Experimental Section

Formation and Coupling of Carbanions.—The anions were prepared by addition of an ethereal solution of the ester or nitrile to 1 equiv of potassium amide in 120–200 ml of liquid ammonia and stirring for 20 min. An ethereal solution of the halide (0.5 molar equiv) was added, the ammonia was allowed to evaporate, and the residue was stirred with water and ether or chloroform and filtered. Individual work-ups follow.

2,3-Diphenylsuccinonitrile (II).—The solid on the funnel was recrystallized from acetic acid to give tan needles of the *meso* nitrile, mp 238° (lit.[6] mp 239–240°). The chloroform solution was diluted with ethanol and chilled, giving the *dl* nitrile, mp 164° (lit.[6] mp 164°). The aqueous solution gave a positive Prussian Blue test for cyanide, which was quantitatively determined.[7]

Tetraphenylsuccinonitrile (V).—The chloroform solution was evaporated, and the residue was recrystallized from chloroform–ethanol to give tetraphenylsuccinonitrile, mp 215°, undepressed by an authentic sample.[8]

Diethyl 2,3-Diphenylsuccinate (VII).—The solid on the funnel was recrystallized from aqueous methanol to give the *meso* ester, mp 140–141° (lit.[9] mp 140–141°). The ethereal solution was evaporated, and the residue was recrystallized from aqueous methanol to give the *dl* ester, mp 79–80° (lit.[9] mp 82–82.5°).

Tetraphenylsuccinic Acid.—Evaporation of the ethereal solution gave an oily solid which several recrystallizations failed to purify. The material was hydrolyzed overnight in refluxing ethanol with 5.7 g (0.1 mol) of potassium hydroxide. The ethanol was evaporated, and the residue was stirred with water and methylene chloride. The aqueous solution was acidified, and the tetraphenylsuccinic acid was recrystallized from methylene chloride–ethanol to give white crystals, mp 271° dec (lit.[10] mp 260–262°).

Reaction of I with Ethylene Bromide.—Evaporation of the ethereal solution gave 21.5 g of a yellow-brown oil, shown by nmr to be a mixture of phenylacetonitrile and 1-phenylcyclopropanecarbonitrile. The mixture was not separated by distillation through a 6 in. helix-packed column, but was satisfactorily separated by gas chromatography (1-m column, 20% methyl silicone SE-30). The nmr spectrum of a sample of pure III, collected from the gas chromatograph, was identical with the published spectrum.[11] The relative yields of the two nitriles (Table I) were estimated from the ratio of the peak areas.

Registry No.—*meso*-II, 15146-07-3; *dl*-II, 19657-49-9; III, 935-44-4; V, 3122-21-2; *meso*-VII, 13638-89-6; *dl*-VII, 24097-93-6; IX, 24097-49-2.

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The *A* Value of Hydroxyl Determined by the Nuclear Magnetic Resonance Peak Area Method at −83°

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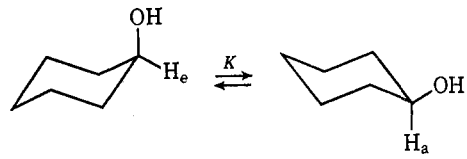
A myriad of reports have appeared concerning the measurement of the *A* value of hydroxyl (eq 1 and 2)



(1)

$$A \text{ value} = -\Delta G^\circ = (RT \ln K)/1000 \quad (2)$$

employing a large variety of techniques.[2–4] The observed *A* values are solvent dependent but deviate seriously from one technique to another in the same solvent.[2] This paper concerns the measurement of the *A* value of hydroxyl in cyclohexanol-2,2,6,6-d_4 at −83° utilizing variable-temperature nuclear magnetic resonance (nmr) spectroscopy, a technique of proven accuracy.[3]

Examination of the nmr spectrum (60 MHz) of cyclohexanol-2,2,6,6-d_4 at −83° in a number of solvents revealed two resonances corresponding to the equatorial HCO proton (H_e, eq 1) at approximately δ 3.85 and to the axial HCO proton (H_a, eq 1) at approximately δ 3.28 (Figure 1). These peak assignments are consistent with those in model compounds[5] and were confirmed by deuteration of the hydroxyl group and investigation of the HCO resonance under conditions of slow, intermediate, and fast rates of exchange on the nmr time scale.

Integration by planimeter and electronic integrator of the peak areas of axial and equatorial HCO resonances at −83° gave the equilibrium constant (*K*, eq 1) of interest and the corresponding *A* values in a variety of solvents (Table I). Since the measured equilibrium constants (*K*, eq 1) are relatively large by nmr standards, a correspondingly large radiofrequency power level was necessary to obtain reasonable reproducibility in peak areas. This introduces the possibility of differential saturation effects on the two H–C–O resonances, but these effects are included in the error limit set on *K* (eq 1, Table I).

TABLE I
A VALUE OF HYDROXYL AS A FUNCTION OF CONCENTRATION AND SOLVENT AT −83°

Group	Solvent	Concn, mol/l.	*K*	*A* value, kcal/mol
-OH	CS_2	3.0	17.0 ± 1.0	1.08 ± 0.06
		2.0	12.9 ± 0.7	0.97 ± 0.05
		1.0	11.6 ± 0.8	0.93 ± 0.05
		0.5	11.3 ± 0.8	0.92 ± 0.05
-OD	CS_2	2.0	11.6 ± 0.8	0.93 ± 0.05
-OH	Toluene	2.0	11.3 ± 0.8	0.92 ± 0.05
	Toluene	1.0	11.0 ± 0.8	0.91 ± 0.05
-OH	CD_3COCD_3	1.0	12.9 ± 0.9	0.97 ± 0.05
-OH	50% CS_2–	2.0	11.6 ± 0.6	0.93 ± 0.05
	50% α-picoline	1.0	11.5 ± 0.7	0.93 ± 0.05
	(by wt)	0.5	11.5 ± 1.8	0.92 ± 0.06
-OD	CD_3OD	1.0	16.1 ± 1.0	1.05 ± 0.06
	CD_3OD	2.0	15.8 ± 1.0	1.04 ± 0.06

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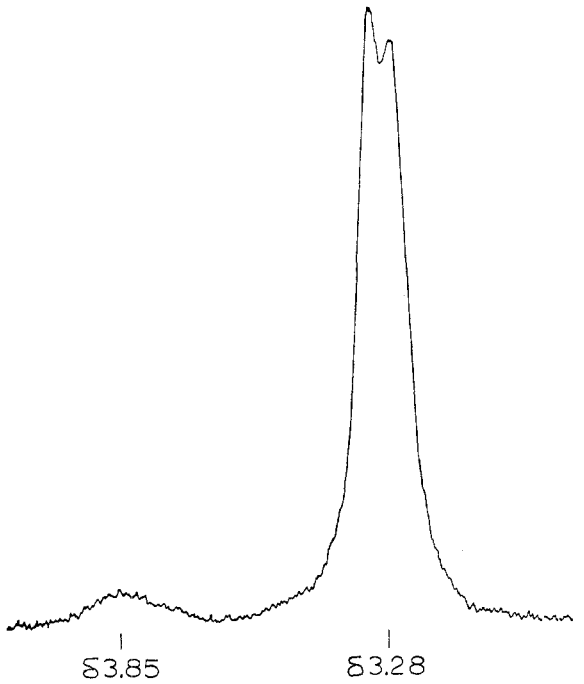

Figure 1.—Axial (δ 3.28) and equatorial (δ 3.85) HCO resonances of cyclohexanol-2,2,6,6-d_4 (3 M in CS_2) at −83°.

Perusal of Table I indicates no dramatic solvent effects (in the solvents used) although some variation is noted. The A value of hydroxyl is larger in the hydroxylic solvent CD_3OD, as expected. Some error is introduced into the A value determined in CD_3OD because of a slight overlap of the CHD_2OD impurity resonance with the axial H–C–O resonance of cyclohexanol-2,2,6,6-d_4.

These data provide an opportunity for a meaningful comparison albeit at a low temperature of the A value of hydroxyl with other oxygen-containing functionalities (Table II). Although the effective group

TABLE II

A VALUES OF VARIOUS OXYGEN-CONTAINING FUNCTIONALITIES

Group	A value, kcal/mol[a]	Group	A value, kcal/mol[a]
–OTs	0.52	–OC(=O)H	0.59
$-OCD_3$	0.55	–OAc	0.71
$-OSO_2CH_3$	0.56	–OH	0.97[b]

[a] All concentrations approximately 2½ M. Solvent is CS_2 except for OTs and OSO_2CH_3 in which case it is approximately 50:50 by volume CS_2–$CDCl_3$; see ref 3. [b] This work.

radius of hydroxyl is almost certainly smaller than the other functionalities, it has a significantly higher A value. The effect of intermolecular association is evident. It is also clear from Table II that the A values of functionalities with oxygen bonded to the cyclohexane ring are not all of the same magnitude.

Experimental Section

Nmr spectra were obtained using a Varian HR-60A spectrometer equipped with a custom-built variable-temperature probe. Spectral calibrations were performed using the audio-modulation technique. Temperature measurements were performed using a calibrated copper-constantan thermocouple.

Cyclohexanol-2,2,6,6-d_4 was prepared by the lithium aluminum hydride reduction of cyclohexanone-2,2,6,6-d_4.[6]

Registry No.—Cyclohexanol-2,2,6,6-d_4, 21273-03-0.

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Biosynthetic Studies with Carbon 13. Piericidin A

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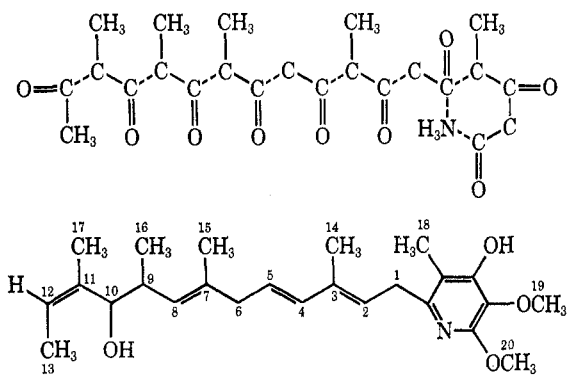
The antibiotic piericidin A is a naturally occurring insecticide which is produced by *Streptomyces mobaraensis*.[1] Its structural and stereochemical formulation (I) is due to the work of Takahashi and coworkers.[2] Biosynthetic studies conducted with carbon 14 labeled precursors indicated that the carbon chain of Piericidin A is formally derived by condensation of five propionate and four acetate units, presumably *via* an acetate starter and the methylmalonyl pathway.[3] A useful



I

procedure for biosynthetic studies of microbial metabolites is the nondegradative ^{13}C proton satellite method.[4] We wish to report that the production of piericidin A in the presence of ^{13}C-methyl labeled propionate ($^{13}CH_3CH_2CO_2Na$) affords direct information on the biological origin of the methyl groups in the antibiotic. This information can be obtained by the ^{14}C method; however, limitations on chemical degradative methods preclude identification of specific labeled carbon atoms.

Streptomyces mobaraensis fermentations in the pre-

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