Therefore, it is rather unlikely that preferred orientation can explain the observed stability differences.

Another possibility is that the CdS layer, which is formed on the electrode in S_x^{2-} solution, will always have the hexagonal structure, which would cause a larger lattice mismatch with an underlying cubic phase than with a hexagonal one. Such a lattice mismatch would influence the "rate matching"[6,21] between hole flux and electrochemical reaction, and by it the ability of S^{2-} oxidation to prevent photoelectrode self-oxidation. However, preliminary experiments on cubic $(CdTe)_{0.25}(ZnSe)_{0.75}$, which show this phase to behave similar to hexagonal Cd(Se,Te) phases in terms of output stability, can be construed as evidence against this possibility.

The most likely explanation may be found in a difference in bond strengths between hexagonal and cubic Cd(Se,Te) of the same chemical composition. The lower band gap of the cubic form, as compared to the hexagonal one (with the same stoichiometry), can be associated with a weaker interaction between the Cd and the chalcogen, something that is expressed in the effective ionic charges on Cd. These charges are much smaller in cubic CdTe than in hexagonal CdS and CdSe.[22] Rough calculations on hexagonal ZnS, by using the method of ref 21, yield an effective ionic charge of ~0.4[23] while that for cubic ZnS is ~0.3[22] (ZnS was chosen because its piezoelectric coefficients are well-known for both phases). Extrapolating to Cd(Se,Te), we expect stronger bonding (higher heat of formation per bond according to Pauling's ionicity) in the hexagonal phase than in the cubic one. Since the stability of Cd chalcogenides in polysulfide solutions depends on the relative probabilities of self-oxidation (which requires breaking a Cd-chalcogen bond) and interfacial charge transfer, a small increase in bond strength can improve the semiconductor stability profoundly.[24]

This explanation of the observed differences in output stability implies that the decomposition potentials[25,26] for the two phases are different. In view of the results from ref 4, as well as our own, a further dependence of these potentials on the orientation of the exposed crystal face is likely as well. Experiments to clarify these points further are now in progress in this laboratory.[27]

(21) Cahen, D.; Manassen, J.; Hodes, G. *Sol. Energy Mater.* **1979**, *1*, 343.
(22) Berlincourt, D.; Jaffe, H.; Shiozawa, L. R. *Phys. Rev.* **1963**, *129*, 1009.
(23) Piezoelectric data from "Landolt-Bornstein Tables", Springer-Verlag: West Berlin, Vol. III/3, 1979.
(24) The difference in effective ionic charge may affect the electrochemical reaction rate as well. However, since the electrode surface is converted rapidly into CdS, especially once current starts flowing, such an effect is unlikely, unless the nature of the newly formed CdS depends on that of the underlying phase.
(25) Gerischer, H. *J. Electroanal. Chem.* **1977**, *82*, 133.
(26) Bard, A. J.; Wrighton, M. S. *J. Electrochem. Soc.* **1977**, *124*, 1706.
(27) Partial support by Ormat Turbines Ltd. and the U.S. Israel Binational Science Foundation, Jerusalem, Israel, is gratefully acknowledged.

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Biosynthesis of Antibiotics of the Virginiamycin Family. 1. Biosynthesis of Virginiamycin M_1: Determination of the Labeling Pattern by the Use of Stable Isotope Techniques

Sir:

Virginiamycin M_1 (**1**) was first isolated from *Streptomyces virginiae*,[1] but the same compound has been isolated from a number of different microorganisms and has also been named as ostreogrycin A, mikamycin A, staphylomycin M_1, pristinamycin IIA, streptogramin A, and PA 114 A1.[2,3] Its structure was originally determined by a combination of chemical and instrumental techniques[4,5] and has been confirmed by X-ray crystallography.[6]

(1) Somer, P. de; Dijck, P. J. van *Antibiot. Chemother. (Washington, D.C.)* **1955**, *5*, 632-639.

Table I. Incorporation of ^3H- and ^{14}C-Labeled Presursors into Virginiamycin M_1

precursor added	% incorporation
sodium [2-^{14}C] acetate	5.0
L-[methyl-^{3}H] methionine	4.0
DL-[3-^{14}C] serine	1.4
L-[3,4-$^3H_2$] proline	5.0
D-[U-^{14}C] glucose	0.6
[2-^{14}C] glycine	0.4, 0.8[a]
L-[U-^{14}C] alanine	*b*

[a] Data from ref 13. [b] No measurable incorporation (ref 13).

The antibiotic is of interest from a biosynthetic viewpoint because it contains the unusual dehydroproline and oxazole ring systems. Although the origin of dehydroamino acids has been discussed previously,[7,8] the only experimental evidence bearing on the formation of the oxazole ring is found in work on the biosynthesis of the alkaloid annuloline[9] and in recent work on the biosynthesis of berninamycin, where it was postulated that ^{14}C-labeled serine and threonine were both incorporated into the oxazole units of this antibiotic.[10] In this communication, we report results which establish the overall biosynthetic origin of the major portion of virginiamycin M_1, including the oxazole ring.

Virginiamycin M_1 was produced in baffled 250-mL Erlenmeyer flasks containing 30 mL of a complex nutrient broth.[11] These were inoculated with a culture of *S. virginiae* strain PDT-30 and shaken at 21 °C. After 1 day, labeled precursors were added, and the broths were worked up 1 day later. Virginiamycin M_1 was isolated by extraction of the broth with hexane followed by ethyl acetate, and purification of the crude ethyl acetate extract by high-performance liquid chromatography.[12] Incorporation of radioactive precursors was determined by counting the chromatographically homogeneous product; this procedure was checked in one instance by addition of authentic antibiotic and recrystallization to constant specific activity.

Table I lists the ^3H- and ^{14}C-labeled precursors fed and the percent incorporation of each. These results broadly corroborate those reported earlier for incorporation of various ^{14}C-labeled substrates into virginiamycin.[13]

Feeding experiments with [2-^{13}C]acetate, [1,2-$^{13}C_2$]acetate, L-[methyl-^{13}C]methionine, and DL-[3-^{13}C]serine gave ^{13}C-labeled antibiotic in yields of 10-20 mg/L, and materials thus obtained were analyzed by ^{13}C NMR spectroscopy. The natural-abundance proton-noise-decoupled ^{13}C NMR spectrum of virginiamycin M_1 shows signals for 28 carbon atoms. Assignment of the relevant signals was made by using characteristic chemical shifts, multiplicities, single-frequency proton decoupling, and analysis of one-bond carbon-carbon couplings in pairs of carbon atoms; our

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(3) Cocito, C. *Microbiol. Rev.* **1979** 145-198.
(4) Delpierre, G. R.; Eastwood, F. W.; Gream, G. E.; Kingston, D. G. I.; Todd, A. R.; Williams, D. H. *J. Chem. Soc. C* **1966**, 1653-1669.
(5) Kingston, D. G. I.; Todd, A. R.; Williams, D. H. *J. Chem. Soc. C* **1966**, 1669-1676.
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(7) Bycroft, B. W. *Nature (London)* **1969**, *224*, 595-597.
(8) Schmidt, V.; Häusler, J.; Öhler, E.; Poisel, H. *Fortschr. Chem. Org. Naturst.* **1979**, *37*, 251-327.
(9) O'Donovan, D. G.; Horan, H. *J. Chem. Soc. C* **1971**, 331-334.
(10) (a) Pearce, C. J.; Rinehart, K. L., Jr. *J. Am. Chem. Soc.* **1979**, *101*, 5069-5070. (b) Rinehart, K. L., Jr.; Weller, D. D.; Pearce, C. J. *J. Nat. Prod.* **1980**, *43*, 1-20.
(11) The broth, designated STA-14, contained the following components (amount expressed as g/L): corn steep solids (20), peanut oil cake (10), yeast autolyzate (5), glucose (5), glycerol (25), linseed oil (10), calcium carbonate (5).
(12) Kingston, D. G. I. *J. Nat. Prod.* **1979**, *42*, 237-260.
(13) Roberfroid, M.; Dumont, P. *Ind. Chim. Belge* **1967**, *32*, 307-309.

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Table II. ^{13}C NMR Spectral Data for Virginiamycin M_1, Including Enrichments from Labeled Precursors

carbon no.	δ_C[a]	multiplicity[b]	I labeled/I unlabeled[c] [2-^{13}C] ac	[3-^{13}C] ser	[Me-^{13}C] met	$^1J_{C-C}$, Hz[d]
1	30.1	d	1.4	0.9	0.9	
1a	18.9	q	1.2	**2.4**	1.1	
1b	19.6	q	1.1	**2.2**	1.2	
2	81.5	d	1.0	0.7	1.1	
3	37.6	d	**4.6**	**1.8**	1.1	42.7
3a	12.2	q	1.0	**3.8**	**3.3**	
4	143.1	d	0.8	0.6	1.1	42.0
5	125.3	d	**4.2**	**1.7**	0.8	64.9
6	167.6	s	0.8	0.4	0.8	65.2
7	40.5	t	1.1	0.9	1.3	
8	126.1	d	1.0	0.8	1.3	
9	133.7	d	**3.7**	**1.7**	1.1	*e*
10	134.7	s	0.7	0.4	0.5	*e*
10a	12.7	q	**4.1**	**3.0**	1.4	
11	131.0	d	**4.1**	**1.8**	1.0	48.0
12	66.0	d	0.9	0.6	1.0	48.0
13	47.7	t	**4.1**	**2.4**	1.2	40.0
14	200.7	s	0.7	0.6	0.6	40.5
15	45.7	t	**3.6**	**2.1**	1.1	60.7
16	156.2	s	0.7	0.5	0.6	60.3
17	136.1 or 137.2	s	1.2 or 1.3	0.7 or 0.8	0.6 or 0.5	
17a	145.4	d	0.8	**7.9**	1.2	
18	160.0 or 160.9	s	1.1 or 0.8	*f*	*f*	
19	50.5	t	1.0	1.0	1.0	
20	29.9	t	1.1	0.9	0.9	
21	122.7	d	1.2	0.9	1.0	
22	136.1 or 137.2	s	1.2 or 1.3	0.7 or 0.8	0.6 or 0.5	
23	160.0 or 160.9	s	1.1 or 0.8	*f*	*f*	

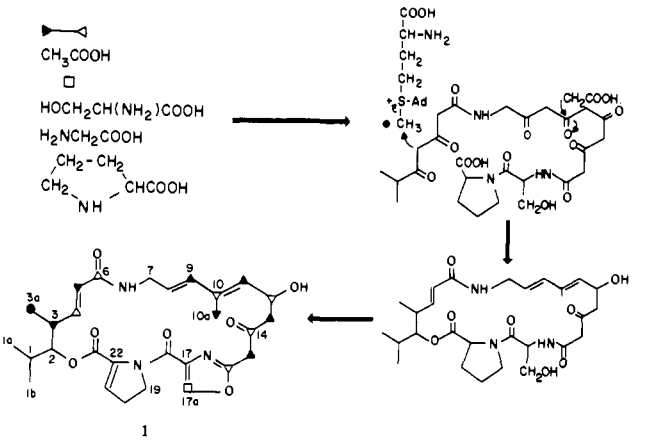
[a] Chemical shifts are downfield from internal Me_4Si in $CDCl_3$. [b] Multiplicities in the off-resonance-decoupled spectrum from ref 14. [c] Intensity of each peak in the labeled antibiotic divided by that of the corresponding peak in the unlabeled antibiotic, normalized to give a ratio of 1.0 for the peak for C-19. Ratios significantly above 1.0 in bold face type. [d] Carbon–carbon coupling observed when [1,2-$^{13}C_2$]acetate was incorporated into virginiamycin M_1. [e] Coupling between carbons 9 and 10 was observable, but the coupling was not first order, and the coupling constant was not determined. [f] Signals for these carbons, which were weak in the unlabeled antibiotic, were lost in the noise in spectra of these labeled compounds.

assignments (Table II) differ in some respects from those reported earlier.[14,15]

The ^{13}C NMR spectrum of virginiamycin M_1 derived from [2-^{13}C]acetate (Table II) showed seven enhanced signals corresponding to carbon atoms 3, 5, 9, 10a, 11, 13, and 15 of the macrocyclic ring. This finding, taken in conjunction with the known incorporation of proline, methionine, serine, and glycine into the antibiotic (Table I), and on the assumption that carbon atoms 1, 1a, 1b, and 2 are derived from valine or isobutyric acid, enables a tentative pathway for the biosynthesis of virginiamycin M_1 to be made (Scheme I). In this scheme, the intermediates shown are hypothetical, but the scheme does serve to indicate the manner in which the carbon skeleton of the molecule is assembled. It is assumed that the dehydroproline ring of the antibiotic arises from proline and that carbon atoms 7 and 8 arise from glycine; work is currently in progress to confirm these assumptions.

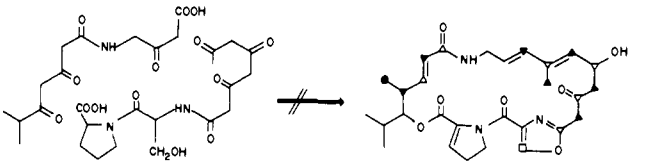
Corroboration of certain details of the pathway of Scheme I was obtained by studies with L-[methyl-^{13}C]methionine and DL-[3-^{13}C]serine. Methionine was found to donate its methyl group specifically to the C-3a methyl group, as indicated in the scheme. Incorporation of DL-[3-^{13}C]serine into virginiamycin M_1

Scheme I



1

Scheme II



proved to be more complex. The carbon atom showing the greatest enrichment was C-17a, consistent with the formation of the oxazole ring from an acyl serine precursor.[8,10] The oxazole ring in the alkaloid annuloline is biosynthesized by a different pathway, involving cyclization of an amide,[9] but this pathway is excluded for virginiamycin by the fact that alanine is not incorporated.[13] Lesser enrichments were observed for a number of other carbon atoms. Carbons 3, 5, 9, 10a, 11, 13, and 15 showed peak intensities approximately double those of unlabeled carbons, and a moderate enrichment of this type is in agreement with the known metabolism of serine via pyruvate to acetyl coenzyme A.[16] The somewhat higher enrichment of C-3a is consistent with the transfer of C-3 of serine to methionine via N^5-methyl tetrahydrofolate.[16] Finally, the enrichment observed for carbons 1a and 1b is consistent with their formation from valine, which in turn is biosynthesized from pyruvate derived from serine;[16] these carbons are not significantly enriched when [2-^{13}C]acetate is the precursor.

One aspect of the biosynthesis that is not established by the preceding experiments is the question as to whether the methyl group at position 10a arises by addition of an acetate unit to the carbonyl group at C-10, followed by decarboxylation, as shown in Scheme I, or whether it arises by condensation of an amino acetoacetate unit with a preformed polyketide chain, as shown in Scheme II. This question was settled by a labeling experiment with [1,2-$^{13}C_2$]acetate. The C–C couplings observed in the product from this experiment (Table II) showed clearly that the C-10a methyl group arose from an external acetate unit, as depicted in Scheme I.

The pathway of Scheme I thus represents our current knowledge of the biosynthesis of virginiamycin M_1. Work is in progress on the detailed pathways by which the oxazole ring and the dehydroproline double bond are formed and on the biosynthesis of other antibiotics of the virginiamycin family.

Acknowledgments. We thank Dr. D. H. Williams, Cambridge University, for a reference sample of virginiamycin M_1, Dr. C. J. DiCuollo, SmithKline Animal Health Products, for a sample of crude virginiamycin M_1 and the strain PDT of *S. virginiae*, T. Glass of this department for recording the NMR spectra, and M. Hanley for some preliminary studies. This work was supported by the College of Arts and Sciences, Virginia Polytechnic Institute

(14) Bycroft, B. W. *J. Chem. Soc., Perkin Trans. 1* **1977**, 2454–2470.
(15) Full details of the ^{13}C NMR assignments will be published elsewhere.
(16) Lehninger, A. L. "Biochemistry", 2nd ed.; Worth Publishers: New York, 1975.

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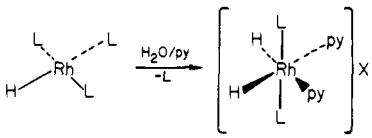
Activation of Water Molecules. 4. Generation of Dihydrogen from Water by Rhodium(I) Hydrido and Rhodium(0) Carbonyl Compounds

Sir:

Previously we reported the oxidative addition of water to monohydridorhodium(I) compounds ligated with electron-donating ligands, e.g., $RhHL_3$ [L = P(*i*-Pr)$_3$].[1] The product formed in a coordinating solvent like pyridine (py) is the cis-dihydride $[RhH_2(py)_2L_2]OH$ (**1a**), which can be isolated as its BPh_4 salt



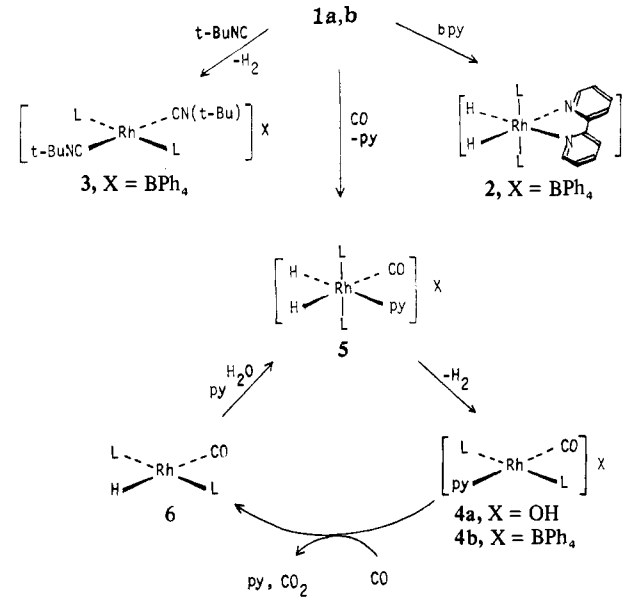
1a, X = OH
1b, X = BPh_4

(**1b**). In spite of its cis ligation, the Rh–H bonds in **1b** were found to be rather thermally stable, in contrast to $[RhH_2(S)_2(PPh_3)_2]^+$ (S = solvent) which dissociates H_2 in vacuo.[2a] When **1a** is heated (90 °C, in dioxane), it merely decomposes into an untractable oil, and irradiation (low-pressure Hg lamp) fails to give any perceptible production of dihydrogen. Our concern then was to produce dihydrogen by utilizing $RhHL_3$ via oxidative addition of water. However, the PPh_3 analogue, $RhH(PPh_3)_3$, which lacks sufficient nucleophilic character to undergo oxidative addition of water, cannot be a candidate for the purpose.

In the absence of systematic information on ligand effects on the Rh–H bond strength in cis-dihydrido compounds of type **1**, we investigated the effect of replacing the equatorial pyridine ligands with other ligands. First, 2,2′-bipyridine (bpy) was examined to see the chelating effect. The reaction of **1b** with bpy (room temperature, in THF) gave $\{RhH_2(bpy)[P(i\text{-}Pr)_3]_2\}BPh_4$[3] (**2**) (Scheme I). The cis-dihydrido ligation in **2** is readily established by its IR [ν(Rh–H) 2080, 2135 cm^{-1}] and ^{1}H NMR [−17.2 (q, Rh–H, $J_{H\text{-}P} = J_{H\text{-}Rh}$ = 15.6 Hz), 1.02 (q, CH$_3$, $^3J_{H\text{-}P} + {}^5J_{H\text{-}P}$ = 12.0 Hz, $J_{H\text{-}H}$ = 6.0 Hz)] data. **2** is somewhat more stable than **1b**, and no dihydrogen evolution is observed upon heating at 90 °C for 10 h in aqueous dioxane, **2** being recovered quantitatively.

Facile dihydrogen evolution from **1b** takes place by treatment with *t*-BuNC. Thus, on addition of *t*-BuNC to a THF solution of **1b** at room temperature, dihydrogen evolution commenced instantaneously with effervescence and was completed within a few minutes. From the solution, *trans*-$\{Rh(t\text{-}BuNC)_2[P(i\text{-}Pr)_3]_2\}BPh_4$[4] (**3**) was isolated as golden yellow crystals (80%). Brisk dihydrogen evolution also occurred on introduction of CO

Scheme I



into a THF solution of **1b** under ambient conditions, *trans*-$\{Rh(CO)(py)[P(i\text{-}Pr)_3]_2\}BPh_4$[5] (**4b**) being produced (80%). The formation of **4b** from **1b** probably involves an intermediate **5**[6] (see Scheme I). These results suggest that electron-donating ligands like pyridine and dipy stabilize the dihydrido coordination in **1a,b** whereas electron-withdrawing *t*-BuNC and CO reduce the bond strength.[7] Higher Rh–H stretching frequencies are observed for cis-dihydridobicarbonato, and -formato complexes, $RhH_2(B)L_2$ (B = HCO_3, ν(Rh–H) 2120, 2140 cm^{-1}; B = HCO_2, ν(Rh–H) 2130, 2145 cm^{-1}).[8] Consistent with these Rh–H stretching frequencies, these compounds do not generate dihydrogen at ambient temperature even in high vacuum.

A hydridocarbonyl compound, *trans*-$[RhH(CO)L_2]$ [**6**, L = P(*i*-Pr)$_3$],[9] prepared as yellow crystals by treating $RhHL_3$ [L = P(*i*-Pr)$_3$] with methanol at room temperature, undergoes oxidative addition of water, producing H_2 (70%) and *trans*-$\{Rh(CO)(py)[P(i\text{-}Pr)_3]_2\}OH$ (**4a**); the latter was isolated as **4b** (55%). Initial formation of the water adduct **5** must be postulated to account for the dihydrogen generation. Therefore, it is most likely, also reasonable, that the same intermediate **5** is involved for the two routes, **1b** → **4b** and **6** → **4a**.

An attempt to prepare **6** through a direct reaction of $RhHL_3$ [L = P(*i*-Pr)$_3$] with CO in *n*-hexane at ambient temperature failed. Unexpectedly, the product obtained was a binuclear Rh(0) carbonyl compound, $Rh_2(CO)_3L_3$ (**7**), as red crystals. The formulation of **7** is based on the elemental analysis, IR, and ^{1}H NMR data,[10] no indication being obtained for the presence of hydride ligands. The formation apparently proceeds via $[Rh(CO)_3L]_2$

(1) Yoshida, T.; Okano, T.; Saito, K.; Otsuka, S. *Inorg. Chim. Acta* **1980**, *44*, L135–L136.
(2) (a) Schrock, R. R.; Osborn, J. A. *J. Am. Chem. Soc.* **1971**, *93*, 1397–1401. (b) *Ibid.* **1976**, *98*, 2134–2143.
(3) Analytical sample obtained from THF–toluene contains 1 mol of toluene. Anal. Calcd for $C_{59}H_{80}N_2P_2BRh$: C, 71.36; H, 8.12; N, 2.82. Found: C, 71.01; H, 7.84; N, 2.92.
(4) Anal. Calcd for $C_{52}H_{80}N_2P_2BRh$: C, 68.92; H, 8.86; N, 2.87. Found: C, 68.77; H, 8.86; N, 3.08. IR (Nujol), ν(C≡N) 2115 cm^{-1}; ^{1}H NMR (acetone-d_6) 1.49 (s, *t*-Bu), 1.38 (q, CH$_3$, $^3J_{H\text{-}P} + {}^5J_{H\text{-}P}$ = 13.2 Hz, $J_{H\text{-}H}$ = 6.6 Hz), ~2.3 (m, CH).
(5) Analytical sample recrystallized from THF–toluene contains 2 mol of toluene. Anal. Calcd for $C_{62}H_{83}NOP_2BRh$: C, 72.01; H, 8.09; N, 1.35. Found: C, 72.03; H, 8.05; N, 1.43. IR (Nujol), ν(CO) 1985 cm^{-1}; ^{1}H NMR (THF-d_8) 1.25 (q, CH$_3$, $^3J_{H\text{-}P} + {}^5J_{H\text{-}P}$ = 13.0 Hz, $J_{H\text{-}H}$ = 6.5 Hz), ~1.9 (m, CH).
(6) Indirect support is a similar reaction of $[RhH_2(PEt_3)_3]OH$, a water adduct of $RhH(PEt_3)_3$, with CO which gives a hexacoordinate cis dihydride, $[RhH_2(CO)(PEt_3)_3]^+$. However, instead of **5**, a possibility of forming a nonsolvated pentacoordinate cis-dihydrido complex, $\{RhH_2(CO)[P(i\text{-}Pr)_3]_2\}^+$, could not be excluded.
(7) $[RhH_2(S)_2(PPh_3)_2]^+$ with CO is known to give $[Rh(CO)(S)(PPh_3)_2]^+$ while the reaction with $AsMe_2Ph$ or dipy gave $[RhH_2(PPh_3)_2L_2]^+$ (L = $AsMe_2Ph$ or L_2 = dipy).[2]
(8) Yoshida, T.; Thorn, D. L.; Okano, T.; Ibers, J. A.; Otsuka, S. *J. Am. Chem. Soc.* **1979**, *101*, 4212–4221.
(9) Anal. Calcd for $C_{19}H_{43}OP_2Rh$: C, 50.44; H, 9.58. Found: C, 50.49; H, 9.65. IR (Nujol), ν(Rh–H) 1980 cm^{-1}; ν(CO) 1920, 1942 cm^{-1}; ^{1}H NMR (benzene-d_6) −5.9 (dt, Rh–H, $J_{H\text{-}Rh}$ = 14.3 Hz, $J_{H\text{-}P}$ = 20.0 Hz), 1.23 (q, CH$_3$, $^3J_{H\text{-}P} + {}^5J_{H\text{-}P}$ = 13.8 Hz, $J_{H\text{-}H}$ = 6.9 Hz), ~2.0 (m, CH).
(10) Anal. Calcd for $C_{30}H_{63}O_3P_3Rh_2$: C, 46.57; H, 8.34. Found: C, 46.48; H, 8.19. IR (Nujol), ν(CO) 1732, 1768, 1957 cm^{-1}; ^{1}H NMR (benzene-d_6) ~1.0 (m, CH$_3$), ~1.8 (m, CH).

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