 20% sodium hydroxide while keeping the solution cold. A precipitate formed. A saturated potassium carbonate solution (1 ml) was added. The resulting mixture was stirred with chloroform (15 ml) for 1 hr and then was extracted with four 20-ml portions of additional chloroform. The chloroform solution was dried and concentrated under reduced pressure. A crystalline product was obtained, which was shown to contain some starting alcohol by an infrared spectrum. The solid was then oxidized as described for the preparation of **24** with Jones reagent. Following the same work-up procedure, a crystalline product was obtained. Crystallization from Skellysolve B gave two crops (0.288 g, 0.00188 mol, 60%) of colorless crystals, mp 120–122°. Two recrystallizations from Skellysolve B gave colorless needles: mp 121–123°; RD (*c* 0.644, CH_3OH), $[\phi]_{370}$ +24.6°; $[\phi]_{320}$ +59.6, $[\phi]_{310}$ +95°, $[\phi]_{301}$ +114, $[\phi]_{290}$ +102°, $[\phi]_{282}$ +86°, $[\phi]_{267}$ + 102°, $[\phi]_{250}$ +176°; ν_{NH} 3220, 3210; $\nu_{C=O}$ 1710 cm^{-1} in Nujol; *m/e* 153 (M^+).

Anal. Calcd for $C_9H_{15}NO$: C, 70.55; H, 9.87; N, 9.14. Found: C, 70.38; H, 9.73; N, 9.88.

Registry No.—(+)-**2**, 16878-36-7; (−)-**2**, 5681-50-5; (−)-**3**, 16878-16-3; (±)-**4**, 16878-38-9; (+)-**4**, 16878-35-6; (−)-**4**, 16878-34-5; (+)-**5**, 16878-39-0; **6**, 16878-17-4; (±)-**9**, 16878-18-5; **10**, 16878-19-6; (±)-**10**, 16959-97-0; **11**, 16915-92-7; **12**, 16878-20-9; perchlorate salt of **12**, 16878-21-0; **13**, 16878-22-1; **15**, 16878-23-2; **17**, 16878-24-3; **18**, 16878-25-4; **19**, 16878-26-5; **21**, 16878-27-6; **22**, 16878-28-7; (±)-**22**, 16878-29-8; **23**, 16878-30-1; **24**, 16878-31-2; **25**, 16878-32-3; **26**, 16878-33-4.

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Stereochemistry of Microbiological Hydroxylation

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Several observations concerning the stereochemistry of a number of rigid molecules, that have been hydroxylated by *Sporotrichum sulfurescens*, in relationship to possible enzyme–substrate interactions are discussed. Rigid molecules containing the 1-benzoylpiperidine ring are hydroxylated at positions outside of the piperidine ring, supporting the idea that a 5.5-Å distance between the electron-rich center and point of hydroxylation is preferred in substrates containing the amide functional group. The hydroxyl group introduced into the substrate molecule by the microorganism has been found to be oriented *trans* with respect to the amide functional group. A spatial orientation for the methylene group which is hydroxylated has been defined on the basis of a coordinate system. Mapping of the enzyme contours may then be carried out indirectly by observing the volume of space occupied by rigid molecules when they are placed into this arbitrary orientation. Preliminary results based on optically active products obtained from hydroxylation of 1-benzoyl-*trans*-decahydroquinoline indicate a preference for placing the bulk of the molecules in the upper right (UR) rear octant of the coordinate system. The dihydroxylation of certain 1-adamantanamine derivatives is observed to result from increased lipophilic character in the amide group. Finally it is suggested that the oxidation state (alcohol or ketone) of the oxygenation products depends upon the conformational mobility of the molecule in question.

A recent report from these laboratories proposed a hypothetical enzyme–substrate model to account for the preferential hydroxylation at certain sites observed during the oxygenation of macrocyclic alcohols by the microorganism, *Sporotrichum sulfurescens*.[1] This model suggests that an electron-rich center of the cyclic substrate molecule becomes attached to the hydroxylating enzyme and that hydroxylation then occurs at a carbon atom approximately 5.5 Å distant from the attachment site.[1] In the case of the macrocyclic alcohols, the hydroxyl oxygen serves as the electron-rich center. Substrates containing other electron-rich groups are also oxygenated by *S. sulfurescens* and the amide functional group has been particularly useful in this respect.[2] Among the types of amide-containing molecules, all of a cyclic nature, which we have studied are amides of azacycloalkanes,[3] azabicycloalkanes,[4]

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