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Studies of *O,O*-Dimethyl α-(2,4-Dichlorophenoxyacetoxy)ethylphosphonate (HW02) as a New Herbicide. 1. Synthesis and Herbicidal Activity of HW02 and Analogues as Novel Inhibitors of Pyruvate Dehydrogenase Complex

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Supporting Information

ABSTRACT: On the basis of the previous work for optimization of *O,O*-diethyl α-(substituted phenoxyacetoxy)alkylphosphonates, further extensive synthetic modifications were made to the substituents in alkylphosphonate and phenoxy moieties of the title compounds. New *O,O*-dimethyl α-(substituted phenoxyacetoxy)alkylphosphonates were synthesized as potential inhibitors of pyruvate dehydrogenase complex (PDHc). Their herbicidal activity and efficacy in vitro against PDHc were examined. Some of these compounds exhibited significant herbicidal activity and were demonstrated to be effective inhibitors of PDHc from three different plants. The structure−activity relationships of these compounds including previously reported analogous compounds were studied by examining their herbicidal activities. Both inhibitory potency against PDHc and herbicidal activity of title compounds could be increased greatly by optimizing substituent groups of the title compounds. *O,O*-Dimethyl α-(2,4-dichlorophenoxyacetoxy)ethylphosphonate (**I-5**), which acted as a competitive inhibitor of PDHc with much higher inhibitory potency against PDHc from *Pisum sativum* and *Phaseolus radiatus* than from *Oryza sativa*, was found to be the most effective compound against broadleaf weeds and showed potential utility as herbicide.

KEYWORDS: α-(substituted phenoxyacetoxy)alkylphosphonate, herbicidal activity, pyruvate dehydrogenase complex, inhibitor, structure−activity relationships

INTRODUCTION

Pyruvate dehydrogenase complex (PDHc) is already known to be one of the target enzymes attacked by some herbicidally active compounds.[1−5] It plays a pivotal role in cellular metabolism, catalyzing the oxidative decarboxylation of pyruvate and the subsequent acetylation of coenzyme A (CoA) to acetyl coenzyme A.[6−8] The complex consists of three enzymes and a number of cofactors. Decarboxylation of pyruvate is the first step in this conversion. This step is catalyzed by pyruvate decarboxylase (PDHc E1), which promotes the decarboxylation of pyruvate using thiamin pyrophosphate (TPP) and Mg^{2+} as cofactors;[8,9] therefore, PDHc E1 as a target is of interest from the point of view of agrochemical design.

There are some reports about the design of inhibitors of PDHc.[7] An attempt to design inhibitors of PDHc E1 as herbicides using biochemical reasoning was reported by Baillie et al.[1] A series of acylphosphinates and acylphosphonates as analogues of pyruvate have been prepared as mechanism-based inhibitors of PDHc. Although these acylphosphinates and acylphosphonates were not active enough for full development as herbicides,[1,2] this provided a clue to the rational design of PDHc inhibitor. These findings prompted us to perform our own study for novel PDHc inhibitors with potential as herbicides.

Considering the above results of Baillie et al.'s work, the structural unit of the phosphonate molecule was kept, and an aryl or a heterocycle group was introduced into the phosphonate molecule to form 10 series of α-oxophosphonic acid derivatives, which have been synthesized and reported.[10] It was found that some *O,O*-diethyl α-(substituted phenoxyacetoxy)alkylphosphonates $\mathbf{I_o}$ exhibited significant herbicidal activity against dicotyledonous plants among 10 series of compounds. Compound $\mathbf{I_o}$ as a lead structure for the control of broadleaf weeds was modified, and it was extended to a general structure **I** (Scheme 1). In structure **I**, there are six different substituents, which can be changed by chemical modifications. It is possible that the biochemical properties of compound **I** will be remarkably variable by combinations of R^1, R^2, R^3, R^4, X, and Y.

In our previous work, some α-(substituted phenoxyacetoxy) alkylphosphonates **I** have been prepared. From the assay results of those compounds, it has been found that the herbicidal activity was greatly improved when the PrO or EtO group attached to phosphorus was replaced by a MeO group and H oas R^4 in the molecule. This suggested that smaller R^1O, R^2O, and R^4 groups are beneficial to herbicidal activity. Therefore, Me group as R^1 and R^2 and H as R^4 were kept in structure **I**, and further optimization was focused on the modification of substituents R^3, X, and Y. It is expected that the herbicidal activity would be improved by the chemical modifications of R^3, X, and Y. Here, we report the synthesis of 35 new *O,O*-dimethyl

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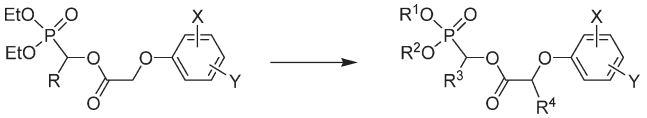
Scheme 1. Chemical Modification of Lead Structure I_o



Lead structure I_o **I**

α-(substituted phenoxyacetoxy)alkylphosphonates **I-1**−**I-35**. Structure−activity relationships of the synthesized compounds including previously reported analogous compounds **I-36**−**I-70** [11−22] were studied by examining their herbicidal activities. The detailed structure information of **I-1**−**I-70** can be found in the Supporting Information. The influence on herbicidal activity by the chemical modification of R^1, R^2, R^3, R^4, X, and Y in compound **I** was discussed.

MATERIALS AND METHODS

Synthesis Procedures. Chemicals and reagents were obtained from commercial sources, and all solvents were anhydrous. Column chromatography was carried out with Merck silica gel (230−400 mesh). Thin-layer chromatography (TLC) was performed on silica gel GF-254. Melting points (mp) were measured on an electrothermal melting-point apparatus and uncorrected. IR spectra were recorded as KBr pellets on a Perkin-Elmer FT-IR spectrophotometer or on a Nicolet Avatar360 FT-IR spectrometer; only the most significant absorption bands were recorded. ^{1}H NMR was recorded in deuterochloroform solution on a Varian Mercury-Plus 200 spectrometer at 200 MHz, a Varian XL-300 spectrometer at 300 MHz, or a Varian Mercury-Plus 400 spectrometer at 400 MHz, using tetramethylsilane as an internal standard. Chemical shifts (δ) are given in parts per million, and coupling constants (J) in hertz, and multiplicities are indicated by s (singlet), d (doublet), t (triplet), q (quartet), and m (multiplet). MS was analyzed on a Finnigen TRACE spectrometer and API2000LC/MS. Elemental analyses were performed with a Vario EL III elemental analyzer. The results of elemental analyses for C, H, and N were within ±0.5% of the theoretical values.

Phosphorus trichloride, triethylamine, and thionyl chloride were distilled before the reaction. Samples were purified by flash chromatography with silica gel.

Dimethyl phosphite **1** was used directly as obtained commercially. *O*,*O*-Dimethyl α-hydroxyalkylphosphonates **2** could be prepared by addition reaction of dimethyl phosphite **1** and several kinds of aldehydes using triethylamine as a catalyst according to the literature[23] or using potassium fluoride and alumina (mass ratio of 1:1) as a catalyst according to the literature.[24] Substituted phenoxyacetic acid **3** and substituted phenoxyacetyl chloride **4**, fluorophenoxyacetic acid or 3-trifluoromethyl phenoxyacetic acid **5**, and fluorophenoxyacetyl chloride or 3-trifluoromethyl phenoxyacetyl chloride **6** were prepared according to the methods previously reported.[11,19,21]

General Procedure for the Preparation of *O*,*O*-Dimethyl α-(Substituted phenoxyacetoxy)alkylphosphonates I-1−I-5, I-9−I-14, I-19, I-21−I-26, I-33, and I-34. A solution of substituted phenoxyacetyl chlorides **4** (0.022 mol) in trichloromethane (10 mL) was added to a stirred mixture of *O*,*O*-dimethyl α-hydroxyalkylphosphonates **2** (0.02 mol) and pyridine (0.022 mol) in trichloromethane (25 mL) at 10−25 °C. The resultant mixture was stirred for 3−5 h at room temperature and then for 1−2 h at 40−42 °C. The trichloromethane layer was washed with 0.1 M hydrochloric acid, saturated sodium hydrogen carbonate solution, and brine, dried, and concentrated. The residue was purified by column chromatography on silica gel and elution with petroleum ether/acetone (2:1, v/v) to give the corresponding pure title compounds **I-1**−**I-5**, **I-9**−**I-14**, **I-19**, **I-21**−**I-26**, **I-33**, and **I-34**.

Their structures were confirmed by ^{1}H NMR, IR, and MS spectra and elemental analysis. Physicochemical and spectroscopic data for compounds **I-1**−**I-5**, **I-9**−**I-14**, **I-19**, **I-21**−**I-26**, **I-33**, and **I-34** are as follows.

*O,O-Dimethyl α-(2,3-dimethylphenoxyacetoxy)ethylphosphonate (**I-1**):* yellow liquid; yield, 85%; n_D^{20} 1.5020; IR (KBr, cm^{-1}) ν 2965 (R—H), 1771 (C═O), 1250 (Ar—O—C), 1186 (C—O—C), 1031 (P—O—C), 771 (P—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.48−1.54 (m, 3H, CH_3—CH), 2.20−2.27 (d, 6H, 2 × CH_3), 3.69−3.80 (dd, 6H, 2 × OCH_3), 4.69 (s, 2H, OCH_2CO), 5.37−5.42 (m, 1H, CH—CH_3), 6.65−7.04 (m, 3H, C_6H_3). Anal. Calcd for $C_{14}H_{21}O_6P$: C, 53.16; H, 6.69. Found: C, 53.66; H, 6.29.

*O,O-Dimethyl α-(4-bromophenoxyacetoxy)ethylphosphonate (**I-2**):* yellow liquid; yield, 84%; n_D^{20} 1.4920; IR (KBr, cm^{-1}) ν 3092 (Ar—H), 2957 (R—H), 1770 (C═O), 1581(Ar C—C), 1175 (C—O—C), 1080 (C—Br), 700 (P—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.48−1.54 (m, 3H, CH_3—CH), 3.76−3.81 (dd, 6H, 2 × OCH_3), 4.66 (s, 2H, OCH_2CO), 5.38−5.42 (m, 1H, CH—CH_3), 6.79−7.41 (m, 4H, C_6H_4); Anal. Calcd for $C_{12}H_{16}BrO_6P$: C, 39.26; H, 4.39. Found: C, 39.21; H, 4.33.

*O,O-Dimethyl α-(2,4-dibromophenoxyacetoxy)ethylphosphonate (**I-3**):* yellow liquid; yield, 84%; n_D^{20} 1.5032; IR (KBr, cm^{-1}) ν 3069 (Ar—H), 2956 (R—H), 1770 (C═O), 1579 (Ar C—C), 1080 (C—Br); ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.48−1.54 (m, 3H, CH_3—CH), 3.76−3.81 (dd, 6H, 2 × OCH_3), 4.78 (s, 2H, OCH_2CO), 5.37−5.41 (m, 1H, CH—CH_3), 6.70−7.70 (m, 3H, C_6H_3). Anal. Calcd for $C_{12}H_{15}Br_2O_6P$: C, 32.31; H, 3.39. Found: C, 32.20; H, 3.59.

*O,O-Dimethyl α-(4-chloro-2-nitrophenoxyacetoxy)ethylphosphonate (**I-4**):* yellow liquid; yield, 84%; n_D^{20} 1.5173; IR (KBr, cm^{-1}) ν 3085 (Ar—H), 2958 (R—H), 1768 (C═O), 1534 (Ar C—C), 1032 (C—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.26−1.54 (m, 3H, CH_3—CH), 3.76−3.81 (dd, 6H, 2 × OCH_3), 4.83 (s, 2H, OCH_2CO), 5.36−5.40 (m, 1H, CH—CH_3), 6.98−7.88 (m, 3H, C_6H_3). Anal. Calcd for $C_{12}H_{15}ClNO_8P$: C, 39.20; H, 4.11; N, 3.81. Found: C, 39.42; H, 4.50; N, 3.18.

*O,O-Dimethyl α-(2,4-dichlorophenoxyacetoxy)ethylphosphonate (**I-5**):* light yellow liquid; yield, 89%; n_D^{20} 1.5172; IR (KBr, cm^{-1}) ν 3030 (Ar—H), 2980, 2890 (R—H), 1745 (C═O), 1580 (C═C), 1260 (P═O), 1170, 1060 (P—O—C), 810 (Ar—Cl), 719 (P—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.48−1.60 (t, 3H, CH_3), 3.76−3.85 (dd, 6H, 2 × OCH_3), 4.75 (s, 2H, OCH_2CO), 5.25−5.40 (m, 1H, CH), 6.8−7.5 (m, 3H, C_6H_3). Anal. Calcd for $C_{12}H_{15}Cl_2O_6P$: C, 40.36; H, 4.23. Found: C, 40.28; H, 4.12.

*O,O-Dimethyl 1-(2,4-dichlorophenoxyacetoxy)-2,2,2-trichloroethylphosphonate (**I-9**):* colorless solid; yield, 62%; mp 77−78 °C; IR (KBr, cm^{-1}) ν 3038 (Ar—H), 1794 (C═O), 1615, 1507 (C═C), 1250 (P═O), 1040 (P—O), 800 (Ar—Cl), 750 (P—C); ^{1}H NMR (200 MHz, $CDCl_3$) δ 3.80−3.90 (dd, 6H, 2 × OCH_3), 4.90 (s, 2H, OCH_2CO), 6.0 (d, 1H, CHP), 6.8−7.4 (m, 3H, C_6H_3); ^{31}P NMR (81 MHz, $CDCl_3$) δ 12.653. Anal. Calcd for $C_{12}H_{12}Cl_5O_6P$: C, 31.30; H, 2.63. Found: C, 31.19; H, 2.72.

*O,O-Dimethyl α-(2,4-dichlorophenoxyacetoxy)propylphosphonate (**I-10**):* colorless solid; yield, 68%; mp 58−60 °C; IR (KBr, cm^{-1}) ν 3010 (Ar—H), 1740 (C═O), 1580, 1490 (C═C), 1240 (P═O), 800 (Ar—Cl), 738 (P—C); ^{1}H NMR (200 MHz, $CDCl_3$) δ 1.0 (t, 3H, CH_3), 1.9 (m, 2H, CH_2), 3.80−3.90 (dd, 6H, 2 × OCH_3), 4.8 (s, 2H, OCH_2CO), 5.3 (m, 1H, CHP), 6.8−7.5 (m, 3H, C_6H_3); ^{31}P NMR (81 MHz, $CDCl_3$) δ 22.706. Anal. Calcd for $C_{13}H_{17}Cl_2O_6P$: C, 42.07; H, 4.62. Found: C, 42.36; H, 4.34.

*O,O-Dimethyl α-phenoxyacetoxybenzylphosphonate (**I-11**):* white solid; yield, 79%; mp 63.0−64.0 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.63−3.71 (dd, 6H, 2 × OCH_3), 4.75 (d, 1H^a, OCH_2CO), 4.78

(d, 1H^b, OCH_2CO), 6.26−6.29 (d, 1H, OCHP), 6.88−7.45 (m, 10H, $C_6H_5 + C_6H_5$). Anal. Calcd for $C_{17}H_{19}O_6P$: C, 58.29; H, 5.47. Found: C, 57.92; H, 5.13.

*O,O-Dimethyl α-(3-methylphenoxyacetoxy)benzylphosphonate (**I-12**):* yellow liquid; yield, 88%; n_D^{20} 1.5273; ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.18 (s, 3H, CH_3), 3.63−3.72 (dd, 6H, 2 × OCH_3), 4.73 (d, 1H^a, OCH_2CO), 4.76 (d, 1H^b, OCH_2CO), 6.25−6.28 (d, 1H, CHP), 6.77−7.47 (m, 9H, $C_6H_4 + C_6H_5$). Anal. Calcd for $C_{18}H_{21}O_6P$: C, 59.34; H, 5.81. Found: C, 59.09; H, 5.50.

*O,O-Dimethyl α-(4-methylphenoxyacetoxy)benzylphosphonate (**I-13**):* white solid; yield, 84%; mp 80.3−81.0 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.27 (s, 3H, CH_3), 3.62−3.70 (dd, 6H, 2 × OCH_3), 4.71 (d, 1H^a, OCH_2CO), 4.74 (d, 1H^b, OCH_2CO), 6.25−6.28 (d, 1H, OCHP), 6.77−7.47 (m, 9H, $C_6H_4 + C_6H_4$). Anal. Calcd for $C_{18}H_{21}O_6P$: C, 59.34; H, 5.81. Found: C, 59.16; H, 5.49.

*O,O-Dimethyl α-(2,3-dimethylphenoxyacetoxy)benzylphosphonate (**I-14**):* yellow liquid; yield, 84%; n_D^{20} 1.5270; IR (KBr, cm^{-1}) ν 3015 (Ar—H), 1772 (C═O), 1264 (Ar—O—C), 1177 (C—O—C), 1032 (P—O—C), 770 (P—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.20−2.27 (d, 6H, 2 × CH_3), 3.63−3.70 (dd, 6H, 2 × OCH_3), 4.75 (d, 1H^a, OCH_2CO), 4.77 (d, 1H^b, OCH_2CO), 6.25−6.28 (d, 1H, CHP), 6.54−7.46 (m, 8H, $C_6H_3 + C_6H_5$). Anal. Calcd for $C_{19}H_{23}O_6P$: C, 60.31; H, 6.13. Found: C, 60.38; H, 6.29.

*O,O-Dimethyl α-(2-nitrophenoxyacetoxy)benzylphosphonate (**I-19**):* light yellow solid; yield, 61%; mp 157−158 °C; IR (KBr, cm^{-1}) ν 3072 (Ar—H), 2961, 2858 (C—H), 1770 (C═O), 1591, 1448 (ArC—C), 1173 (C—O), 1048, 922 (P—O—C), 748 (P—C), 1267 (P═O), 1507, 1346 (ArC—NO_2); ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.58−3.66 (dd, 6H, 2 × OCH_3), 4.82 (s, 2H, OCH_2CO), 6.19 (d, 1H, CHP), 6.84−8.16 (m, 9H, $C_6H_4 + C_6H_5$). Anal. Calcd for $C_{17}H_{18}O_8NP$: C, 51.65; H, 4.56; N, 3.54. Found: C, 51.60; H, 4.57; N, 3.47.

*O,O-Dimethyl α-(3-methyl-4-chlorophenoxyacetoxy)-3-nitrobenzylphosphonate (**I-21**):* colorless solid; yield, 71%; mp 72.0−73.2 °C; IR (KBr, cm^{-1}) ν 3071 (Ar—H), 2957, 2856 (C—H), 1766 (C═O), 1530, 1485 (ArC—C), 1087 (C—O—C), 1033 (P—O—C), 724 (P—C), 1161 (Ar—O—C), 1272 (P═O), 1048 (ArC—Cl), 1530, 1352 (ArC—NO_2); ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.43 (s, 3H, CH_3), 3.65−3.68 (m, 6H, 2 × OCH_3), 4.71 (s, 2H, OCH_2CO), 6.25 (d, 1H, CHP), 6.58−8.23 (m, 7H, $C_6H_4 + C_6H_3$); EI-MS m/z (%) 443 (M^+, 48.55), 290 (33.44), 244 (98.01), 199 (62.38), 174 (68.29), 109 (100), 93 (92.39), 77 (43.73). Anal. Calcd for $C_{18}H_{19}O_8ClNP$: C, 48.72; H, 4.32; N, 3.16. Found: C, 48.67; H, 4.14; N, 2.96.

*O,O-Dimethyl α-(5-methyl-2-chlorophenoxyacetoxy)-3-nitrobenzylphosphonate (**I-22**):* light yellow solid; yield, 69%; mp 109.7−109.9 °C; IR (KBr, cm^{-1}) ν 3087 (Ar—H), 2957, 2885 (C—H), 1776 (C═O), 1599, 1495 (ArC—C), 1094 (C—O—C), 1040 (P—O—C), 728 (P—C), 1172 (Ar—O—C), 1268 (P═O), 1057 (ArC—Cl), 1530, 1351 (ArC—NO_2); ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.56 (s, 3H, CH_3), 3.72−3.77 (m, 6H, 2 × OCH_3), 4.86 (s, 2H, OCH_2CO), 6.33 (d, 1H, CHP), 6.63−8.30 (m, 7H, $C_6H_4 + C_6H_3$); EI-MS m/z (%) 443 (M^+, 41.79), 333 (41.05), 261 (13.07), 229 (86.81), 199 (95.60), 109 (78.41), 93 (100), 77 (72.39). Anal. Calcd for $C_{18}H_{19}O_8ClNP$: C, 48.72; H, 4.32; N, 3.16. Found: C, 49.37; H, 4.23; N, 3.16.

*O,O-Dimethyl α-(2-methyl-4-chlorophenoxyacetoxy)-3-nitrobenzylphosphonate (**I-23**):* light yellow solid; yield, 61%; mp 132.2−133.1 °C; IR (KBr, cm^{-1}) ν 3075 (Ar—H), 2958, 2856 (C—H), 1759 (C═O), 1598, 1492 (ArC—C), 1095 (C—O—C), 1036 (P—O—C), 732 (P—C), 1186 (Ar—O—C), 1265 (P═O), 1138, 384 (ArC—Cl), 1533, 1353 (ArC—NO_2); ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.27 (s, 3H, CH_3), 3.71−3.79 (m, 6H, 2 × OCH_3), 4.81 (s, 2H, OCH_2CO), 6.32 (d, 1H, CHP), 6.59−8.29 (m, 7H, $C_6H_4 + C_6H_3$); EI-MS m/z (%) 443 (M^+, 51.09), 260 (6.18), 245 (85.69), 155 (79.07), 125 (86.24), 109 (92.30), 93 (100), 77 (70.47). Anal. Calcd for $C_{18}H_{19}O_8ClNP$: C, 48.72; H, 4.32; N, 3.16. Found: C, 48.69; H, 4.16; N, 2.93.

*O,O-Dimethyl α-(2,3-dichlorophenoxyacetoxy)-3-nitrobenzylphosphonate (**I-24**):* colorless solid; yield, 68%; mp 111.7−113.1 °C; IR (KBr, cm^{-1}) ν 3081 (Ar—H), 2959, 2852 (C—H), 1745 (C═O), 1581, 1459 (ArC—C), 1084 (C—O—C), 1053 (P—O—C), 777 (P—C), 1178 (—C), 1264 (P═O), 1027 (ArC—Cl), 1534, 1353 (ArC—NO_2); ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.72−3.76 (m, 6H, 2 × OCH_3), 4.90 (s, 2H, OCH_2CO), 6.33 (d, 1H, CHP), 6.74−8.29 (m, 7H, $C_6H_4 + C_6H_3$); EI-MS m/z (%) 465 ($M^+ + 1$, 30.88), 464 (M^+, 21.78), 260 (24.84), 199 (3.27), 175 (76.09), 109 (79.63), 93 (100), 777 (29). Anal. Calcd for $C_{17}H_{16}O_8Cl_2NP$: C, 43.99; H, 3.47; N, 3.02. Found: C, 43.57; H, 3.17; N, 2.95.

*O,O-Dimethyl α-(2,6-dichlorophenoxyacetoxy)-3-nitrobenzylphosphonate (**I-25**):* colorless solid; yield, 75%; mp 86.2−87.6 °C; IR (KBr, cm^{-1}) ν 3087 (Ar—H), 2952, 2863 (C—H), 1753 (C═O), 1562, 1456 (ArC—C), 1076 (C—O—C), 1033 (P—O—C), 718 (P—C), 1219 (Ar—O—C), 1252 (P═O), 1013 (ArC—Cl), 1529, 1358 (ArC—NO_2); ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.78−3.82 (m, 6H, 2 × OCH_3), 4.81 (s, 2H, OCH_2CO), 6.40 (d, 1H, CHP), 7.04−8.37 (m, 7H, $C_6H_4 + C_6H_3$); EI-MS m/z (%) 556 ($M^+ + 93$, 4.36), 464 (M^+, 12.76), 260 (50.64), 175 (55.18), 109 (85.08), 93 (100), 77 (33.71). Anal. Calcd for $C_{17}H_{16}O_8Cl_2NP$: C, 43.99; H, 3.47; N, 3.02. Found: C, 44.33; H, 3.22; N, 2.94.

*O,O-Dimethyl α-(4-chlorophenoxyacetoxy)-3-nitrobenzylphosphonate (**I-26**):* light yellow solid; yield, 80%; mp 79.8−83.9 °C; IR (KBr, cm^{-1}) ν 3072 (Ar—H), 2959, 2857 (C—H), 1769 (C═O), 1595, 1492 (ArC—C), 1172 (C—O—C), 1037 (P—O—C), 733 (P—C), 1233 (Ar—O—C), 1223 (P═O), 1095 (ArC—Cl), 1534, 1354 (ArC—NO_2); ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.72−3.82 (m, 6H, 2 × OCH_3), 4.79 (s, 2H, OCH_2CO), 6.34 (d, 1H, CHP), 6.81−8.29 (m, 8H, $C_6H_4 + C_6H_4$); EI-MS m/z (%) 429 (M^+, 80.71), 262 (46.91), 244 (91.98), 186 (98.14), 166 (70.08), 141 (100), 109 (99.22), 75 (99.47). Anal. Calcd for $C_{17}H_{17}O_8ClNP$: C, 47.51; H, 3.99; N, 3.26. Found: C,47.01; H, 3.97; N, 3.02.

*O,O-Dimethyl α-(2,4-dichlorophenoxyacetoxy)-3-bromobenzylphosphonate (**I-33**):* yellow liquid; yield 53%; n_D^{20} 1.4529; IR (KBr, cm^{-1}) ν 3028 (Ar—H), 1750 (C═O), 1580, 1480 (C═C), 1225 (P═O), 1050 (P—O), 800 (Ar—Cl),748 (P−C); ^{1}H NMR (200 MHz, $CDCl_3$) δ 3.7−3.8 (d, 6H, 2 × OCH_3), 4.75 (s, 2H, OCH_2CO), 6.20 (d, 1H, CHP), 6.7−7.5 (m, 7H, $C_6H_3 + C_6H_4$). Anal. Calcd for $C_{17}H_{16}BrCl_2O_6P$: C, 40.99; H, 3.24. Found: C, 41.14; H, 3.25.

*O,O-Dimethyl α-(4-chlorophenoxyacetoxy)-2-hydroxybenzylphosphonate (**I-34**):* yellow liquid; yield, 78%; n_D^{20} 1.4955; IR (KBr, cm^{-1}) ν 3473 (O—H), 2957 (R—H), 1774 (C═O), 1598 (Ar C—C), 1051 (C—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.49−3.86 (dd, 6H, 2 × OCH_3), 4.64 (d, 1H^a, OCH_2CO), 4.74 (d, 1H^b, OCH_2CO), 4.96 (s, 1H, OH), 6.48−6.51 (d, 1H, CHP), 6.78−7.59 (m, 8H, $C_6H_4 + C_6H_4$). Anal. Calcd for $C_{17}H_{18}ClO_7P$: C, 50.95; H, 4.53. Found: C, 50.46; H, 4.82.

General Procedure for the Preparation of *O,O*-Dialkyl α-(Fluorophenoxyacetoxy)alkylphosphonates I-6−I-8, I-15−I-17, I-27−I-29, and I-35. A solution of fluorophenoxyacetyl chloride **6** (0.011 mol) in trichloromethane (15 mL) was added to a stirred mixture of α-hydroxyalkylphosphonate **2** (0.01 mol) and triethylamine (0.011 mol) in trichloromethane (15 mL) at 2−4 °C. The resultant mixture was stirred at ambient temperature for 2−3 h, then washed with 0.1 M HCl, saturated $NaHCO_3$, and brine successively, dried, and concentrated. The residue was chromatographed on silica with 20% acetone in petroleum ether as eluent to give the compounds **I-6**−**I-8**, **I-15**−**I-17**, **I-27**−**I-29**, and **I-35**. Their structures were confirmed by IR, MS, and ^{1}H NMR spectra and elemental analysis. Physicochemical and spectroscopic data for compounds are as follows.

*O,O-Dimethyl α-(2-fluorophenoxyacetoxy)ethylphosphonate (**I-6**):* colorless crystal; yield, 59%; mp 62−63 °C; IR (KBr, cm^{-1}) ν 3074 (ArC—H), 2965 (R—H), 2840 (R—H), 1769 (C═O), 1614

(ArC—C), 1509 (ArC—C), 736 (P—C), 1275 (P═O), 1186 (C—O—C), 1046 (P—O—C), 1123 (ArC—F); ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.51 (dd, 3H, CH_3), 3.76−3.81 (d, 6H, 2 × OCH_3), 4.77 (s, 2H, OCH_2CO), 5.39−5.43 (m, 1H, CHP), 6.92−7.13 (m, 4H, C_6H_4); EI-MS *m/z* (%) 306 (M^+, 17.51), 138 (100), 125 (59.61), 109 (73.03), 93 (48.23), 79 (26.37). Anal. Calcd for $C_{12}H_{16}FO_6P$: C, 47.07; H, 5.27. Found: C, 47.27; H, 5.07.

*O,O-Dimethyl α-(3-chloro-4-fluorophenoxyacetoxy)ethylphosphonate (**I-7**):* yellowish liquid; yield, 82%; n_D^{20} 1.4894; IR (KBr, cm^{-1}) ν 3088 (ArC—H), 2989 (R—H), 2856 (R—H), 1768 (C═O), 1597 (ArC—C), 763 (P—C), 1277 (P═O), 1187 (C—O—C), 1050 (P—O—C), 1124 (ArC—F), 690 (ArC—Cl); ^{1}H NMR ($CDCl_3$) δ 1.50 (m, 3H, CH_3), 3.84−3.75 (dd, 6H, 2 × OCH_3), 4.67 (s, 2H, OCH_2CO), 5.43−5.39 (m, 1H, CHP), 7.10−6.77 (m, 3H, C_6H_3); EI-MS *m/z* (%) 340 (M^+, 39.48), 195 (36.53), 138 (84.96), 109 (100), 93 (60.59), 79 (46.94). Anal. Calcd for $C_{12}H_{15}ClFO_6P$: C, 42.31; H, 4.44. Found: C, 42.33; H, 4.74.

*O,O-Dimethyl 1-(3-chloro-4-fluorophenoxyacetoxy)-2,2,2-trichloroethylphosphonate (**I-8**):* yellow liquid; yield, 80%; n_D^{20} 1.5152; IR (KBr, cm^{-1}) ν 3075 (Ar—H), 2961, 2857 (R—H), 1785 (C═O), 1598 (C═C), 1278 (P═O), 1158 (C—O—C), 1110 (Ar—F), 1048 (P—O—C), 734 (P—C), 689 (Ar—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.87−3.92 (m, 6H, 2 × OCH_3), 4.84 (s, 2H, OCH_2CO), 5.98 (d, 1H, CHP), 6.82−7.29 (m, 3H, C_6H_3); EI-MS *m/z* (%) 442 (M^+, 41.53), 205 (77.24), 139 (24.89), 109 (100), 93 (93.28), 79 (72.63). Anal. Calcd for $C_{12}H_{12}Cl_4FO_6P$: C, 32.46; H, 2.72. Found: C, 32.59; H, 2.54.

*O,O-Dimethyl α-(2,4-difluorophenoxyacetoxy)benzylphosphonate (**I-15**):* colorless solid; yield, 73%; mp 64.5−65.4 °C; IR (KBr, cm^{-1}) ν 3069 (Ar—H), 2950, 2852 (C—H), 1765 (C═O), 1603, 1456 (ArC—C), 1263 (P═O), 1113 (C—O—C), 1027 (P—O—C), 1188 (Ar—O—C), 772 (P—C), 1194 (Ar—F); ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.63−3.72 (m, 6H, 2 × OCH_3), 4.78 (s, 2H, OCH_2CO), 6.25 (d, 1H, CHP), 6.75−7.47 (m, 8H, C_6H_5 + C_6H_3); EI-MS *m/z* (%) 479 (M^+ + 93, 7.27), 386 (M^+, 59.50), 215 (57.79), 199 (67.39), 109 (61.94), 93 (100), 77 (52.70). Anal. Calcd for $C_{17}H_{17}O_6F_2P$: C, 52.86; H, 4.44. Found: C, 53.33; H, 4.29.

*O,O-Dimethyl α-(3-chloro-4-fluorophenoxyacetoxy)benzylphosphonate (**I-16**):* colorless solid; yield, 80%; mp 74.5−75.1 °C; IR (KBr, cm^{-1}) ν 3059 (Ar—H), 2962, 2852 (R—H), 1772 (C═O), 1602, 1455 (C═C), 1266 (P═O), 1088 (C—O—C), 1026 (P—O—C), 783 (P—C); ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.63−3.73 (m, 6H, 2 × OCH_3), 4.72 (s, 2H, OCH_2CO), 6.25 (d, 1H, CHP), 6.75−7.46 (m, 8H, C_6H_5 + C_6H_3); EI-MS *m/z* (%) 402 (M^+, 36.03), 215 (42.56), 199 (98.14), 159 (65.03), 129 (49.3), 109 (49.2), 93 (100), 77 (49.5). Anal. Calcd for $C_{17}H_{17}O_6ClFP$: C, 50.70; H, 4.25. Found: C, 51.31; H, 4.25.

*O,O-Dimethyl α-(2-chloro-4-fluorophenoxyacetoxy)benzylphosphonate (**I-17**):* colorless solid; yield, 69%; mp 86.8−87.1 °C; IR (KBr, cm^{-1}) ν 3054 (Ar—H), 2960, 2859 (R—H), 1773 (C═O), 1597, 1442 (C═C), 1260 (P═O), 1086 (C—O—C), 1086 (P—O—C), 742 (P—C); ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.63−3.72 (m, 6H, 2 × OCH_3), 4.79 (s, 2H, OCH_2CO), 6.25 (d, 1H, CHP), 6.80−7.46 (m, 8H, C_6H_5 + C_6H_3); EI-MS *m/z* (%) 402 (M^+, 17.52), 238 (26.13), 215 (18.87), 159 (66.68), 109 (48.43), 93 (100), 77 (31.60). Anal. Calcd for $C_{17}H_{17}O_6ClFP$: C, 50.70; H, 4.25. Found: C, 51.20; H, 4.01.

*O,O-Dimethyl α-(2,4-difluorophenoxyacetoxy)-3-nitrobenzyl phosphonate (**I-27**):* light yellow liquid; yield, 75%; n_D^{20} 1.6010; IR (KBr, cm^{-1}) ν 3091 (Ar—H), 2959, 2856 (C—H), 1756 (C═O), 1586, 1461 (ArC—C), 1278 (P═O), 1068 (C—O—C), 1039 (P—O—C), 1188 (Ar—O—C), 732 (P—C), 1285 (Ar—F), 1533, 1354 (ArC—NO_2); ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.74−3.81 (m, 6H, 2 × OCH_3), 4.84 (s, 2H, OCH_2CO), 6.33 (d, 1H, CHP), 6.78−8.30 (m, 7H, C_6H_4 + C_6H_3); EI-MS *m/z* (%) 431 (M^+, 0.05), 261 (6.62), 244 (34.61), 188 (44.62), 123 (5054), 109 (13.57), 93 (100), 77 (61.63). Anal. Calcd for $C_{17}H_{16}O_8F_2NP$: C, 47.34; H, 3.74; N, 3.25. Found: C, 47.36; H, 3.58; N, 3.12.

*O,O-Dimethyl α-(3-chloro-4-fluorophenoxyacetoxy)-3-nitrobenzylphosphonate (**I-28**):* light yellow solid; yield, 75%; mp 74.9−75.3 °C; IR (KBr, cm^{-1}) ν 3093 (Ar—H), 2958, 2856 (C—H), 1775 (C═O), 1595, 1444 (ArC—C), 1082 (C—O—C), 1036 (P—O—C), 733 (P—C), 1169 (Ar—O—C), 1266 (P═O), 1224 (ArC—Cl), 684 (ArC—F), 1533, 1353 (ArC—NO_2); ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.73−3.78 (m, 6H, 2 × OCH_3), 4.79 (s, 2H, OCH_2CO), 6.34 (d, 1H, CHP), 6.84−8.30 (m, 7H, C_6H_4 + C_6H_3); EI-MS *m/z* (%) 449 (M^+ + 1, 5.02), 448 (M^+, 10.52), 261 (24.56), 129 (36.42), 109 (100), 93 (86.65), 77 (54.23). Anal. Calcd for $C_{17}H_{16}O_8ClFNP$: C, 45.60; H, 3.60; N, 3.13. Found: C, 46.88; H, 3.76; N, 3.18.

*O,O-Dimethyl α-(2-chloro-4-fluorophenoxyacetoxy)-3-nitrobenzylphosphonate (**I-29**):* colorless solid; yield, 70%; mp 111.2−112.0 °C; IR (KBr, cm^{-1}) ν 3080 (Ar—H), 2966, 2859 (C—H), 1756 (C═O), 1601, 1491 (ArC—C), 1080 (C—O—C), 1058 (P—O—C), 747 (P—C), 1191 (Ar—O—C), 1259 (P═O), 1137 (ArC—Cl), 1302 (ArC—F), 1529, 1353 (ArC—NO_2); ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.74−3.76 (m, 6H, 2 × OCH_3), 4.85 (s, 2H, OCH_2CO), 6.33 (d, 1H, CHP), 6.83−8.30 (m, 7H, C_6H_4 + C_6H_3); EI-MS *m/z* (%) 449 (M^+ + 1, 3.97), 447 (M^+ − 1, 10.23), 245 (9.97), 159 (25.24), 129 (36.8), 109 (100), 93 (31.47), 77 (15). Anal. Calcd for $C_{17}H_{16}O_8ClFNP$: C, 45.60; H, 3.60; N, 3.13. Found: C, 46.05; H, 3.34; N, 3.03.

*O,O-Dimethyl α-(2-chloro-4-chlorophenoxyacetoxy)-2-hydroxybenzylphosphonate (**I-35**):* yellow liquid; yield, 55%; n_D^{20} 1.4885; IR (KBr, cm^{-1}) ν 3467 (O—H), 3076 (Ar—H), 2959 (R—H), 1770 (C═O), 1602 (ArC—C), 1175 (C—F), 1032 (C—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.60−3.72 (dd, 6H, 2 × OCH_3), 4.75 (d, 1H^a, OCH_2CO), 4.80 (d, 1H^b, OCH_2CO), 5.02 (s, 1H, OH), 6.50−6.53 (d, 1H, CHP), 6.72−7.85 (m, 7H, C_6H_4 + C_6H_3). Anal. Calcd for $C_{17}H_{17}ClFO_7P$: C, 48.76; H, 4.09. Found: C, 48.94; H, 3.66.

General Procedure for the Preparation of *O,O*-Dialkyl α-(Trifluoromethylphenoxyacetoxy)alkylphosphonates I-18, I-20, and I-30−I-32. A solution of trifluoromethylphenoxyacetyl chloride **6** (1.19 g, 0.005 mol) in trichloromethane (10 mL) was added to a stirred mixture of α-hydroxyalkylphosphonates **2** (0.006 mol) and triethylamine (0.606 g, 0.006 mol) in trichloromethane (25 mL) at 2−4 °C. The resultant mixture was stirred at ambient temperature for 3−5 h and then stirred at 35−36 °C for 1−2 h. The trichloromethane solution was washed with 0.1 M HCl, saturated $NaHCO_3$, and brine successively, dried, and concentrated. The residue was purified by column chromatography on silica gel and eluted with petroleum ether/acetone = 2:1 (v/v) to give the corresponding pure compounds **I-18**, **I-20**, and **I-30**−**I-32**.

The structures of compounds **I-18**, **I-20**, and **I-30**−**I-32** were confirmed by IR, MS, and ^{1}H NMR spectra and elemental analysis. Physicochemical and spectroscopic data for compounds **I-18**, **I-20**, and **I-30**−**I-32** are as follows.

*O,O-Dimethyl α-(4-trifluoromethylphenoxyacetoxy)benzylphosphonate (**I-18**):* colorless solid; yield, 76%; mp 103.6−104.8 °C; IR (KBr, cm^{-1}) ν 3069 (Ar—H), 2963, 2860 (C—H), 1781 (C═O), 1591, 1456 (ArC—C), 1250 (P═O), 1092 (C—O—C), 1064 (P—O—C), 1175 (Ar—O—C), 747 (P—C), 1334 (ArC—F); ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.62−3.72 (m, 6H, 2 × OCH_3), 4.81 (s, 2H, OCH_2CO), 6.25 (d, 1H, CHP), 6.94−7.55 (m, 9H, C_6H_5 + C_6H_4); EI-MS *m/z* (%) 418 (M^+, 15.42), 216 (30.12), 125 (80.36), 109 (56.87), 93 (100), 77 (45.63). Anal. Calcd for $C_{18}H_{18}O_6F_3P$: C, 51.68; H, 4.34. Found: C, 51.18; H, 4.08.

*O,O-Dimethyl α-(2-nitro-4-trifluoromethylphenoxyacetoxy)benzylphosphonate (**I-20**):* light yellow solid; yield, 73%; mp 134−135 °C; IR (KBr, cm^{-1}) ν 3070 (Ar—H), 2947 (R—H), 2857 (R—H), 1763 (C═O), 1630 (ArC—C), 1483 (ArC—C), 833 (P—C), 1230 (Ar—O—C), 1264 (P═O), 1194 (C—O—C), 1033 (P—O—C), 1506 (NO_2),

1335 (Ar—CF_3); ^{1}H NMR (200 MHz, $CDCl_3$) δ 3.59−3.72 (dd, 6H, 2 × OCH_3), 4.96 (s, 2H, OCH_2), 6.17−6.23 (d, 1H, CHP), 6.97−8.11 (m, 8H, C_6H_3 + C_6H_5). Anal. Calcd for $C_{18}H_{17}F_3NO_8P$: C, 46.66; H, 3.70; N, 3.02. Found: C, 46.69; H, 3.79; N, 2.70.

*O,O-Dimethyl α-(3-trifluoromethylphenoxyacetoxy)-3-nitrobenzylphosphonate (**I-30**):* yellow liquid; yield, 71%; n_D^{20}, 1.5233; IR (KBr, cm^{-1}) ν 3080 (Ar—H), 2954, 2853 (C—H), 1764 (C═O), 1595, 1492 (ArC—C), 826 (P—C), 1228, 1101 (Ar—O—C), 1267 (P═O), 1171 (C—O—C), 1050, 907 (P—O—C), 1525, 1354 (ArC—NO_2), 1330 (ArC—CF_3); ^{1}H NMR (200 MHz, $CDCl_3$) δ 3.60−3.84 (dd, 6H, 2 × OCH_3), 4.80 (s, 2H, OCH_2), 6.20−6.40 (d, 1H, CHP), 6.92−7.76 (m, 8H, C_6H_4 + C_6H_4). Anal. Calcd for $C_{18}H_{17}F_3NO_8P$: C, 46.65; H, 3.67; N, 3.02. Found: C, 46.48; H, 3.19; N, 2.50.

*O,O-Dimethyl α-(4-trifluoromethylphenoxyacetoxy)-3-nitrobenzylphosphonate (**I-31**):* light yellow liquid; yield, 74%; n_D^{20} 1.4985; IR (KBr, cm^{-1}) ν 3084 (Ar—H), 2961, 2857 (C—H), 1770 (C═O), 1615, 1445 (ArC—C), 1264 (P═O), 1090 (C—O—C), 1040 (P—O—C), 1163 (Ar—O—C), 733 (P—C), 1342 (ArC—F), 1533, 1329 (ArC—NO_2); ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.73−3.75 (m, 6H, 2 × OCH_3), 4.70 (s, 2H, OCH_2CO), 6.35 (d, 1H, PCHO), 6.98−8.30 (m, 8H, C_6H_4 + C_6H_4); EI-MS *m/z* (%) 463 (M^+, 10.42), 444 (46.03), 260 (8.21), 245 (57.79), 145 (96.22), 109 (71.42), 93 (100), 77 (46.41). Anal. Calcd for $C_{18}H_{17}O_8F_3NP$: C, 46.66; H, 3.70; N, 3.02. Found: C, 46.73; H, 3.69; N, 2.97.

*O,O-Dimethyl α-(2-nitro-4-trifluoromethylphenoxyacetoxy)-3-nitrobenzylphosphonate (**I-32**):* light yellow solid; yield, 70%; mp 107−108 °C; IR (KBr, cm^{-1}) ν 3087 (Ar—H), 2958 (R—H), 2857 (R—H), 1770 (C═O), 1627 (ArC—C), 1463 (ArC—C), 832 (P—C), 1228 (Ar—O—C), 1083 (Ar—O—C), 1267 (P═O), 1166 (C—O—C), 1043 (P—O—C), 1534 (NO_2), 1326 (Ar—CF_3); ^{1}H NMR (200 MHz, $CDCl_3$) δ 3.71−3.76 (dd, 6H, 2 × OCH_3), 5.02 (s, 2H, OCH_2), 6.27−6.33 (d, 1H, CHP), 7.05−8.23 (m, 8H, C_6H_3 + C_6H_5). Anal. Calcd for $C_{18}H_{16}F_3N_2O_{10}P$: C, 42.53; H, 3.17; N, 5.51. Found: C, 43.00; H, 3.14; N, 4.91.

Herbicide Activity Assay. *Test in Laboratory Greenhouse.* The pre-emergence and postemergence herbicidal activities of title compounds were evaluated in a set of experiments in a greenhouse. Barnyard grass (*Echinochloa crusgalli*), ascendant crabgrass (*Digitaria sanguinalis*), amaranth pigweed (*Amaranthus retroflexus*), carrot (*Daucus carota*), clover (*Medicago sativa*), cucumber (*Cucumis sativus*), chingma abutilon (*Abutilon theophrasti*), spiny amaranth (*Amaranthus spinosus*), lambsquarters (*Chenopodium album*), giant foxtail (*Setaria viridis*), white eclipta (*Eclipta prostrata*), Siberia cocklebur (*Xanthium strumarium*), Pennsylvania bittercress (*Cardamine hirsuta*), common purslane (*Portulaca oleracea*), rape (*Brassica campestris*), rice (*Oryza sativa*), wheat (*Triticum aestivum*), and maize (*Zea mays*) were chosen as test plants.

Plastic pots were packed with sandy clay loam soil, and water was added up to 3 cm in depth. For the postemergence study, about 15−20 seeds of plant were sown in the soil at a depth of 5 mm and grown at 20−25 °C for a few days. At pre-emergence and postemergence, the diluted formulation of each compound containing acetone and Tween 80 was applied into the pots at 18.75−1500 ai g/ha. Twenty days later, the pre-emergence herbicidal activity against each weed was visually evaluated.

At postemergence, the solution of the chemicals tested was applied to the foliage of plants grown at 2−3-leaf stage with a sprayer at the test rate with a spilling volume of 1000 L/ha.

All of the treatments were replicated three times in a completely randomized design. The test plants were harvested 20 days after sowing and determined for fresh weight. The postemergence herbicidal activity against each weed was evaluated. The percentage growth inhibition of roots and aerial parts was calculated in relation to the mass of the roots and aerial parts of the control, respectively.

Inhibitory Effect (%). The inhibitory effect of compounds on the growth of plants at a dose was measured as percentage change in each plant weight compared to that of the control, as 0% (no effect or not significantly different from control), 100% (completely killed). According to the corresponding inhibition percentage, the inhibitory effect was expressed as a four scale: A, 90−100%; B, 75−89%; C, 50−74%; D, ≤50%.

Determination of IC_{50} Values (Inhibition on the Growth of Cucumis sativa). The IC_{50} values of the title compounds were determined as the toxicity of inhibition to root growth of *Cucumis sativa* by the following method.

A defined amount of test compound dissolved in acetone was poured on a filter paper in Petri dishes (9 cm), and 10 cucumber (*Cucumis sativa*) seeds after soaking in water for 6 h were placed on the filter paper. The Petri dishes with cucumber seeds were placed in a Lighting Incubator (LRH-250-G) at 28 °C for 3 days with 10 h of lighting and 14 h in the dark. After 3 days of treatment, the inhibition percentage was calculated by the corresponding control using the length of the taproot as indicator. Three replications per concentration were performed. According to the average percentage of inhibition of cucumber root at five concentrations for each test compound (a different IC_{50} value was determined at different range of five concentrations), the IC_{50} values were estimated by regression analysis using the logarithm of concentration and probit of corresponding inhibition percentage; the detailed data can be found in the Supporting Information.

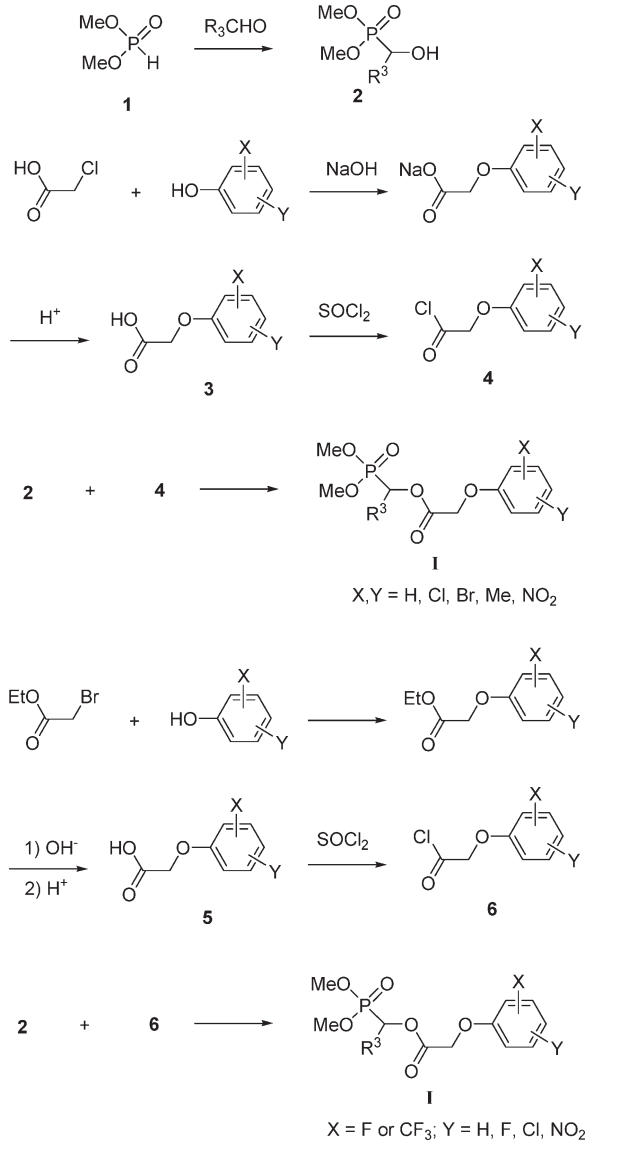
Enzyme Assays. The activity of PDHc could be determined by a sensitive spectrophotometric assay according to refs 1 and 25−27. In this procedure, PDHc could be conveniently assayed by measuring the rate of appearance of product reduced nicotinamide adenine dinucleotide (NADH), which absorbs ultraviolet (UV) at 340 nm.[28] If the reaction is prevented by an inhibitor, there will be a corresponding decrease in absorbance compared with control. Thus, the PDHc activity was determined by measuring the formation of NADH.

Preparation of Pisum sativum (Pea) Mitochondria. The preparation of pea mitochondria was carried out according to the procedure of Reid et al.[29] *P. sativum* (pea) seeds were soaked in water overnight and grown at room temperature in the dark until the shoots were 20−30 cm in height. The shoots were cut into small pieces (about 10 mm) and frozen for 1 h in a refrigerator. Then the shoots were ground, using a mortar and pestle, in three times volume of 50 mM Tris-HCl buffer (pH 7.4) containing 0.25 M glucose, 3 mM EDTA, and 0.75 mM BSA. The homogenate was filtered through several layers of cheesecloth and centrifuged at 10000*g* for 15 min. The supernatant was centrifuged at 27000*g* for 45 min. The resulting pellet was resuspended in the grinding medium and centrifuged at 27000*g* for 45 min. The surface of the pellet was washed with ice water and then resuspended in acetone or ethanol at −20 °C, centrifuged at 27000*g* for 20 min, and resuspended and centrifuged an additional three times in cold acetone. The final pellet was dried with a stream of air and stored at −20 °C.

Preparation of Enzyme Sample. The powder of mitochondria in acetone was resuspended in 25 mM Tes buffer (pH 7.4) containing 200 μM thiamin pyrophosphate (TPP), 5 mM dithiothreitol (DTT), and 2 mM $MgSO_4$ at a final concentration of 20 mg/mL and ground thoroughly in glass homogenizer. The homogenate was centrifuged at 27000*g* for 15−20 min. The supernatant was used as an enzyme solution.

Inhibition of PDHc from Pisum sativum (Pea). Six samples of enzyme solution (0.5 mL) were taken. One of them was added to water as control, and the rest were mixed with test compounds at final concentrations of 10^{-3}, 10^{-4}, 10^{-5}, 10^{-6}, and 10^{-7} g/mL separately. The mixtures were incubated at 25 °C for 25 min, and they were added respectively at various times to 1 mL of reaction mixture containing 25 mM Tes (pH 7.4), 1 mM $MgCl_2$, 1 mM cysteine, 1 mM NAD^+ (oxidized nicotinamide adenine dinucleotide), 0.13 mM CoA (Na salt), and 4 mM TPP, and then 0.3 mL (1 mM) sodium pyruvate as a substrate was added to start the reaction in a total volume of 3 mL. After 1 min at

Scheme 2. Synthetic Route of *O,O*-Dimethyl α-(Substituted phenoxyacetoxy)alkylphosphonates I-1—I-35



25 °C for temperature equilibration, the rate of formation of NADH was continuously monitored at 340 nm by a spectrophotometer. Three or five replications per concentration were performed and averaged, and the inhibitor concentration giving 50% inhibition (IC_{50}) was calculated.

Assays of PDHc from mung bean and rice were also carried out according to the same method as that used for peas.

Kinetic Experiment of PDHc from Phaseolus radiatus (Mung Beans). The maximum velocity V_{max} and Michaelis constant K_m were determined by measuring the enzyme catalytic effect of variations in the concentration of sodium pyruvate as a substrate in the presence of a fixed concentration of inhibitor.

In this kinetic experiment, the concentration of inhibitor **I-5** was kept in 0.028 mM (10^{-5} g/mL); V_{max} and K_m were determined experimentally by measuring V at different substrate concentrations in 0.1, 0.2, 0.3, 0.4, and 0.5 mM of sodium pyruvate separately. Under the same conditions, this set of experiments was also done in the absence of inhibitor **I-5**. Detailed data for the enzyme kinetic experiment of **I-5** against PDHc can be found in the Supporting Information.

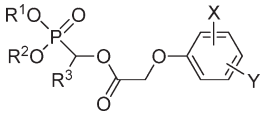
RESULTS AND DISCUSSION

Synthesis. Two synthetic routes could be used to obtain *O,O*-dialkyl α-(substituted phenoxyacetoxy)alkylphosphonates. They can be synthesized by the reaction of sodium substituted phenoxide with corresponding α-(chloroacetoxy)alkyl phosphonates in the presence of sodium iodide (route 1) or by the condensation of *O,O*-dialkyl α-hydroxyalkyl phosphonates and substituted phenoxyacetyl chlorides (route 2). Generally, the yield of title compounds using route 2 was better than that of route 1. Therefore, we chose synthetic route 2 to obtain new *O,O*-dimethyl α-(substituted phenoxyacetoxy)alkylphosphonates **I-1**—**I-35** (Scheme 2). The target phosphonate derivatives **I** contain a carboxylic ester group, which is sensitive to acid or base; the reaction required a temperature near room temperature in anhydrous solvents. We performed the reaction in two steps: first, the reaction was carried out at room temperature for several hours and then at higher temperature under mild reaction conditions. **I-1**—**I-5**, **I-9**—**I-14**, **I-19**, **I-21**—**I-26**, **I-33**, and **I-34** were prepared by condensation of *O,O*-dimethyl α-hydroxyalkylphosphonates with substituted phenoxyacetyl chlorides in the presence of pyridine as a base. Considering that *O,O*-dimethyl α-hydroxyalkylphosphonates are easily regenerated to the starting carbonyl compounds in strong alkaline medium,[30] pyridine, a weak base, was chosen for this reaction. *O,O*-Dimethyl α-(fluoro-substituted phenoxyacetoxy)alkylphosphonates **I-6**—**I-8**, **I-15**—**I-17**, **I-27**—**I-29**, and **I-35** and *O,O*-dimethyl α-(trifluoromethyl-substituted phenoxyacetoxy)alkyl phosphonates **I-18**, **I-20**, and **I-30**—**I-32** were obtained by the condensation of *O,O*-dimethyl α-hydroxyalkylphosphonates **2** with the corresponding fluorophenoxyacetyl chloride and trifluoromethyl phenoxyacetyl chloride **6**, respectively.

In this method substituted phenoxyacetic acids **3** were prepared by condensation of corresponding substituted phenols with chloroacetic acid in the presence of alkali such as sodium hydroxide. However, the yield of the reaction of chloroacetic acid with fluoro-substituted phenols or trifluoromethyl-substituted phenols in the presence of sodium hydroxide was very poor because of the strong electron-withdrawing nature of the substituent. Therefore, according to the method of Brayer et al.,[31] fluoro-substituted phenoxyacetic acids and trifluoromethyl-substituted phenoxyacetic acids **5** were prepared in satisfactory yields by the reaction of fluoro-substituted phenols or trifluoromethyl-substituted phenols with ethyl bromoacetate in the presence of K_2CO_3 in DMSO followed by alkaline hydrolysis (Scheme 2).

Herbicidal Activity and Structure—Activity Relationships. The herbicidal activities of title compounds **I** were evaluated at different doses in a set of experiments in a greenhouse. As a preliminary bioassay all compounds were tested at a dose of 1500 ai g/ha for pre-emergence and postemergence herbicidal activity on *Echinochloa crusgalli*, *Digitaria sanguinalis*, *Brassica campestris*, *Amaranthus retroflexus*, and *Medicago sativa*. The results are shown in Tables 1 and 2. By comparison of herbicidal activities among compounds **I** in Tables 1 and 2, substituents X and Y on the benzene ring greatly affected activity when R^1, R^2, R^3, and R^4 were kept the same. The herbicidal activities of most compounds with substituents on the phenoxybenzene ring were higher than those of compounds with no substituent on the phenoxybenzene ring, such as **I-11** and **I-50**, which were almost inactive. The 2,4-Cl_2 or 2-Cl 4-F substitution on the phenoxybenzene ring was the most promotive followed by 2-Me,4-Cl; 4-Cl; and 3-Me,4-Cl. Herbicidal activity could be greatly enhanced by introducing

Table 1. Structures and Herbicidal Activities of *O,O*-Dimethyl α-(Substituted phenoxyacetoxy)alkylphosphonates I[a]



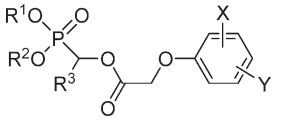
					Ech[b]		*Dig*[b]		*Bra*[b]		*Ama*[b]		*Med*[b]	
no.	R^1R^2	R^3	X	Y	pre	post	pre	post	pre	post	pre	post	pre	post
I-1	Me	Me	2-Me	3-Me	B	C	D	D	A	B	B	C	B	C
I-2	Me	Me	H	4-Br	D	D	D	D	D	B	D	C	D	C
I-3	Me	Me	2-Br	4-Br	D	D	D	D	D	A	D	A	B	A
I-4	Me	Me	2-NO_2	4-Cl	D	D	D	D	D	D	D	D	D	D
I-5	Me	Me	2-Cl	4-Cl	B	C	B	C	A	A	A	A	A	A
I-6	Me	Me	H	2-F	D	D	D	D	D	D	D	D	D	D
I-7	Me	Me	3-Cl	4-F	C	D	C	D	D	B	D	B	B	B
I-8	Me	CCl_3	3-Cl	4-F	C	C	B	D	D	B	C	A	C	C
I-9	Me	CCl_3	2-Cl	4-Cl	A	C	A	C	A	A	A	A	A	C
I-10	Me	Et	2-Cl	4-Cl	B	C	B	C	A	A	A	A	A	A
I-36	Me	Me	H	4-Me	C	C	D	D	B	C	B	C	B	C
I-37	Me	Me	H	4-Cl	A	D	A	D	A	B	A	A	A	C
I-38	Me	Me	2- Me	4-Cl	A	C	A	C	A	A	A	A	A	C
I-39	Me	Me	3- Me	4-Cl	B	D	B	D	A	C	B	C	B	C
I-40	Me	Me	2-Cl	5-Me	D	D	D	C	D	D	D	D	D	D
I-41	Me	Me	2-Cl	3-Cl	D	D	D	D	D	D	D	C	D	D
I-42	Me	Me	H	3-F	D	D	D	D	D	D	D	D	D	D
I-43	Me	Me	H	4-F	D	D	A	D	B	C	C	B	B	C
I-44	Me	Me	2-F	4-F	D	D	B	C	D	B	D	B	C	C
I-45	Me	Me	3-F	5-F	D	D	D	D	D	D	D	D	D	D
I-46	Me	Me	2-Cl	4-F	A	B	A	B	A	A	A	A	A	A
I-47	Me	H	2-Cl	4-Cl	D	D	A	D	A	A	A	A	B	A
I-48	Me	Me	H	3-CF_3	C	D	B	D	B	A	C	D	B	A
I-49	Me	Me	H,	4-CF_3	D	D	D	D	D	D	D	B	D	D
I-50	Me	Me	H	H	D	D	D	D	D	D	D	D	D	D
2,4-D					B	C	A	C	A	A	A	A	A	A

[a] Synthetic methods for compounds: **I-36**, **I-37** (ref 16); **I-47** (ref 18);**I-41**, **I-44**, **I-45** (ref 19; **I-46**, **I-49**, **I-54** (ref 20); **I-42**, **I-48**, **I-50** (ref 21); **I-38**, **I-39**, **I-40**, **I-43** (ref 22). [b] Inhibitory potency (%) on the growth of plants at a dose of 1500 ai g/ha in greenhouse was expressed as a four scale: A, 90−100%; B, 75−89%; C, 50−74%; D, ≤50%. *Ech*, *Echinochloa crusgalli*; *Dig*, *Digitaria sanguinalis*; *Bra*, *Brassica campestris*; *Ama*, *Amaranthus retroflexus*; *Med*, *Medicago sativa*.

2,4-Cl_2 or 2-Cl 4-F on the phenoxybenzene ring. However, it is very interesting that the introduction of 2,4-F_2 or 2-F,4-Cl as X or Y resulted in a sharp decrease in herbicidal activity. The compounds with 2,4-Cl_2 or 2-Cl 4-F as X or Y (such as **I-5**, **I-10**, **I-33**, **I-35**, and **I-46**) irrespective of the difference of substituents in the R^3 moiety, exhibited notable herbicidal activity against dicotyledons for pre-emergence and postemergence at a dose of 1500 ai g/ha. The effects of the introduction of other substituents as X and Y on herbicidal activity was not beneficial compared with that of 2,4-Cl_2 or 2-Cl 4-F; that is, the herbicidal activities of compounds with 2-F; 3-F; 2-F,3-F; 3-F, 5-F; 2-Cl,5-Me; 2-Cl,3-Cl; 2-Cl,6-Cl; 4-CF_3; 2-NO_2; 4-NO_2; 2-NO_2,4-Cl; or 2-NO_2,4-CF_3 as X and Y on the benzene ring were very weak or inactive.

To compare the effect of substituents R^1, R^2, R^3, and R^4 in title compounds **I** on their herbicidal activities, the IC_{50} values of a part of the title compounds with 2,4-Cl_2 as X and Y were examined against the growth of cucumber (*Cucumis sativa*). From the data in Table 3, it can be found that when 2,4-Cl_2 as X and Y was kept constant, great differences for cucumber toxicity could be also made due to the changes of structure of R^1, R^2, and R^3 in the phosphonate moiety and R^4. Inhibitory activity decreased with the increase in the size of both R^1 and R^2 groups attached to phosphorus and R^4. When PrO or EtO groups attached to phosphorus were replaced by a smaller group (MeO), the inhibitory potency of compounds was greatly improved. Compounds with methyl as R^1 and R^2 groups and H as R^4 displayed higher inhibitory potency. By the chemical modifications of R^1, R^2, R^3, and R^4 in the structure **I**, compounds **I-5**, **I-51**, **I-57**, **I-58**, and **I-47** displayed higher activities against the growth of the root and stem of *Cucumis sativa* than did commercial herbicide 2,4-D. However some compounds with larger R^1, R^2, and R^4 groups showed lower activity against the growth of *Cucumis sativa* than did

Table 2. Structures and Herbicidal Activities of *O,O*-Dimethyl α-(Substituted phenoxyacetoxy)benzylphosphonates I[a]



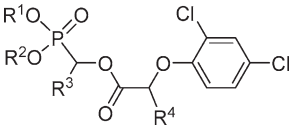
					Ech[b]		*Dig*[b]		*Bra*[b]		*Ama*[b]		*Med*[b]	
no.	R^1R^2	R^3	X	Y	pre	post	pre	post	pre	post	pre	post	pre	post
I-11	Me	Ph	H	H	D	D	D	D	D	D	D	D	D	D
I-12	Me	Ph	H	3-Me	D	D	D	D	B	C	B	C	B	C
I-13	Me	Ph	H	4-Me	D	D	D	D	B	C	B	C	B	C
I-14	Me	Ph	2-Me	3-Me	B	C	D	D	A	B	C	D	B	C
I-15	Me	Ph	2-F	4-F	B	D	D	D	D	D	C	A	C	D
I-16	Me	Ph	3-Cl	4-F	B	C	A	D	A	B	A	B	B	C
I-17	Me	Ph	2-Cl	4-F	A	B	A	C	A	B	A	A	A	B
I-18	Me	Ph	H	4-CF_3	D	D	D	D	D	D	D	D	D	D
I-19	Me	Ph	H	2-NO_2	D	D	D	D	D	D	D	D	D	D
I-20	Me	Ph	2- NO_2	4-CF_3	D	D	D	D	D	D	D	D	D	D
I-21	Me	3-NO_2Ph	3- Me	4-Cl	D	D	C	D	C	D	B	B	B	B
I-22	Me	3-NO_2Ph	2-Cl	5-Me	D	D	D	D	D	D	D	D	D	D
I-23	Me	3-NO_2Ph	2- Me	4-Cl	A	B	A	D	C	B	B	A	B	C
I-24	Me	3-NO_2Ph	2-Cl	3-Cl	D	D	D	D	D	D	D	D	D	D
I-25	Me	3-NO_2Ph	2-Cl	6-Cl	D	D	D	D	D	D	D	D	D	D
I-26	Me	3-NO_2Ph	H	4-Cl	A	B	A	C	A	B	A	B	A	B
I-27	Me	3-NO_2Ph	2-F	4-F	D	D	C	C	D	B	D	B	C	C
I-28	Me	3-NO_2Ph	3-Cl	4-F	C	C	D	B	D	C	C	B	C	C
I-29	Me	3-NO_2Ph	2-Cl	4-F	A	B	A	C	A	B	A	A	A	C
I-30	Me	3-NO_2Ph	H	3-CF_3	D	C	D	D	C	C	C	D	C	D
I-31	Me	3-NO_2Ph	H	4-CF_3	D	D	D	D	D	D	D	C	D	D
I-32	Me	3-NO_2Ph	2-NO_2	4-CF_3	D	D	D	D	D	D	D	D	D	D
I-33	Me	3-Brph	2-Cl	4-Cl	C	C	A	D	A	A	A	A	A	A
I-34	Me	2-OHPh	H	4-Cl	D	D	C	D	D	B	D	B	A	B
I-35	Me	2-OHPh	2-Cl	4-F	C	C	A	D	A	A	A	A	A	A
I-51	Me	Ph	2-Cl	4-Cl	C	D	C	D	A	A	A	A	A	A
I-52	Me	3-NO_2Ph	H	2-F	D	A	D	D	D	D	D	D	D	D
I-53	Me	3-NO_2Ph	H	3-F	D	D	D	D	D	D	D	D	D	D
I-54	Me	3-NO_2Ph	H	4-F	C	C	A	C	D	C	B	B	B	C
I-55	Me	3-NO_2Ph	H	2- NO_2	D	D	D	D	D	D	D	D	D	D
I-56	Me	3-NO_2Ph	H	4- NO_2	D	D	D	D	D	D	D	D	D	D
I-57	Me	3-NO_2Ph	2-Cl	4-Cl	C	D	D	D	A	A	A	A	A	A
2,4-D				B	C	A	C	A	A	A	A	A	A	

[a] Synthetic methods for compounds: **I-55**, **I-56** (ref 15); **I-51**, **I-57**, **I-58** (ref 18); **I-52**, **I-53** (ref 19); **I-54** (ref 20). [b] Inhibitory potency (%) on the growth of plants at a dose of 1500 ai g/ha in a greenhouse was expressed as a four scale: A, 90−100%; B, 75−89%; C, 50−74%; D, ≤50%. *Ech*, *Echinochloa crusgalli*; *Dig*, *Digitaria sanguinali*; Bra, *Brassica campestris*; *Ama*, *Amaranthus retroflexus*; Med, *Medicago sativa*.

2,4-D. These results indicate that the title compounds **I** themselves are responsible for the inhibitory potency against *Cucumis sativa*, not the possible metabolic product 2,4-D.

On the basis of the preliminary bioassays, some of the title compounds were chosen for further herbicide activity assay at lower dose for pre-emergence and postemergence herbicidal activity against *Abutilon theophrasti*, *Amaranthus spinosus*, *Chenopodium album*, *Digitaria sanguinalis*, and *Setaria viridis*. As shown in Table 4, the tested compounds displayed much higher herbicidal activity against dicotyledonous plants than monocotyledons for postemergence. However, they displayed very poor herbicidal activity against tested plants for pre-emergence at a lower dose of 450 or <450 ai g/ha. It is also found that the compounds with 2,4-Cl_2 as X and Y showed higher herbicidal activity than compounds with 2-Me, 4-Cl, or H, 3-CF_3 as X and Y. The tested compounds with 2,4-Cl_2 as X and Y exhibited potent herbicidal activity against dicotyledons for postemergence at a dose of 75 ai g/ha. It was also found that when the EtO group attached to phosphorus in the compound was replaced by MeO, the herbicidal activity of the compound

Table 3. IC_{50} Values of *O,O*-Dialkyl α-(2,4-Dichlorophenoxyacetoxy)alkylphosphonates I against *Cucumis sativa*[a]



no.	R^1	R^2	R^3	R^4	stem length IC_{50} (μM)	no.	R^1	R^2	R^3	R^4	root length IC_{50} (μM)
I-57	Me	Me	3-NO_2Ph	H	5.02	**I-5**	Me	Me	Me	H	0.0025
I-64	Me	Me	Me	Me	6.74	**I-57**	Me	Me	3-NO_2Ph	H	0.0034
I-5	Me	Me	Me	H	7.56	**I-58**	Me	Me	Pr	H	0.0075
I-47	Me	Me	H	H	7.79	**I-51**	Me	Me	Ph	H	0.0142
I-51	Me	Me	Ph	H	8.95	**I-62**	Et	Et	Ph	H	0.0488
I-58	Me	Me	Pr	H	9.34	**I-47**	Me	Me	H	H	0.0577
2,4-D					10.14	2,4-D					0.2280
I-59	Et	Et	H	H	13.4	**I-60**	Et	Et	Me	H	0.3
I-65	Me	Me	Pr	Me	13.7	**I-61**	Et	Et	Et	H	0.31
I-10	Me	Me	Et	H	14	**I-10**	Me	Me	Et	H	0.327
I-68	Pr	Pr	Me	Me	18	**I-67**	Et	Et	Me	Me	1.08
I-61	Et	Et	Et	H	25.70	**I-64**	Me	Me	Me	Me	1.4200
I-66	Me	Me	Ph	Me	30.1	**I-63**	Et	Et	2-ClPh	H	1.49
I-69	Pr	Pr	Ph	Me	44.3	**I-68**	Pr	Pr	Me	Me	2.4400
I-62	Et	Et	Ph	H	109.00	**I-65**	Me	Me	Pr	Me	2.9800
I-70	Pr	Pr	2-ClPh	Me	164.00	**I-69**	Pr	Pr	Ph	Me	4.5200
						I-70	Pr	Pr	2-ClPh	Me	12.1000

[a] Synthetic methods for compounds: **I-58** (ref 18); **I-59**, **I-60**, **I-61** (ref 11); **I-62** (refs 12 and 17); **I-63**, **I-70** (ref 16); **I-64**—**I-68** (ref 14); **I-69** (ref 13).

could be improved. Compounds **I-5** and **I-60** both have 2,4-Cl_2 as X and Y; however, compound **I-5** with a MeO group attached to the phosphorus exhibited better herbicidal activity than did compound **I-60** at a dose of 75 ai g/ha. Compound **I-5** was found to exhibit promising herbicidal activity among the tested compounds.

To evaluate the postemergence herbicidal activity and herbicidal spectrum, compound **I-5** was selected for further examination at the dose of 18.75−450 ai g/ha against *Eclipta prostrata*, *Amaranthus retroflexus*, *Xanthium strumarium*, *Cardamine hirsuta*, *Portulaca oleracea*, and *Medicago sativa* for postemergence. As shown in Table 5, compound **I-5** exhibited excellent herbicidal activity against broadleaf weeds by postemergence application at a dose of 18.75−450 ai g/ha. However, it showed no activity against monocot weeds *Echinochloa crusgalli* and *Digitaria sanguinalis* at the above dose. Crop selectivity of **I-5** at the dose of 37.5−600 ai g/ha was further examined with *Brassica campestris*, *Daucus carota*, *Triticum aestivum*, *Zea mays*, and *Oryza sativa* as some representative crops tested for compound **I-5**'s selectivity (Table 6). Among the tested crops, rape and carrot as dicotyledonous crops are very susceptible at doses as low as 37.5 ai g/ha, whereas wheat, maize, and rice exhibited higher tolerance to compound **I-5** even at the dose of 600 ai g/ha. The data in Tables 5 and 6 further show all tested broadleaf plants are highly sensitive to compound **I-5** by postemergence application at lower dose. **I-5** has a broad-spectrum herbicidal activity against broadleaf weeds and a higher level of selectivity in wheat, maize, and rice.

Inhibition on PDHc. Inhibitory potencies of compound **I-5** and some compounds against PDHc from three different plants were examined. The IC_{50} values are listed in Table 7.

As shown in Table 7, compound **I-5** exhibited powerful inhibitory potency against PDHc from both *Phaseolus radiatus* and *Pisum sativum* (mung bean and pea) and showed relatively lower inhibitory potency against PDHc from *Oryza sativa* (rice). The above observations showed that **I-5** could selectively inhibit the PDHc from dicotyledonous plant.

To recognize the type of mechanism of the inhibitor **I-5**, the maximum velocity (V_{max}) and Michaelis constant (K_m) were determined by measuring the effect of variations in the concentration of sodium pyruvate as a substrate on PDHc in the presence or absence of the inhibitor **I-5**. According to the enzyme kinetic experiments of **I-5**, V_{max} and K_m were obtained

in the presence of inhibitor **I-5**:

$$K_m = 0.03262 \text{ mM}, \quad V_{max} = 0.4349 \text{ mM}$$

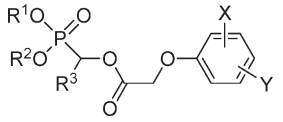
in the absence of inhibitor **I-5**:

$$K_m' = 0.01329 \text{ mM}, \quad V_{max} = 0.4373 \text{ mM}$$

The data showed that V_{max} almost remains constant and that K_m is increased in the presence of inhibitor **I-5**. When the concentration of substrate sodium pyruvate was increased to 0.5 mM, the inhibition almost disappeared. This indicates that the inhibition of PDHc by **I-5** can be overcome by increasing the concentration of substrate. K_m increased in the presence of inhibitor **I-5**, indicating that the apparent affinity of the enzyme for its substrate decreases in the presence of the inhibitor. **I-5** could diminish the rate of catalysis by reducing the proportion of enzyme molecules bound to the substrate. However, the V_{max} values were almost the same in the presence and absence of **I-5**. On the basis of these results, compound **I-5** was found to act as a competitive inhibitor of PDHc.

Relationships of Herbicidal Activity and Inhibitory Potency against PDHc. By comparison of the inhibitory potency against PDHc and herbicidal activity of tested compounds in

Table 4. Structures and Herbicidal Activities of Some Title Compounds



no.	g ai/ha	treatment	*Abu*[a]	*Amas*[a]	*Che*[a]	*Dig*[a]	*Set*[a]
I-5	75	post	97.5 ± 2.5	90 ± 2.5	92.5 ± 2.5	0	0
		pre	0	0	0	0	0
	150	post	100 ± 0	100 ± 0	96 ± 2.0	0	0
		pre	0	0	0	0	0
	450	post	100 ± 0	100 ± 0	100 ± 0	37.5 ± 2.5	0
		pre	0	0	0	45 ± 5.0	35 ± 5.0
I-38	150	post	82 ± 2.0	10 ± 2.0	60 ± 5.0	0	0
		pre	0	0	0	0	0
	450	post	96 ± 2.0	100 ± 0	80 ± 5.0	10 ± 2.0	0
		pre	0	30 ± 2.0	0	16 ± 2.0	0
I-23	75	post	50 ± 0	50 ± 0	52.5 ± 2.5	0	0
		pre	0	0	0	0	0
	150	post	90 ± 5.0	100 ± 0	82.5 ± 2.5	0	0
		pre	0	0	0	0	0
	450	post	100 ± 0	100 ± 0	100 ± 0	10 ± 2.0	0
		pre	0	0	0	0	0
I-47	75	post	82.5 ± 2.5	62.5 ± 2.5	82.5 ± 2.5	0	0
		pre	0	0	0	0	0
	150	post	99 ± 1.0	92.5 ± 2.5	99.5 ± 0.5	0	0
		pre	0	0	0	0	0
	450	post	100 ± 0	97.5 ± 2.5	100 ± 0	0	0
		pre	47.5 ± 2.5	52.5 ± 2.5	52.5 ± 2.5	0	5.0 ± 5.0
I-48	75	post	27.5 ± 2.5	0	32.5 ± 2.5	0	0
		pre	0	0	0	0	0
	150	post	42.5 ± 2.5	0	42.5 ± 2.5	0	0
		pre	0	0	0	0	0
	450	post	87.5 ± 7.5	57.5 ± 2.5	82.5 ± 2.5	0	0
		pre	0	0	0	35 ± 5.0	35 ± 5.0
I-51	75	post	82.5 ± 2.5	62.5 ± 2.5	82.5 ± 2.5	0	0
		pre	0	0	0	0	0
	150	post	92.5 ± 2.5	70 ± 5.0	92.5 ± 2.5	0	0
		pre	0	0	0	0	0
	450	post	100 ± 0	87.5 ± 2.5	97.5 ± 2.5	0	0
		pre	0	0	0	45 ± 5.0	30 ± 0
I-60	75	post	97.5 ± 2.5	52.5 ± 2.5	62.5 ± 2.5	0	0
		pre	0	0	0	0	0
	150	post	97.5 ± 2.5	82.5 ± 2.5	92.5 ± 2.5	0	0
		pre	0	0	0	0	0
	450	post	100 ± 0	100 ± 0	97.5 ± 2.5	0	0
		pre	47.5 ± 2.5	50 ± 0	52.5 ± 2.5	10 ± 0	5.5 ± 4.5
2,4-D	75	post	72.5 ± 2.5	60 ± 5.0	72.5 ± 2.5	0	0
		pre	0	0	0	0	0
	150	post	100 ± 0	92.5 ± 2.5	92.5 ± 2.5	0	0
		pre	0	0	0	10 ± 0	5.5 ± 4.5
	450	post	100 ± 0	92.5 ± 2.5	100 ± 0	0	0
		pre	0	0	0	65 ± 5.0	67.5 ± 2.5

[a] Evaluation in greenhouse test. *Abu, Abutilon theophrasti*; *Amas, Amaranthus spinosus*; *Che, Chenopodium album*; *Dig, Digitaria sanguinalis*; *Set, Setaria viridis*.

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Table 5. Herbicidal Activity of Compound I-5 (HW02) for Post-emergence (Relative Inhibiton of Growth %)[a]

g ai/ha (post)	*Ecl*[a]	*Ama*[a]	*Xan*[a]	*Car*[a]	*Por*[a]	*Med*[a]	*Ech*[a]	*Dig*[a]
18.75	56 ± 0	65 ± 2	50 ± 0	75 ± 0	50 ± 0	60 ± 0	0	0
37.5	72 ± 2	82.5 ± 2.5	60 ± 0	84 ± 1	65 ± 0	72 ± 2.	0	0
75	82.5 ± 2.5	90 ± 0	72 ± 2	88 ± 0	75 ± 0	84 ± 2	0	0
150	90 ± 2	96 ± 2	90 ± 1	92.5 ± 2.5	85 ± 2.5	90 ± 2	0	15 ± 5
225	96. ± 1.5	100 ± 0	95 ± 0	95 ± 0	90 ± 1	95 ± 0	10 ± 0	22.5 ± 2.5
300	100 ± 0	100 ± 0	100 ± 0	100 ± 0	95 ± 0	97 ± 0	20 ± 5	32 ± 5
450	100 ± 0	100 ± 0	100 ± 0	100 ± 0	100 ± 0	100 ± 0	35 ± 4.0	37.5 ± 2.5

[a] Evaluation in greenhouse test. *Ecl, Eclipta prostrata*; *Ama, Amaranthus retroflexus*; *Xan, Xanthium strumarium*; *Car, Cardamine hirsuta*; *Por, Portulaca oleracea*; *Med, Medicago sativa*; *Ech, Echinochloa crusgalli*; *Dig, Digitaria sanguinalis*.

Table 6. Crop Selectivity of Compound I-5 (HW02)

g ai/ha (post)	*Bra*[a]	*Dau*[a]	*Tri*[a]	*Zea*[a]	*Ory*[a]
37.5	90 ± 0	85 ± 0	N[b]	N	N
75	92.5. ± 2.5	90 ± 0	N	N	N
150	97. ± 2.5	95 ± 2.5	N	0	N
225	100 ± 0	97. ± 2.5	0	0	0
300	100 ± 0	100 ± 0	0	0	0
450	100 ± 0	100 ± 0	0	0	0
600	100 ± 0	100 ± 0	0	0	0

[a] Evaluation in greenhouse test. *Bra, Brassica campestris*; *Dau, Daucus carota*; *Tri, Triticum aestivum*; *Zea, Zea mays*; *Ory, Oryza sativa*. [b] N, not tested.

Table 7. Herbicidal Activity and Inhibitory Activity against PDHc of HW02 and Some Analogues

no.	*Ech*[a]	*Ory*[a]	*Ama*[a]	*Bra*[a]	*Pis*[a]	IC_{50} (μM)
I-60	5.0 ± 5.0	37.5 ± 2.5	100 ± 0	97.5 ± 2.5	82.5 ± 2.5	23.12[b]
I-47	0	0	97.5 ± 2.5	100 ± 0	87.5 ± 2.5	19.70[b]
I-5 (HW02)	35 ± 4.0	0	100 ± 0	100 ± 0	97.5 ± 2.5	12.75[b] 18.19[c] 867.3[d]
I-51	25 ± 5.0	10 ± 0	87.5 ± 2.5	97.5 ± 2.5	96.5 ± 1.5	95.38[b]
I-48	0	32.5 ± 2.5	57.5 ± 2.5	82.5 ± 2.5	10 ± 0	442[c]
2,4-D	35 ± 4.0	37.5 ± 2.5	100 ± 0	100 ± 0	97.5 ± 2.5	>4530[c]

[a] Inhibitory potency (%) on the growth of plants at a dose of 450 ai g/ha in greenhouse. *Ech, Echinochloa crusgalli*; *Ory, Oryza sativa*; *Ama, Amaranthus retroflexus*; *Bra, Brassica juncea*; *Pis, Pisum sativum*. [b] IC_{50} values against PDHc from *Phaseolus radiatus*. [c] IC_{50} values against PDHc from *Pisum sativum*. [d] IC_{50} values against PDHc from *Oryza sativa*.

Table 7, the herbicidal activity correlated well to the inhibitory potency against PDHc. It was found that the 2- and 4-positions in the benzene ring were the most essential sites for substitution, which could greatly enhance both inhibitory potency against PDHc and herbicidal activity. The inhibitory potency against PDHc from *Pisum sativum* could be greatly enhanced by introducing 2,4-Cl_2 into the phenoxybenzene ring (such as **I-5** > **I-48**). This result is in accord with the results of molecular docking and 3D-QSAR studies on the binding modes of title compounds to PDH E1.[22] When 2,4-Cl_2 is kept constant as X and Y, the inhibitory potency of the compound could be also greatly enhanced by the chemical modification of R^1, R^2, and R^3 in the phosphonate moiety. Compounds with MeO as the R^1O and R^2O groups displayed higher inhibitory potency than those with EtO as the R^1O and R^2O groups (such as **I-5** > **I-60**, against PDHc from *Phaseolus radiatus*). When X, Y, R^1, R^2, and R^4 are kept constant, the introduction of Me at R^3 seems to have a favorable effect on inhibitory potency, such as the inhibitory potency against PDHc from *Phaseolus radiatus* (**I-5** > **I-47** > **I-51**). It is shown that the methyl group is needed for good binding to the enzyme. The above results indicate that the improvement of inhibitory potency against PDHc also requires a reasonable combination of R^1, R^2, R^3, X, and Y.

Compound **I-5** with 2,4-Cl_2 as X and Y, Me as R^1, R^2, and R^3, and H as R^4 showed higher inhibitory activity against PDHc from dicotyledons and also higher herbicidal activity against dicotyledons than other compounds. It is worth noting that **I-5** showed higher inhibitory potency against PDHc from *Pisum sativum* than *Oryza sativa* in vitro; it also showed higher inhibitory activity against *Pisum sativum* than *Oryza sativa* in vivo by the herbicide activity assay in the greenhouse test (Table 7). These results indicate that **I-5** showed higher inhibitory potency against dicotyledons than that against monocotyledons, and the results of bioactivity evaluation in vitro and in vivo could be in good accordance. 2,4-D, as a metabolic product of compound **I-5**, also showed higher activity against *Pisum sativum* and other tested dicotyledonous plants in vivo; however, it was almost inactive against PDHc from *Pisum sativum* in vitro (Table 7). These results indicate that **I-5** and 2,4-D have different action modes, and compound **I-5** showed herbicidal activity due to its inhibitory activity against PDHc, but not by the action of 2,4-D. Furthermore, compound **I-5** had a higher safety level to the monocot crops in wheat, maize, and rice fields due to its weak activity against PDHc from monocotyledons. The results indicate that compound **I-5** (HW02) could be used as a selective herbicide for broadleaf weed control in monocot crop fields. To the best of our knowledge, none of the PDHc inhibitors prepared was active enough to be commericalized as herbicide. Up to now, some reported PDHc inhibitors, such as acylphosphinates, acylphosphonates, and relative compounds, did not show sufficient commercial potential for full development as herbicides because their activity was demonstrated at an impressive level in the field.[1,2] Compound **I-5** (HW02) seems to be the first compound that shows practical herbicidal activity as a PDHc inhibitor.

In summary, our results showed that satisfactory herbicidal activity of *O*,*O*-dialkyl α-(substituted phenoxyacetoxy)alkylphosphonates **I** could be achieved by a reasonable combination of both phosphonate and phenoxyacetate moieties. The results indicate that the degree of herbicidal activity of tested compounds positively correlated with that of inhibition of PDHc. The herbicidal activity and inhibitory potency against PDHc of the title compounds could be increased greatly by optimizing R^1, R^2, R^3, R^4, X, and Y in parent compound **I**. Two chlorine atoms or one chlorine atom and one fluorine atom as X and Y should be

substituted at the 2- and 4-positions on a phenoxybenzene ring. Another condition was that R^1 and R^2 attached to the phosphorus atom should not be bulky. A smaller group such as MeO was the best. Besides, smaller groups such as H as R^4 and Me as R^3 were beneficial to herbicidal activity. Therefore, *O*,*O*-dimethyl α-(2,4-dichlorophenoxyacetoxy)ethyl phosphonate **I-5** (HW02), which acted as a competitive inhibitor of PDHc with much higher inhibitory potency against PDHc from *Pisum sativum* and *Phaseolus radiatus* than *Oryza sativa*, was found to be the most effective compound against broadleaf weeds. The results indicated that compound **I-5** (HW02) could be developed as a postemergence herbicide with good selectivity in monocot crops. Further field trials are under way.

■ ASSOCIATED CONTENT

Supporting Information. Structures of compounds **I-1**−**I-70**, source of seeds used for bioassays, and regression equation and *R* values for the IC_{50} values along with detailed data for enzyme kinetic experiment of **I-5** against PDHc. This material is available free of charge via the Internet at http://pubs.acs.org.

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