Synthesis of (*E*)- and (*Z*)-[2-((Dimethoxyphosphinyl)oxy)-3-ethoxy-3-oxo-1-propenyl]bromobis-(triphenylphosphine)palladium and the X-ray Crystal Structure Determination for the *E*-Isomer

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We report the preparation of (*E*)- and (*Z*)-[2-((dimethoxyphosphinyl)oxy)-3-ethoxy-3-oxo-1-propenyl]-bromobis(triphenylphosphine)palladium from the corresponding ethyl (*E*)- and(*Z*)-3-bromo-2-[(dimethoxyphosphinyl)oxy]propenoate and the X-ray crystal structure determination for (*E*)-[2-((dimethoxyphosphinyl)oxy)-3-ethoxy-3-oxo-1-propenyl]bromobis(triphenylphosphine)palladium. The (*E*)-palladium complex was reduced to (*E*)-3-[^{2}H]phosphoenolpyruvate by treatment with a mixture of trifluoroacetic acid-*d*/trifluoroacetic anhydride under strict anhydrous conditions. The ^{1}H-NMR spectrum of the *E*-isomer obtained from the reduction of the palladium complex was identical to the ^{1}H-NMR spectra of (*E*)-3-[^{2}H]phosphoenolpyruvate previously prepared by two different synthetic strategies. The X-ray structure of the palladium phosphoenolpyruvate analogue is the first X-ray structure of any 3-substituted phosphoenolpyruvate analogue reported and along with its synthesis and transformation interrelates the stereochemistry of several 3-substituted phosphoenolpyruvate analogues.

The exploration of the topology of the active site of an enzyme(s) with substrate analogs has proven to be productive in the design of enzyme inhibitors. The successful utilization of this methodology requires access to a library of substrate analogs in which not only the functionality but also their stereochemical disposition relative to each other are both varied and known with absolute certainty. Such an analysis of the active site terrain of several phosphoenolpyruvate (PEP)-utilizing enzymes has shown that the position of both the carboxyl and phosphate groups (attached to the same C-2 sp^2 carbon atom) as well as the bond angle (120°) between them plays an important role in the binding of substrate analogs to the active site of these enzymes.[1] Further studies involving the replacement of one of the two C-3 hydrogens of PEP with substituents of varying size and electronegativity have also proven extremely useful in deciphering the sometimes cryptic mechanism of these various PEP-utilizing enzymes.[2-11] Two problems limit the full utilization of this approach with PEP-type substrate analogs: (1) the inaccessibility on many occasions to both geometric isomers of the PEP analog, i.e., both the *E*- and *Z*-isomers, and (2) the lack of an unambiguous methodology for the assignment of the stereochemistry of the geometric isomer(s) available. In this paper we wish to report (1) the first direct proof of the stereochemistry of a 3-substituted PEP via X-ray crystallography of a PEP analog and (2) the development of a potentially general methodology for the synthesis of both geometric isomers of 3-substituted PEP analogs and utilization of the methodology in the synthesis of (*E*)-[3-^{2}H]PEP (Figure 1).

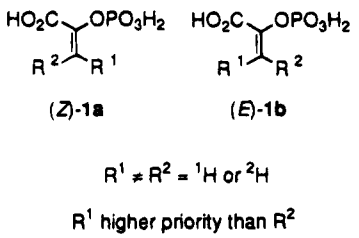

Figure 1. Structure of (*E*)- and (*Z*)-[3-^{2}H]PEP.

Results

(a) Synthesis. The synthetic route to (*E*)- and (*Z*)-[2-((dimethoxyphosphinyl)oxy)-3-ethoxy-3-oxo-1-propenyl]-bromobis(triphenylphosphine)palladium (Scheme 1) began with the bromination of commercially available ethyl bromopyruvate using *N*-bromosuccinimide in carbon tetrachloride to give ethyl 3,3-dibromopyruvate (**2**). Treatment of the ethyl 3,3-dibromopyruvate with trimethyl phosphite, under Perkow-type reaction conditions, gave the corresponding ethyl (*Z*)-3-bromo-2-[(dimethoxyphosphinyl)oxy]propenoate (**3a**) and ethyl (*E*)-3-bromo-2-[(dimethoxyphosphinyl)oxy]propenoate (**3b**) (28:72, *E*:*Z*). Separation of this mixture of ethyl bromophosphoenolpyruvates via medium pressure liquid chromatography afforded three distinct fractions: a major fraction *Z* > 98% stereochemically pure, a minor fraction *E* > 98%

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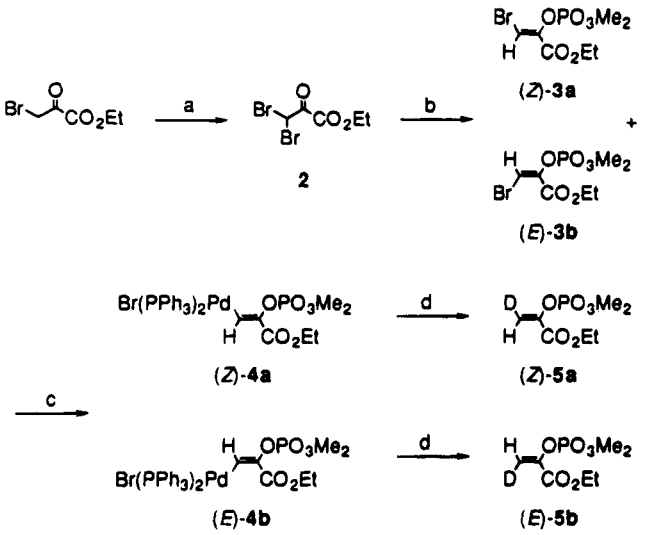
Scheme 1. Synthesis and Reaction of Vinylpalladium PEP Stereochemical Isomers[a]



[a] (a) *N*-Bromosuccinimide/CCl_4; (b) $P(OCH_3)_3/Et_2O$; (c) $Pd(PPh_3)_4$/benzene; (d) CF_3CO_2D, $(CF_3CO)_2O/CH_2Cl_2$.

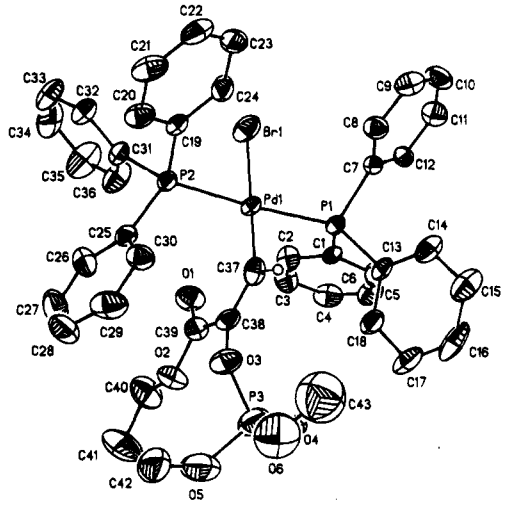


Figure 2. ORTEP representation of **4b** with crystallographic numbering system.

stereochemically pure, and a large mixed fraction containing both pyruvates (43:57, *E*:*Z*). Each of these ethyl bromophosphoenolpyruvate fractions was treated separately with a molar stoichiometric amount of tetrakis(triphenylphosphine)palladium(0) in benzene, except the mixed fraction was treated with a molar amount equal to that of only the *E*-isomer. In all three reactions, the palladium complex was formed in quantitative yields with respect to the palladium reagent. In the case of the reaction of the *E*/*Z*- mixture with tetrakis(triphenylphosphine)palladium(0), the (*Z*)-**4a** and (*E*)-**4b** palladium complexes (25:75) were easily separable by normal gravity silica gel chromatography. The pure (*E*)-palladium complex was reduced to the corresponding ethyl (*E*)-3-deuterio-2-[(dimethoxyphosphinyl)oxy]propenoate **5b** by treatment under strict anhydrous conditions with a mixture of trifluoroacetic acid-*d*/trifluoroacetic anhydride.

(b) X-ray Analysis. The X-ray crystal structure of the (*E*)-[2-((dimethoxyphosphinyl)oxy)-3-ethoxy-3-oxo-1-propenyl]bromobis(triphenylphosphine)palladium (**4b**) with its atomic numbering scheme is shown in Figure 2 and the data collection and refinement parameters are summarized in Table 1.[12]

Discussion

Analogs of PEP monosubstituted on the C-3 vinylic carbon have been synthesized for the most part by reacting β-halo-α-keto acids, bearing the desired substituent on the β-carbon, with a trialkyl phosphite in a Perkow-type reaction.[7-9] This synthetic route is nonstereospecific and yields a mixture of geometric isomers. The stereochemical assignments of these compounds have been determined from the ^1H NMR spectra of the vinylic protons. The vinyl proton(s) for both PEP and phosphoenolbutyrate (MePEP) were assigned from their respective 1-^{13}C labeled compounds by determining the H−C−C−^{13}C coupling constants.[7,13] In substituted alkenes the *trans* coupling constant has been shown to be greater than the *cis* coupling constant.[14] However, subsequent assignments of monosubstituted PEP analogs have been based upon relative chemical shifts and phosphorus coupling constants of the respective vinyl protons in comparison to MePEP and the *assumption* that the *Z*-isomer is always the major isomer obtained from the Perkow reaction.[7,8,10] The reliability of stereochemical assignments on such basis is suspect. Attempts at separation of the two geometric isomers often lead to the purification of only one isomer and an isomeric mixture, and/or loss of one isomer altogether. The loss of one of the isomers is either purposely, through enzymatic degradation with an enzyme of known stereospecificity,[15] or because of the instability of one of the isomers during chemical workup.[10] A number of substituents at C-3 of PEP such as bromo, chloro, fluoro, cyano, and methyl are known to produce an enhancement in both the binding and activity, depending on steric and electronic requirements of the PEP-utilizing enzyme.[5,7-10,16] However, due to the difficulty in obtaining both isomers pure and the ambiguity in the stereochemical assignments, little is known about the influence of the configuration at the C-3 position.

In order to develop a general methodology for the synthesis of both geometric isomers of monosubstituted C-3 PEP analogs of *known* stereochemistry from a common precursor, various metalation reactions, in which the stereochemistry of formation and reaction are known, were sought. Low oxidation state metal complexes, particularly those having d^8 and d^{10} configurations, such as palladium, are known to undergo oxidative addition with vinylic halides with retention of configuration to give σ-vinylpalladium complexes. For example, (*Z*)-[2-acetoxy-3-ethoxy-3-oxo-1-propenyl]bromobis(triphenylphosphine)palladium has been synthesized from ethyl (*Z*)-2-acetoxy-3-bromoacrylate (a PEP analog in which the phosphinyloxy moiety has been replaced by an

(12) The authors have deposited the atomic coordinates for this structure with the Cambridge Crystallographic Data Centre. The coordinates can be obtained, on request, from the Director, Cambridge Crystallographic Data Centre, 12 Union Road, Cambridge CB2 1EZ, UK.

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Table 1. Summary of Crystallographic Data for 4b

Crystal Data	
empirical formula	$C_{43}H_{42}O_6P_3BrPd$
formula weight	934.036 amu
crystal color and habit	yellow plate
crystal dimensions (mm)	0.38 × 0.22 × 0.04
crystal system	monoclinic
space group	$P2_1/c$(#14)
Z	4
unit cell dimensions from 35 reflctns ($8.1° \leq 2\theta \leq 34.0°$)	
a	13.794(8) Å
b	10.169(2) Å
c	30.190(9) Å
α	90.000°
β	91.07(3)°
γ	90.000°
volume	4234(3) Å^3
density (calcd)	1.46_5 g cm^{-3}
F(000)	1896 electrons
linear absorption coefficient (μ)	15.21 cm^{-1}
Data Collection	
diffractometer	Syntex $P2_1$m/v
radiation type	Mo Kα, λ = 0.71073 Å, Lp corrected, graphite monochromator
temperature	ambient
scan type	$\theta/2\theta$ scan
2θ scan range	5−50°
octants used	$+h, +k, +l$ (h: 0/17; k: 0/13; l; 36/36)
scan rate	1.5−5.0 deg per min, variable
scan width	0.8° below Kα1 to 0.8° above Kα2
background/scan ratio	0.5
standard reflections	3 measd every 97 reflctns, linear decay ~3%
no. of data collected	8515
no. of unique reflections	6780, R_{int} = 0.0384
absorption correction	semiempirical, psi scans
R merge before/after correction	0.0401/0.0275
max/min transmission	0.974/0.720
Solution and Refinement	
system used	Siemens SHELXTL PLUS, VAXStation 3500
solution	Patterson
refinement method	full-matrix least-squares
function minimized	$\Sigma w(\|F_o - F_c\|)^2$
hydrogen atoms	riding model, d_{C-H} = 0.96 Å, common isotropic U(H) refined to 0.074(7)
refined reflections with $(F_o) \geq 3\sigma(F)$	4632
no. of parameters refined	483
data/parameter ratio	9.6
$R = \Sigma(\|F_o - F_c\|)/\Sigma(\|F_o\|)$	0.0829
$R_w = [\Sigma(w\|F_o - F_c\|)^2/\Sigma w(F_o)^2]^{1/2}$	0.0926
$w^{-1} = \sigma^2(F_o) + 0.001503(F_o)^2$	
GOF	1.55
mean shift/error	<0.001
max shift/error	<0.001
secondary extinction	6 reflctns excluded from refinement
residual electron density	+1.13/−1.47 e/Å^3

acetoxy group), indicating that the metal insertion reaction occurs with retention of configuration.[17] Therefore, the metalation of the PEP analogs ethyl (*Z* and *E*)-3-bromo-2-[(dimethoxyphosphinyl)oxy]propenoate (**3a** and **3b**) with tetrakis(triphenylphosphine)palladium(0) was chosen as the first target since the synthesis of **3a** and **3b** is straightforward and high yielding and a simple chromatographic method for the separation of **3a** from **3b** had been developed in our laboratory.[18] The σ-vinylpalladium PEP compounds **4a** and **4b** were synthesized individually from pure **3a** and **3b**, respectively. The ^{1}H NMR spectra of **4a** and **4b** indicate that they are stereochemically pure and of opposite configuration, as judged by the resonance of the C-3 vinylic hydrogen atoms. The σ-vinylpalladium complexes derived from the reaction of a mixture of **3a** and **3b** with tetrakis(triphenylphosphine)palladium(0) proved easily separable by silica gel chromatography to give 25% pure *Z* (**4a**) and 75% pure *E* (**4b**). The complexes **4a** and **4b** form excellent crystals from methylene chloride/diethyl ether/cyclohexane. The X-ray crystal structure of **4b** shown in Figure 2 constitutes the first direct proof of the stereochemistry of a 3-monosubstituted PEP analog.

Fryzuk and Bosnich[17] have synthesized (*Z*)-2-acetoxy-3-deuterioacrylate by reduction of (*Z*)-[2-acetoxy-3-ethoxy-3-oxo-1-propenyl]bromobis(triphenylphosphine)palladium with trifluoroacetic acid-*d*/trifluoroacetic anhydride in methylene chloride. The stereochemistry of the deuteriated acrylate was unequivocally demonstrated. Identical results were obtained with the *E*-isomer. It can therefore be concluded that the cleavage of the σ-vinylpalladium bond with trifluoroacetic acid-*d*/trifluoroacetic anhydride proceeds with complete retention of configuration. By analogy, the conversion of **4a** and/or

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4b to their corresponding 3-deuterio compounds should occur with retention of configuration. This reduction of **4a** and/or **4b** would therefore complete a cyclic proof since ethyl (*E*)- and (*Z*)-3-deuterio-2-[(dimethoxyphosphinyl)-oxy]propenoate have been previously synthesized by Bartlett and Chouinard[19] utilizing two different stereo-controled routes, both of which are independent of a Perkow-type reaction and involve reactions with well-documented stereochemistry. The *E*-isomer was prepared via a process that involved the *anti* elimination of HBr from a phosphorylated bromohydrin and the *Z*-isomer via hydrogen peroxide oxidation of a benzeneselenide with concomitant *syn* elimination of the selenoxide. In addition, our laboratory has reported the reduction of the ethyl (*Z*)-3-bromo-2-[(dimethoxyphosphinyl)oxy]propenoate (**3a**) and ethyl (*E*)-3-bromo-2-[(dimethoxyphosphinyl)oxy]propenoate (**3b**) with Zn/Ag couple in THF/^2H_2O (4:1) at 25 °C to give the corresponding ethyl (*Z*)- and (*E*)-3-deuterio-2-[(dimethoxyphosphinyl)oxy]propenoates.[18] The reduction of vinylic halides via Zn/Ag couple in THF/^2H_2O has been shown to occur with retention of configuration.[17,20] The ^{1}H-NMR assignment of the vinyl hydrogens of the two geometric isomers of [3-^{2}H]PEP prepared by both the Bartlett and the Woodard methods are in agreement. The stereochemistry of the product obtained from the reduction of **4b** with trifluoroacetic acid-*d*/trifluoroacetic anhydride was identical to that of the *E*-isomer obtained by both Bartlett and Chouinard and our laboratory, indicating that our σ-vinylpalladium bond cleavage reaction proceeded with retention of configuration. On the basis of our previous studies of the reduction of **3a** and **3b** with Zn/Ag couple in THF/^2H_2O[18] and comparison of the stereochemistry of the resulting deuterated PEP with the Bartlett and Chouinard work, as well as the data presented in this paper, the absolute stereochemistry of **3a, 3b, 4a,** and **4b** have been interrelated and firmly verified.

Since σ-vinylpalladium compounds react with various nucleophiles, such as alkyllithium, alkylzinc, alkylcopper, Grignard reagents, and potassium cyanide/18-crown-6, to yield nucleophile coupled olefin with retention of configuration at the olefinic double bond, compounds **4a** and **4b** may serve as intermediates leading to the synthesis of geometric pairs of 3-monosubstituted PEP analogs. Work is in progress to explore the scope of these conversions.

Experimental Section

All melting points are uncorrected. All literature melting points are for the nondeuteriated compounds. Elemental analyses are within ±0.4%. The incorporation of deuterium in the various compounds was determined via ^{1}H NMR.

Inorganic and organic reagents were purchased from the usual chemical sources and used without further purification. All organic solvents were of reagent grade and were used without further purification. Solvents were dried over anhydrous Na_2SO_4 and evaporated in vacuo using a rotary evaporator (water aspirator vacuum) at 40 °C unless stated otherwise. Medium grade silica gel (Merck 70–230 mesh) was used for column chromatography. TLC plates (silica, Analtech) were visualized by ultraviolet irradiation from a Mineralight short wave UV lamp or iodine chamber visualization.

Ethyl (*Z*)-3-Bromo-2-[(dimethoxyphosphinyl)oxy]propenoate (3a) and Ethyl (*E*)-3-Bromo-2-[(dimethoxyphosphinyl)oxy]propenoate (3b). The title compounds were prepared as a 28/72 (*E*:*Z*) mixture of geometric isomers, determined by high field NMR, and separated by the method previously described by this lab.[18] **E-Isomer:** ^{1}H NMR ($CDCl_3$) δ 6.96 (d, J = 2.9 Hz, 1 H), 4.34 (q, J = 7.3 Hz, 2 H), 3.87 (d, J = 11.4 Hz, 6 H), 1.26 (t, J = 7.3 Hz, 3 H). **Z-isomer:** ^{1}H NMR ($CDCl_3$) δ 7.24 (d, J = 1.7 Hz, 1 H), 4.30 (q, J = 7.3 Hz, 2 H), 3.95 (d, J = 11.5 Hz, 6 H), 1.33 (t, J = 7.3 Hz, 3 H); exact mass 302.9626 (302.9633 calcd for $C_7H_{12}{}^{79}BrPO_6$).

(*E*)-[2-((Dimethoxyphosphinyl)oxy)-3-ethoxy-3-oxo-1-propenyl]bromobis(triphenylphosphine)palladium (4b). A solution of the pure (*E*)-vinyl bromide **3b** (262.5 mg, 0.86 mmol) in benzene (1 mL) was added by syringe to a stirred solution of $Pd(PPh_3)_4$ (1 g, 0.86 mmol) in benzene (6 mL) under nitrogen. The reaction mixture was heated at 65–68 °C for 2 h. The reaction was allowed to cool to 25 °C and 50 mL of anhydrous Et_2O added. The precipitate which formed was filtered and washed with an additional 10 mL (2 × 5 mL) of anhydrous diethyl ether. The yellow solid was taken up in $CHCl_3$ (5 mL) and filtered through a 1 × 4 cm silica gel column, and the column was washed with 50 mL of 5% MeOH in $CHCl_3$. The solvent was removed from the clear yellow filtrate. The yellow solid obtained was recrystallized from CH_2Cl_2/anhydrous Et_2O/cyclohexane (3:5:10) to give the title (*E*)-vinylpalladium derivative **4b** in quantitative yield as yellow plates (mp 138 °C decomp): ^{1}H NMR ($CDCl_3$) δ 7.68 (bs, 12 H), 7.36 (m, 18 H), 6.97 (bs, Hz, 1 H), 3.87 (q, J = 7.1 Hz, 2 H), 3.40 (d, J= 11.3 Hz, 6 H), 1.08 (t, J = 7.1 Hz, 3 H); ^{13}C NMR (90 MHz, $CDCl_3$) δ 14.1, 53.9, 60.7, 128.0, 130.0, 131.2, 134.0, 134.8, 151.3 (carbonyl carbon unresolved).

(*Z*)-[2-((Dimethoxyphosphinyl)oxy)-3-ethoxy-3-oxo-1-propenyl]bromobis(triphenylphosphine)palladium (4a). The (*Z*)-vinyl bromide **3a** (262.5 mg, 0.86 mmol) was reacted with $Pd(PPh_3)_4$ (1.0 g, 0.86 mmol) in benzene (6 mL) as described above for the *E*-derivative to give the (*Z*)-vinylpalladium compound **4a** as a yellow solid (mp 170–171 °C) in quantitative yield: ^{1}H NMR ($CDCl_3$) δ 7.71 (m, 12 H), 7.36 (m, 18 H), 7.10 (t, J = 6.0 Hz, 1 H), 3.77 (q, J = 7.1 Hz, 2 H), 3.67 (d, J = 11.4 Hz, 6 H), 1.00 (t, J = 7.1 Hz, 3 H); ^{13}C NMR (90 MHz, $CDCl_3$) δ 14.3, 54.5, 60.1, 128.0, 130.3, 130.8, 135.0, 138.8, 155.6, 159.2; MS 934 (M^+, 0.9), 853 (15.9), 630 (22.9), 591 (91.5), 485 (100), 367 (20.5), 262 (51.5). Anal. Calcd for $C_{43}H_{42}BrO_6P_3Pd$: C, 55.29; H, 4.54. Found: C, 55.28; H, 4.61.

(*Z*)-[2-((Dimethoxyphosphinyl)oxy)-3-ethoxy-3-oxo-1-propenyl]bromobis(triphenylphosphine)palladium (4a) and (*E*)-[2-((Dimethoxyphosphinyl)oxy)-3-ethoxy-3-oxo-1-propenyl]bromobis(triphenylphosphine)palladium (4b). A mixture of the vinyl bromides (920 mg, 3.04 mmol, *E*/*Z* (43/57)), obtained as a middle fraction from silica gel MPLC (30% ethyl acetate/hexanes) of a 28/72 (*E*/*Z*) mixture, was reacted with $Pd(PPh_3)_4$ (1.51 g, 1.31 mmol; 1 equiv with respect to the (*E*)-vinyl bromide) in benzene (5 mL) as described above for the individual isomers. The reaction was worked up as described above to yield a 75:25 (*E*:*Z*) mixture of the title σ-vinylpalladium compounds in quantitative yield with respect to $Pd(PPh_3)_4$. The remaining ether filtrate contained only triphenylphosphine and (*Z*)-vinyl bromide **3a**, but no (*E*)-vinyl bromide **3b**, as determined by ^{1}H NMR. The (*E*)- and (*Z*)-vinylpalladium derivatives were separated by silica gel (30 g) flash chromatography using a step gradient of 0, 1, 2, and 3% MeOH in $CHCl_3$ (100 mL each). The elution was monitored by TLC developed with 3% MeOH in chloroform and visualized by UV. The (*E*)-vinylpalladium **4b** eluted first. Both isomers were recrystallized as described above and displayed the same physical properties.

Ethyl (*E*)-3-Deuterio-2-[(dimethoxyphosphinyl)oxy]-propenoate (5b). To an oven-dried (120 °C) 25 mL round-bottomed flask containing the (*E*)-vinylpalladium compound **4b** (0.40 g, 0.433 mmol) in CH_2Cl_2 (5 mL; distilled over CaH_2) under nitrogen was added, via syringe, a solution of CF_3CO_2D 99.5 atom % D (750 mg, 6.49 mmol; vial open under a heavy flow of nitrogen) and $(CF_3CO)_2O$ (91 mg, 0.433 mmol). The reaction was heated, under nitrogen, at reflux for 24 h. The reaction was allowed to cool to 25 °C and the solvent removed by rotary evaporation. The residue was taken up in CCl_4 (5 mL), the solvent was removed by rotary evaporation, and the whole procedure was repeated twice. Finally, the residue was

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taken up in CCl_4 (2.5 mL), and Et_2O (5 mL) was added. After refrigeration for 12 h, yellow crystals formed. The crystals were filtered and washed with Et_2O (2 × 2.5 mL). All the solvent washes were combined and evaporated to give a yellow oil from which a second crop of yellow crystals was obtained. The yellow oil was taken up in Et_2O and extracted with an aqueous 0.1 M thiourea solution (5 × 5 mL) to remove the final traces of the palladium complexes. The ether layer was dried and removed under vacuum to give the title compound (**5b**) as a colorless oil (0.078g, 80% yield): ^{1}H NMR ($CDCl_3$) δ 5.62 (d, J = 2.4 Hz, 1 H), 4.28 (q, J = 7.3 Hz, 2 H), 3.88 (d, J = 11.4 Hz, 6 H), 1.33 (t, J = 7.3 Hz, 3 H). Analysis of the vinylic proton resonances in the ^{1}H NMR indicated that there was less than 5% of the *Z* isomer present.

Cyclohexylammonium (*E*)-3-Deuteriophosphoenolpyruvate (1b). To a stirred solution of **5b** (0.039 g, 0.173 mmol) in CH_2Cl_2 (5 mL) was added TMSBr (0.066 g, 0.433 mmol). The reaction was allowed to stir at 25 °C for 2 h, and the solvent was removed. The resulting residue was dissolved in 0.5 mL of H_2O and titrated with 3.5 equiv of 0.1 N KOH, added over a 4 h period, with the pH never exceeding 10. After the addition of the base was completed, the solution was stirred for 12 h and desalted by passing through a 1 × 10 cm column of Dowex 50W resin (100–200 mesh) in the H^+ form. One equivalent of cyclohexylamine (0.173 mmol) was added to the column eluate and the solution freeze-dried to give 0.02 g (69% yield) **1b** as the monocyclohexylamine salt, mp 148 °C; lit.[19] mp 150–151 °C. Analysis of the vinylic proton resonances in the ^{1}H NMR spectra indicated that some isomerization occurred upon base hydrolysis of the ethyl ester, resulting in 18% contamination by the *Z*-isomer.

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