DOI 10.5012/bkcs.2010.31.8.2315

Spirodiclofen Analogues as Potential Lipid Biosynthesis Inhibitors: A Convenient Synthesis, Biological Evaluation, and Structure-Activity Relationship

Shaoyong Ke,* Tingting Sun, Zhigang Zhang, Ya-Ni Zhang, Ying Liang, Kaimei Wang, and Ziwen Yang

Hubei Biopesticide Engineering Research Center, Hubei Academy of Agricultural Sciences, Wuhan 430064, P. R. China
**E-Mail: keshaoyong@163.com*
Received February 8, 2010, Accepted June 30, 2010

Twenty spirodiclofen analogues have been designed and conveniently synthesized *via* three steps including esterification, one-pot heterocyclization, and acylation reactions. The target molecules have been identified on the basis of analytical spectra (^{1}H NMR, ^{13}C NMR and ESI-MS) data. All newly synthesized compounds have been screened for their potential insecticidal and herbicidal activity by standard method. The preliminary assays indicated that some of analogues displayed moderate to good insecticidal activity against *Plutella xylostella* compared with spirodiclofen, and some compounds showed obvious activity against *Brassica chinensis*. Structure-activity relationship (SAR) is also discussed based on the experimental data.

Key Words: Spirodiclofen analogues, Synthesis, Biological activity, Structure-activity relationship

Introduction

Highly efficacious acaricides/insecticides with novel modes of action are becoming increasingly important during the course of integrated pest management (IPM) strategy.[1,2] Nowadays, since mites usually acquire resistance rapidly, the useful life of acaricides on the market is becoming shorter.[3] Thus the focus of novel acaricides is shifting towards more effective, eco-friendly new structures with novel modes of action. Recently, spirodiclofen (Trade name: Envidor®, and Daniemon®; Figure 1) derived from natural tetronic acid core, a novel broad spectrum, non-systemic acaricide, was discovered by Bayer CropScience,[4-6] which belong to the novel chemical class of spirocyclic phenyl-substituted tetronic acid derivatives. Spirodiclofen controls a broad range of phytophagous mite species such as *Tetranychus*, *Phyllocoptruta*, *Panonychus*, *etc*. at all stages of growth and has a unique mode of action differed from other acaricides,[7,8] which interferes the lipid biosynthesis and blocks the enzyme acetyl-coenzyme A carboxylase that allows mites to form important fatty acids.[9-12] Bayer Company has already developed the three tetronic acids analogues as acaricides and insecticides named spirodiclofen,[5,6] spiromesifen,[13-16] and spirotetramat (Figure 1),[17-21] respectively.

However, both the mode of action and the structure of receptor for these compounds are not yet sure, and the systemic and detailed structure-activity relationships are unreported, which restrict the creation and optimization for these kinds of novel acaricides to some extent. Owing to the unique natural keto-enol structure, novel mode of action and potential significance in insect resistance management[22-26] for spirocyclic tetronic acid derivatives, so it is important to establish the structure-activity relationships and extend activity profile for these compounds.

The aim of the present study was to develop more effective or broad-spectrum bioactive derivatives of spirodiclofen that could serve as potential lipid biosynthesis inhibitors for agrochemicals and explore the preliminary structure-activity relationships for these derivatives. As part of our agrochemistry program aimed at the search for novel heterocycle-based bioactive molecules, we wish to report herein the convenient synthesis, and biological evaluation of series of spiro-tetronic acid derivatives (Figure 2).

Experimental

Instrumentation and chemicals. All melting points (mp) were obtained using a digital model X-5 apparatus and are uncorrected. ^{1}H NMR and ^{13}C NMR spectra were recorded on a Brucker AM-400 (400 MHz) spectrometer with $CDCl_3$ or DMSO-d_6 as the solvent and TMS as the internal standard. Chemical shifts are reported in δ (parts per million) values. Coupling constants ^nJ are reported in Hz. Mass spectra were performed on a Micro-

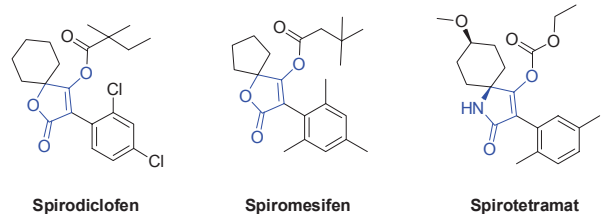


Figure 1. Structures of novel insecticides/acaricides derived from natural tetronic acid.

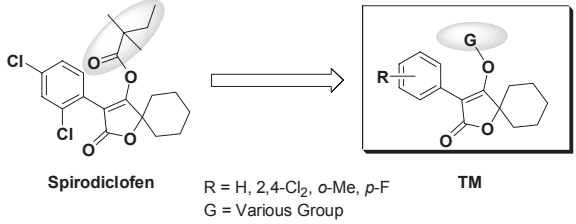


Figure 2. Design strategy of the target compounds.

Mass Quattro *micro*™ API instrument. Analytical thin-layer chromatography (TLC) was carried out on precoated plates (silica gel 60 F254), and spots were visualized with ultraviolet (UV) light. All chemicals or reagents were purchased from standard commercial supplies. Anhydrous CH_2Cl_2 and THF were prepared by standard methods. All other solvents and reagents were analytical reagent and used directly without purification.

General synthetic procedure for the intermediates esters 2 and 4. The various esters **2** and **4** were prepared *via* normal esterification reactions of an acid with methanol under catalytic amount of sulfuric acid. Following the brief descriptions: the various acids (0.10 mol) is dissolved in 50 mL anhydrous methanol, 3 mL of conc. sulfuric acid is added, and the mixture refluxed for 5 - 8 h. Part of the methanol is distilled in vacuo and the residue is poured into 80 mL of ice water. The mixture is extracted three times with 30 mL of ethyl acetate. The combined extracts are washed with 30 mL of water, dried over anhydrous sodium sulfate, evaporated in vacuo to obtain the residue, which was used for the next step reaction without further purification.

General synthetic procedure for the tetronic acid 5a-d. The important intermediate tetronic acid **5a-d** is conveniently obtained by the modified one-pot cyclization method developed by Thierry Le Gall *et al*.[27,28] The modified synthetic method are as follows: To a solution of substituted phenylacetate (0.01 mol, 1 equiv) and methyl 1-hydroxycyclohexanecarboxylate 1.90 g (0.012 mol, 1.2 equiv) in anhydrous THF was added a solution of potassium *tert*-butoxide 2.46 g (0.022 mol, 2.2 equiv) in THF. The suspension obtained was then refluxed under nitrogen overnight. After cooling to ambient temperature, the reaction mixture was acidified with 5% HCl aqueous to pH 2 - 3, and then was extracted three times with ethyl acetate. The combined organic layers were then washed with water and dried over Na_2SO_4. After filtration and concentration under vacuum, the residue was taken up into little ethyl acetate (3 - 5 mL) and filtered in vacuo to give the corresponding tetronic acid **5a-d** as white powder. Their physico-chemical properties and the spectra data are as follows:

4-Hydroxy-3-phenyl-1-oxaspiro[4.5]dec-3-en-2-one (5a): This compound was obtained following the above-described method as white powder, yield 76%, mp 256 - 257 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.40 (bs, 1H, OH), 7.21-7.87 (m, 5H, Ph-H), 1.19-2.06 (m, 10H, Cyclohexyl-H); ESI-MS: calcd for $C_{15}H_{16}O_3$ ($[M+1]^+$), 245.3; found, 245.8.

4-Hydroxy-3-*o*-tolyl-1-oxaspiro[4.5]dec-3-en-2-one (5b): This compound was obtained following the above-described method as white powder, yield 78%, mp 223 - 225 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.08 (bs, 1H, OH), 7.11-7.24 (m, 4H, Ph-H), 2.15 (s, 3H, Ph-CH_3), 1.23-1.96 (m, 10H, Cyclohexyl-H); ESI-MS: calcd for $C_{16}H_{18}O_3$ ($[M+1]^+$), 259.3; found, 259.8.

3-(2,4-Dichlorophenyl)-4-hydroxy-1-oxaspiro[4.5]dec-3-en-2-one (5c): This compound was obtained following the above-described method as white powder, yield 80%, mp 249 - 250 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.46 (bs, 1H, OH), 7.28-7.63 (m, 3H, Ph-H), 1.04-1.85 (m, 10H, Cyclohexyl-H); ESI-MS: calcd for $C_{15}H_{14}Cl_2O_3$ ($[M+1]^+$), 314.2; found, 313.8.

3-(4-Fluorophenyl)-4-hydroxy-1-oxaspiro[4.5]dec-3-en-2-one (5d): This compound was obtained following the above-described method as white powder, yield 72%, mp > 260 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.45 (bs, 1H, OH), 7.91-7.95 (m, 2H, Ph-H), 7.21 (t, J = 9 Hz, 2H, Ph-H), 1.19-2.05 (m, 10H, Cyclohexyl-H); ESI-MS: calcd for $C_{15}H_{15}FO_3$ ($[M+1]^+$), 263.3; found, 263.8.

General synthetic procedure for the target compounds 6a-t. The typical process of synthesis of spirodiclofen analogues **6a-t** is shown as following: To a stirred solution of 4-hydroxy-3-(substituted phenyl)-1-oxaspiro[4.5]dec-3-en-2-one **5a-d** (1 mmol) and Et_3N (1.5 mmol) in anhydrous dichloromethane (10 mL) under an ice bath, and various acyl chloride (1.1 mmol) in dichloromethane (5 mL) was added dropwise. The mixture was stirred at low temperature for about 1 h and then was allowed to react at ambient temperature for another 1 - 4 hours, which was detected by TLC. After this, the reaction mixture was poured into water and extracted with dichloromethane. The combined organic layer was washed extensively with aqueous sodium bicarbonate, brine and water, and then dried over anhydrous sodium sulfate. The solvent was evaporated, and the residue was purified by column chromatography on silica gel using petroleum ether/ethyl acetate as eluent to afford the target compounds **6a-t**. Their physico-chemical properties and the spectra data are as follows:

3-(2,4-Dichlorophenyl)-2-oxo-1-oxaspiro[4.5]dec-3-en-4-yl methanesulfonate (6a): This compound was obtained following the above-described method as white solid, yield 81%, mp 138 - 140 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.49 (d, ^4J = 1.2 Hz, 1H, Ph-H), 7.34-7.39 (m, 2H, Ph-H), 2.99 (s, 3H, CH_3-S), 1.73-1.94 (m, 10H, Cyclohexyl-H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.68, 168.28, 136.41, 134.59, 132.87, 129.58, 127.57, 125.58, 84.59, 40.31, 33.10, 32.11, 32.09, 24.23, 21.62; ESI-MS: calcd for $C_{16}H_{16}Cl_2O_5S$ (M^+), 391.3; found, 391.8.

3-(2,4-Dichlorophenyl)-2-oxo-1-oxaspiro[4.5]dec-3-en-4-yl ethyl carbonate (6b): This compound was obtained following the above-described method as white powder, yield 86%, mp 79 - 80 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.41 (d, ^4J = 2 Hz, 1H, Ph-H), 7.38 (d, J = 8.4 Hz, 1H, Ph-H), 7.31 (dd, ^4J = 2 Hz, J = 8 Hz, 1H, Ph-H), 4.15 (q, J = 7 Hz, 2H, $\underline{CH_2}CH_3$), 1.72-1.91 (m, 10H, Cyclohexyl-H), 1.24 (t, J = 7 Hz, 3H, $CH_2\underline{CH_3}$); ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.53, 170.58, 149.94, 136.62, 135.68, 133.07, 130.38, 128.48, 128.26, 111.42, 84.86, 67.66, 47.05, 34.15, 25.55, 22.91, 15.06, 9.80; ESI-MS: calcd for $C_{18}H_{18}Cl_2O_5$ ($[M+1]^+$), 386.2; found, 385.8.

3-(2,4-Dichlorophenyl)-2-oxo-1-oxaspiro[4.5]dec-3-en-4-yl 2-phenylacetate (6c): This compound was obtained following the above-described method as white crystal, yield 68%, mp 85 - 86 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.24-7.26 (m, 3H, Ph-H), 7.19 (s, 1H, Ph-H), 7.08-7.17 (m, 4H, Ph-H), 3.69 (s, 2H, CH_2CO), 1.53-1.70 (m, 10H, Cyclohexyl-H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.57, 163.85, 134.29, 133.12, 130.75, 130.70, 128.42, 128.32, 128.20, 128.16, 127.87, 127.65, 126.84, 126.24, 125.73, 111.13, 82.83, 39.83, 31.86, 23.35, 20.62; ESI-MS: calcd for $C_{23}H_{20}Cl_2O_4$ (M^+), 431.3; found, 431.9.

3-(2,4-Dichlorophenyl)-2-oxo-1-oxaspiro[4.5]dec-3-en-4-yl 2-(2,4-dichloro phenyl)acetate (6d): This compound was obtained following the above-described method as white flocculent solid, yield 88%, mp 142 - 143 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.42 (s, 1H, Ph-H), 7.37 (s, 1H, Ph-H), 7.28-7.33 (m, 2H, Ph-H), 7.22 (d, J = 8 Hz, 1H, Ph-H), 7.13 (d, J = 8 Hz, 1H,

Ph-H), 3.91 (s, 2H, CH_2CO), 1.63-1.87 (m, 10H, Cyclohexyl-H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.27, 169.20, 163.52, 135.49, 135.23, 134.86, 134.13, 132.19, 131.86, 129.61, 129.21, 128.87, 127.58, 127.39, 126.78, 112.09, 83.80, 38.00, 32.94, 30.93, 24.42, 21.68; ESI-MS: calcd for $C_{23}H_{18}Cl_4O_4$ ($[M+1]^+$), 501.2; found, 501.5.

3-(2,4-Dichlorophenyl)-2-oxo-1-oxaspiro[4.5]dec-3-en-4-yl 2-*o*-tolylacetate (6e): This compound was obtained following the above-described method as yellow powder, yield 83%, mp 102 - 104 °C; ^1H NMR (400 MHz, $CDCl_3$) δ 7.34 (d, ^4J = 1.6 Hz, 1H, Ph-H), 7.20-7.24 (m, 3H, Ph-H), 7.15 (t, J = 7.8 Hz, 2H, Ph-H), 7.10 (d, J = 6.8 Hz, 1H, Ph-H), 3.78 (s, 2H, CH_2CO), 2.17 (s, 3H, Ph-CH_3), 1.59-1.79 (m, 10H, Cyclohexyl-H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.64, 169.26, 164.63, 136.79, 135.33, 134.18, 131.77, 130.66, 130.45, 130.24, 129.21, 128.19, 127.29, 126.84, 126.44, 112.16, 83.80, 38.62, 32.89, 24.40, 21.67, 19.29; ESI-MS: calcd for $C_{24}H_{22}Cl_2O_4$ (M^+), 445.3; found, 445.7.

3-(2,4-Dichlorophenyl)-2-oxo-1-oxaspiro[4.5]dec-3-en-4-yl 3,4,5-trimethoxybenzoate (6f): This compound was obtained following the above-described method as white powder, yield 75%, mp 183 - 185 °C; ^1H NMR (400 MHz, $CDCl_3$) δ 7.44 (d, J = 8.4 Hz, 1H, Ph-H), 7.36 (d, ^4J = 1.2 Hz, 1H, Ph-H), 7.32 (dd, J = 8.4 Hz, 1H, Ph-H), 7.25 (s, 2H, Ph-H), 3.94 (s, 3H, CH_3O), 3.90 (s, 6H, CH_3O), 1.24-1.88 (m, 10H, Cyclohexyl-H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.80, 169.40, 159.84, 153.17, 144.02, 135.30, 134.20, 131.81, 129.31, 127.36, 127.02, 121.60, 112.00, 108.04, 83.93, 60.98, 56.39, 33.18, 24.39, 21.71; ESI-MS: calcd for $C_{25}H_{24}Cl_2O_7$ (M^+), 507.4; found, 507.9.

3-(2,4-Dichlorophenyl)-2-oxo-1-oxaspiro[4.5]dec-3-en-4-yl 4-fluorobenzoate (6g): This compound was obtained following the above-described method as white powder, yield 82%, mp 155 - 156 °C; ^1H NMR (400 MHz, $CDCl_3$) δ 7.96-8.00 (m, 2H, Ph-H), 7.09-7.39 (m, 5H, Ph-H), 1.19-1.84 (m, 10H, Cyclohexyl-H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.37, 169.45, 167.97, 165.41, 158.98, 135.30, 134.19, 133.36, 133.26, 131.80, 129.21, 127.34, 127.06, 123.33, 123.30, 116.40, 116.18, 111.58, 83.84, 33.12, 24.35, 21.69; ESI-MS: calcd for $C_{22}H_{17}Cl_2FO_4$ ($[M+1]^+$), 436.3; found, 435.9.

2-Oxo-3-*o*-tolyl-1-oxaspiro[4.5]dec-3-en-4-yl 2,2-dimethylbutanoate (6h): This compound was obtained following the above-described method as white powder, yield 85%, mp 87 - 89 °C; ^1H NMR (400 MHz, $CDCl_3$) δ 7.13-7.23 (m, 3H, Ph-H), 7.06 (d, J = 7.6 Hz, 1H, Ph-H), 2.27 (s, 3H, Ph-CH_3), 1.71-1.85 (m, 10H, Cyclohexyl-H), 1.50 (q, J = 7.6 Hz, 2H, $\underline{CH_2}CH_3$), 1.07 (s, 6H, CH_3), 0.62 (t, 3H, J = 7.6 Hz, $CH_2\underline{CH_3}$); ^{13}C NMR (100 MHz, $CDCl_3$) δ 172.24, 171.39, 169.83, 137.27, 130.88, 130.31, 129.49, 129.00, 127.51, 125.61, 117.98, 84.00, 43.29, 33.26, 32.87, 30.92, 24.57, 24.38, 21.78, 19.57, 8.83; ESI-MS: calcd for $C_{23}H_{18}Cl_4O_4$ ($[M+1]^+$), 357.5; found, 358.0.

2-Oxo-3-*o*-tolyl-1-oxaspiro[4.5]dec-3-en-4-yl methanesulfonate (6i): This compound was obtained following the above-described method as colorless crystal, yield 94%, mp 118 - 121 °C; ^1H NMR (400 MHz, $CDCl_3$) δ 7.29-7.37 (m, 2H, Ph-H), 7.22-7.25 (m, 2H, Ph-H), 2.59 (s, 3H, CH_3-S), 2.30 (s, 3H, Ph-CH_3), 1.74-1.93 (m, 10H, Cyclohexyl-H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.79, 168.87, 138.05, 130.64, 130.09, 129.93, 126.87, 126.02, 124.91, 118.80, 84.45, 40.46, 32.97, 32.48, 24.25, 21.68, 19.51; ESI-MS: calcd for $C_{17}H_{20}O_5S$ ($[M+1]^+$), 337.4; found, 337.8.

Ethyl 2-oxo-3-*o*-tolyl-1-oxaspiro[4.5]dec-3-en-4-yl carbonate (6j): This compound was obtained following the above-described method as pale yellow solid, yield 74%, mp 72 - 74 °C; ^1H NMR (400 MHz, $CDCl_3$) δ 7.07-7.19 (m, 4H, Ph-H), 3.94 (q, J = 7.2 Hz, 2H, $\underline{CH_2}CH_3$), 2.19 (s, 3H, Ph-CH_3), 1.65-1.82 (m, 10H, Cyclohexyl-H), 1.03 (t, J = 7.2 Hz, 3H, $CH_2\underline{CH_3}$); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.35, 169.72, 148.98, 137.09, 130.39, 129.25, 129.04, 127.27, 125.78, 115.74, 83.51, 66.14, 32.93, 24.33, 21.70, 19.54, 13.64; ESI-MS: calcd for $C_{19}H_{22}O_5$ ($[M+1]^+$), 331.4; found, 331.9.

2-Oxo-3-*o*-tolyl-1-oxaspiro[4.5]dec-3-en-4-yl 2-(2,4-dichlorophenyl)acetate (6k): This compound was obtained following the above-described method as white crystal, yield 90%, mp 122 - 125 °C; ^1H NMR (400 MHz, $CDCl_3$) δ 7.37 (d, J = 2 Hz, 1H, Ph-H), 7.03-7.26 (m, 5H, Ph-H), 6.94 (d, J = 8.4 Hz, 1H, Ph-H), 3.77 (s, 2H, CH_2CO), 2.20 (s, 3H, Ph-CH_3), 1.69-1.82 (m, 10H, Cyclohexyl-H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 206.92, 170.42, 169.41, 164.36, 137.15, 135.10, 134.58, 131.92, 130.39, 129.47, 129.21, 129.04, 128.97, 127.45, 127.25, 125.74, 118.16, 83.86, 50.75, 37.94, 33.06, 30.86, 24.42, 21.71, 19.48; ESI-MS: calcd for $C_{24}H_{22}Cl_2O_4$ (M^+), 445.3; found, 445.9.

2-Oxo-3-*o*-tolyl-1-oxaspiro[4.5]dec-3-en-4-yl 2-o-tolylacetate (6l): This compound was obtained following the above-described method as colorless crystal, yield 72%, mp 103 - 105 °C; ^1H NMR (400 MHz, $CDCl_3$) δ 6.99-7.23 (m, 8H, Ph-H), 3.65 (s, 2H, CH_2CO), 2.14 (s, 3H, Ph-CH_3), 1.99 (s, 3H, Ph-CH_3), 1.61-1.78 (m, 10H, Cyclohexyl-H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.52, 169.60, 165.19, 137.09, 136.84, 130.59, 130.31, 130.11, 128.00, 127.39, 126.30, 125.72, 117.82, 83.76, 38.72, 32.94, 24.40, 21.70, 19.37, 19.04; ESI-MS: calcd for $C_{25}H_{26}O_4$ ($[M+1]^+$), 391.5; found, 391.9.

2-Oxo-3-*o*-tolyl-1-oxaspiro[4.5]dec-3-en-4-yl 3,4,5-trimethoxybenzoate (6m): This compound was obtained following the above-described method as white powder, yield 76%, mp 170 - 172 °C; ^1H NMR (400 MHz, $CDCl_3$) δ 7.08-7.15 (m, 6H, Ph-H), 3.85 (s, 3H, CH_3O), 3.81 (s, 6H, CH_3O), 2.24 (s, 3H, Ph-CH_3), 1.73-1.80 (m, 10H, Cyclohexyl-H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.52, 168.84, 159.51, 152.13, 142.74, 135.90, 129.38, 128.24, 127.93, 126.63, 124.81, 120.93, 116.77, 106.77, 82.99, 59.99, 55.37, 32.33, 23.42, 20.80, 18.70; ESI-MS: calcd for $C_{26}H_{28}O_7$ ($[M+1]^+$), 453.5; found, 454.0.

2-Oxo-3-*o*-tolyl-1-oxaspiro[4.5]dec-3-en-4-yl 4-fluorobenzoate (6n): This compound was obtained following the above-described method as white powder, yield 90%, mp 162 - 163 °C; ^1H NMR (400 MHz, $CDCl_3$) δ 7.97-8.00 (m, 2H, Ph-H), 7.11-7.19 (m, 6H, Ph-H), 2.28 (s, 3H, CH_3), 1.79-1.86 (m, 10H, Cyclohexyl-H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.31, 169.70, 159.76, 136.89, 133.09, 133.00, 130.33, 129.18, 128.92, 127.54, 125.74, 123.58, 117.72, 116.27, 116.05, 83.90, 33.24, 24.38, 21.75, 19.62; ESI-MS: calcd for $C_{23}H_{21}FO_4$ ($[M+1]^+$), 381.4; found, 381.9.

3-(4-Fluorophenyl)-2-oxo-1-oxaspiro[4.5]dec-3-en-4-yl 2,2-dimethylbutanoate (6o): This compound was obtained following the above-described method as white powder, yield 80%, mp 98 - 99 °C; ^1H NMR (400 MHz, $CDCl_3$) δ 7.51-7.55 (m, 2H, Ph-H), 7.07 (t, J = 8.6 Hz, 2H, Ph-H), 1.66-1.77 (m,

11H, Cyclohexyl-H and $\underline{CH_2}CH_3$), 1.25 (s, 6H, CH_3), 0.87 (t, 3H, J = 7.4 Hz, $CH_2\underline{CH_3}$); ESI-MS: calcd for $C_{21}H_{25}FO_4$ ($[M+1]^+$), 361.4; found, 361.9.

3-(4-Fluorophenyl)-2-oxo-1-oxaspiro[4.5]dec-3-en-4-yl methanesulfonate (6p): This compound was obtained following the above-described method as white powder, yield 75%, mp 111 - 112 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.58-7.62 (m, 2H, Ph-H), 7.14-7.18 (m, 2H, Ph-H), 3.09 (s, 3H, CH_3), 1.26-1.99 (m, 10H, Cyclohexyl-H); ESI-MS: calcd for $C_{16}H_{17}F O_5S$ ($[M+1]^+$), 341.4; found, 341.8.

Ethyl 3-(4-fluorophenyl)-2-oxo-1-oxaspiro[4.5]dec-3-en-4-yl carbonate (6q): This compound was obtained following the above-described method as white powder, yield 84%, mp 187 - 188 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.02-8.06 (m, 2H, Ph-H), 6.98-7.02 (m, 2H, Ph-H), 3.07 (q, J = 7.4 Hz, 2H, $\underline{CH_2}CH_3$), 1.57-2.14 (m, 10H, Cyclohexyl-H), 1.35 (t, J = 7.4 Hz, 3H, $CH_2\underline{CH_3}$); ESI-MS: calcd for $C_{18}H_{19}FO_5$ ($[M+1]^+$), 335.3; found, 335.9.

2-Oxo-3-phenyl-1-oxaspiro[4.5]dec-3-en-4-yl 2,2-dimethylbutanoate (6r): This compound was obtained following the above-described method as white powder, yield 83%, mp 105 - 106 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.26-7.46 (m, 5H, Ph-H), 1.56-1.75 (m, 12H, Cyclohexyl-H and $\underline{CH_2}CH_3$), 1.17 (s, 6H, CH_3), 0.79 (t, 3H, J = 7.4 Hz, $CH_2\underline{CH_3}$); ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.35, 169.03, 127.82, 127.40, 127.20, 127.10, 115.54, 82.63, 42.32, 32.16, 31.79, 23.58, 23.42, 20.73, 8.09; ESI-MS: calcd for $C_{21}H_{26}O_4$ ($[M+1]^+$), 343.4; found, 343.9.

2-Oxo-3-phenyl-1-oxaspiro[4.5]dec-3-en-4-yl methanesulfonate (6s): This compound was obtained following the above-described method as white powder, yield 92%, mp 112 - 113 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.50 (d, J = 6.8 Hz, 2H, Ph-H), 7.35-7.40 (m, 3H, Ph-H), 2.89 (s, 3H, CH_3), 1.17-1.94 (m, 10H, Cyclohexyl-H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.95, 169.38, 130.65, 130.02, 129.75, 128.37, 119.51, 85.23, 41.92, 33.68, 25.21, 22.69; ESI-MS: calcd for $C_{16}H_{18}O_5S$ ($[M+1]^+$), 323.4; found, 323.8.

Ethyl 2-oxo-3-phenyl-1-oxaspiro[4.5]dec-3-en-4-yl carbonate (6t): This compound was obtained following the above-described method as light yellow powder, yield 93%, mp 71 - 72 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.55 (d, J = 7.6 Hz, 2H, Ph-H), 7.25-7.35 (m, 3H, Ph-H), 4.07 (q, J = 7.2 Hz, 2H, $\underline{CH_2}$ CH_3), 1.66-1.79 (m, 10H, Cyclohexyl-H), 1.12 (t, J = 7.2 Hz, 3H, $CH_2\underline{CH_3}$); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.58, 169.27, 149.10, 128.96, 128.62, 128.07, 127.73, 114.70, 83.14, 66.30, 32.73, 24.34, 21.68, 13.84; ESI-MS: calcd for $C_{18}H_{20}O_5$ ($[M+1]^+$), 317.3; found, 371.9.

Biology assay. All bioassays were performed on representative test organisms reared in the laboratory. The bioassay was repeated at 25 ± 1 °C according to statistical requirements. All compounds were dissolved in EtOH (AP, Shanghai Chemical Reagent Co., Ltd., Shanghai, China) and diluted with distilled water containing Triton X-100 (0.1 μg/mL) to obtain series concentrations of 1000, 333, and 111 μg/mL and others for bioassays. For comparative purposes, spirodiclofen was tested under the same conditions.

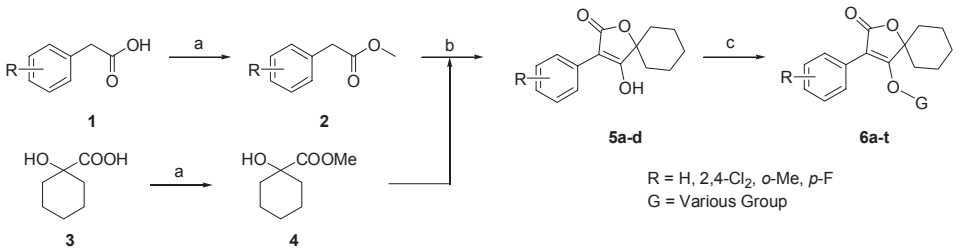
Results and Discussion

Synthesis of substituted spiro-tetronic acid derivatives 6a-t. Considering the convenient synthesis to construct the spirocycle scaffold, spirodiclofen analogues **6a-t** was prepared in a new and simple route, which is outlined in Scheme 1.

As shown in Scheme 1, the target compounds were conveniently obtained *via* three steps including esterification, heterocyclization and acylation reactions. The key intermediate 3-(substituted phenyl)-4-hydroxy-Δ^3-dihydrofuran-2-one **5a-d** was accessed conveniently by the modified one-pot heterocyclization reaction[27,28] from various substituted methyl 2-phenylacetate **2** and methyl 1-hydroxycyclohexanecarboxylate **4** in 70 - 83% yield, which involve a transesterification step, followed by the Dieckmann condensation. According the above-described experimental conditions, the key intermediates **5a-d** were achieved with a high yield and simple disposal compared the reported cyclization methods.[27,28] Meanwhile, this synthetic route leads to a convenient access to spirodiclofen analogues compared the reported traditional methods,[29-34] which may be propitious to large scale production. The structures of all newly synthesized compounds were characterized as 3-(substituted-phenyl)-1-oxaspiro[4.5]dec-3-en-2-one derivatives **6a-t** on the basis of satisfactory analytical and spectral data including ^{1}H NMR, ^{13}C NMR and ESI-MS.

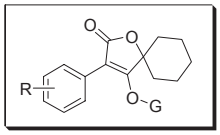
Biological activity evaluation. The bioactivity of synthesized spirodiclofen analogues against *Aphid*, *Plutella xylostella*, *Cotton bollworm* were evaluated according to a slightly modified FAO dip test,[35] and the herbicidal activities of compounds **6a-t** against *Cynodon dactylon* L., *Lemna minor Linn*, *Brassica chinensis* were tested using a previously reported procedure.[36] All the insecticidal and herbicidal activities of synthesized spirotetronic acid derivatives **6a-t** are listed in Table 1 and 2, respectively.

As we can see from Table 1, the preliminary assay showed that some of target molecules (such as compounds **6a**, **6b**, **6c**,



Scheme 1. Reagents and conditions: a. MeOH, Conc. H_2SO_4, reflux 6 - 8 h; b. EtONa or ^{t}BuOK, THF, reflux for 8 - 10 h; c. GCl, Et_3N, CH_2Cl_2, rt for 0.5 - 4 h

Table 1. Effects of substitution on insecticidal activity for compounds **6a-t** at different concentration (μg/mL)



Entry	Compd. No.	Substituents		Efficacy against *Aphid*			Efficacy against *Plutella xylostella*			Efficacy against *Cotton bollworm*		
		R	G	1000	333	111	1000	333	111	1000	333	111
1	**6a**	2,4-Cl_2	SO_2Me	0	0	0	9	9	9	0	0	0
2	**6b**	2,4-Cl_2	COOEt	0	0	0	9	9	9	0	0	0
3	**6c**	2,4-Cl_2	$COCH_2$-C_6H_5	0	0	0	9	9	9	0	0	0
4	**6d**	2,4-Cl_2	$COCH_2$-2,4-Cl_2-C_6H_3	0	0	0	9	9	9	0	0	0
5	**6e**	2,4-Cl_2	$COCH_2$-2-Me-C_6H_4	0	0	0	9	9	9	0	0	0
6	**6f**	2,4-Cl_2	CO-3,4,5-$(MeO)_3$-C_6H_2	0	0	0	9	0	0	0	0	0
7	**6g**	2,4-Cl_2	CO-4-F-C_6H_4	0	0	0	5	5	0	0	0	0
8	**6h**	*o*-Me	$COMe_2Et$	0	0	0	5	5	5	0	0	0
9	**6i**	*o*-Me	SO_2Me	0	0	0	9	9	9	0	0	0
10	**6j**	*o*-Me	COOEt	0	0	0	9	9	9	9	0	0
11	**6k**	*o*-Me	$COCH_2$-2,4-Cl_2-C_6H_3	0	0	0	5	5	5	0	0	0
12	**6l**	*o*-Me	$COCH_2$-2-Me-C_6H_4	0	0	0	9	9	9	0	0	0
13	**6m**	*o*-Me	CO-3,4,5-$(MeO)_3$-C_6H_2	0	0	0	0	0	0	0	0	0
14	**6n**	*o*-Me	CO-4-F-C_6H_4	0	0	0	5	5	5	0	0	0
15	**6o**	4-F	$COMe_2Et$	0	0	0	0	0	0	0	0	0
16	**6p**	4-F	SO_2Me	0	0	0	0	0	0	0	0	0
17	**6q**	4-F	COOEt	0	0	0	0	0	0	0	0	0
18	**6r**	H	$COMe_2Et$	0	0	0	0	0	0	0	0	0
19	**6s**	H	SO_2Me	0	0	0	0	0	0	0	0	0
20	**6t**	H	COOEt	0	0	0	0	0	0	0	0	0
21		**Spirodiclofen**		0	0	0	9	9	9	0	0	0

Scale: 0 (Weak, 0 - 40% mortality); 5 (Moderate, 41 - 80% mortality); 9 (Very good, 81 - 100% mortality).

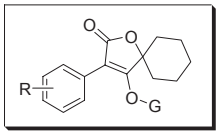
6d, **6e**, **6i**, **6j**, and **6l**) displayed obviously good and selective insecticidal activity against *Plutella xylostella* compared with spirodiclofen (at low concentration of 111 μg/mL, Table 1), but presented low activity against *Aphid* and *Cotton bollworm*. In addition, we introduced electron-withdrawing groups (such as halogen and fluorine atoms) and electron-donating substituents (Me) into the aromatic ring for exploring the influence of structural changes on activity. As described in Table 1, the different substituent at the periphery of the molecules **6a-t** can lead to the obviously different inhibition activities. Compounds containing 2,4-dichloro and *ortho*-methyl substituents **6a-n** and compounds bearing 4-fluoro and hydrogen substituents **6o-t** show a striking contrast, the latter almost lost activities at the same concentration level (Entry **15-20**, Table 1), which may be indicated position changes of the substituent within the aromatic ring dramatically affect the activity, suggesting that the presence of a group in the *ortho*-position of aromatic ring could introduce important steric and electronic effects.

Additionally, we can find from Table 1 that the insecticidal activities of the target compounds **6a-t** were also influenced by the different moiety attached to the oxygen atom of keto-enol cycle. As shown in Table 1, the function groups G are SO_2Me and COOEt, which are more favorable for insecticidal activities. However, when the groups G are substituted-benzoyl such as compounds **6f** and **6g**, **6m** and **6n**, decreases the potency.

From Table 2, which shows the structures and the herbicidal activity data, it can be seen that most of the synthesized spirodiclofen analogues **6a-t** showed good and selective efficacy against *Brassica chinensis* in higher concentrations (1.0×10^{-3}) except compounds **6f**, **6j**, **6k**, and **6m**. It is note worthy that most of target compounds including spirodiclofen exhibited low activity against *Lemna minor Linn.* and *Cynodon dactylon* L. As we know, *Lemna minor Linn.* and *Cynodon dactylon* L. are two difficult species in weed control. However, only the synthesized spirocyclic analogues **6j** revealed obviously better herbicidal performance against these two target species compared with commercial spirodiclofen, which indicated that compound **6j** might be used to further optimize for development novel weeds control agents.

The structure evolution here was to replace moiety A and moiety B, respectively (Figure 3). According to the results shown in Table 1, we can obtain the preliminary structure-activity profile for these spirocyclic tetronic acid analogues. As indicated in Figure 3, for moiety A, the compounds bearing 2,4-Cl_2-phenyl and 2-Me-phenyl substituents such as **6a-n** show better activities, however, when the substituents R are H and 4-F (such as **6o-t**), which present low efficacy. Moiety B was varied among such substituents as -COOEt, -SO_2Me, -$COMe_2Et$, substi-

Table 2. Effects of substitution on herbicidal activity for compounds **6a-t** at different concentration (μg/mL)



Entry	Compd. No.	Substituents		Efficacy against *Lemna minor Linn.*			Efficacy against *Cynodon dactylon* L.			Efficacy against *Brassica chinensis.*		
		R	G	1000	333	111	1000	333	111	1000	333	111
1	**6a**	2,4-Cl_2	SO_2Me	0	0	0	0	0	0	9	0	0
2	**6b**	2,4-Cl_2	COOEt	0	0	0	0	0	0	9	0	0
3	**6c**	2,4-Cl_2	$COCH_2$-C_6H_5	0	0	0	0	0	0	9	0	0
4	**6d**	2,4-Cl_2	$COCH_2$-2,4-Cl_2-C_6H_3	0	0	0	0	0	0	9	0	0
5	**6e**	2,4-Cl_2	$COCH_2$-2-Me-C_6H_4	0	0	0	0	0	0	9	0	0
6	**6f**	2,4-Cl_2	CO-3,4,5-$(MeO)_3$-C_6H_2	0	0	0	0	0	0	0	0	0
7	**6g**	2,4-Cl_2	CO-4-F-C_6H_4	0	0	0	0	0	0	9	0	0
8	**6h**	*o*-Me	$COCMe_2Et$	0	0	0	0	0	0	9	0	0
9	**6i**	*o*-Me	SO_2Me	0	0	0	0	0	0	9	0	0
10	**6j**	*o*-Me	COOEt	9	9	0	9	9	0	0	0	0
11	**6k**	*o*-Me	$COCH_2$-2,4-Cl_2-C_6H_3	0	0	0	0	0	0	0	0	0
12	**6l**	*o*-Me	$COCH_2$-2-Me-C_6H_4	0	0	0	0	0	0	9	0	0
13	**6m**	*o*-Me	CO-3,4,5-$(MeO)_3$-C_6H_2	0	0	0	0	0	0	0	0	0
14	**6n**	*o*-Me	CO-4-F-C_6H_4	0	0	0	0	0	0	9	0	0
15	**6o**	4-F	$COCMe_2Et$	0	0	0	0	0	0	9	0	0
16	**6p**	4-F	SO_2Me	0	0	0	0	0	0	9	0	0
17	**6q**	4-F	COOEt	0	0	0	0	0	0	9	0	0
18	**6r**	H	$COCMe_2Et$	0	0	0	0	0	0	0	0	0
19	**6s**	H	SO_2Me	0	0	0	0	0	0	9	0	0
20	**6t**	H	COOEt	9	0	0	0	0	0	9	0	0
21		**Spirodiclofen**		0	0	0	0	0	0	9	0	0

Scale: 0 (Weak, 0 - 40% mortality); 5 (Moderate, 41 - 80% mortality); 9 (Very good, 81 - 100% mortality).

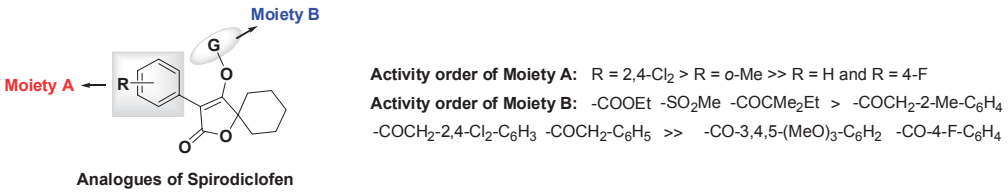


Figure 3. General structure-activity profile for the spirodiclofen analogues.

tuted phenylacetyl and substituted benzoyl, the results testified that compounds containing -COOEt, -SO_2Me, and -$COCMe_2Et$ group exhibited obviously high activity than compounds bearing substituted benzoyl moiety, which may be due to the steric size of phenyl is unfavourable for the binding to receptor.

Conclusion

In summary, we have described the convenient synthesis, biological activities, and SAR of a series of spirodiclofen analogues, and developed a practical and efficient procedure for the large scale synthesis of spiro-tetronic acid derivatives through the direct one-pot heterocyclization reaction of methyl 1-hydroxycyclohexanecarboxylate with various esters in THF. According the above-described experimental conditions, the cyclization reaction reached completion with a very high yield and simple disposal. The preliminary bioassay results indicated that some of the analogues exhibited good insecticidal and herbicidal activity compared to commercialized spirodiclofen. Furthermore, the understanding of structure-activity relationship (SAR) may be advantageous for further structure optimization and thereby provides some insight into the rational design of novel inhibitors targeting lipid biosynthesis.

Acknowledgments. This work was financially supported by the Hubei Biopesticide Engineering Research Centre. The authors also gratefully acknowledge the partial support from Hubei Agricultural Science Innovation Centre (2007-620-001-03). The authors also would like to thank many colleagues of the Hubei Biopesticide Engineering Research Centre and the Hubei Academy of Agricultural Sciences for valuable advice and assistance.

References

1. Wing, K. D.; Sacher, M.; Kagaya, Y.; Tsurubuchi, Y.; Mulderig, L.; Connair, M.; Schnee, M. *Crop Prot.* **2000**, *19*, 537.
2. Qian, X.; Lee, P. W.; Cao, S. *J. Agric. Food Chem.* **2010**, *58*, 2613.
3. Suzuki, J.; Ishida, T.; Kikuchi, Y.; Ito, Y.; Morikawa, C.; Tsukidate, Y.; Tanji, I.; Ota, Y.; Toda, K. *J. Pestic. Sci.* **2002**, *27*, 1.
4. Fischer, R.; Bretschneider, T.; Krueger, B. W.; Bachmann, J.; Erdelen, C.; Wachendorff-Neumann, U.; Santel, H. J.; Luerssen, K.; Schmidt, R. R. *DE 4216814*, 1993.
5. Wachendorff, U.; Brück, E.; Elbert, A.; Fischer, R.; Nauen, R.; Stumpf, N.; Tiemann, R. *Proc. Brighton Crop Protection Conference - Pests & Diseases* **2000**, *2A-6*, 53.
6. Fischer, R.; Benet-Buchholz, J. *Pflanzenschutz-Nachrichten Bayer* **2002**, *55*, 137.
7. Wachendorff, U.; Nauen, R.; Schnorbach, H. J.; Rauch, N.; Elbert, A. *Pflanzenschutz-Nachrichten Bayer* **2002**, *55*, 149.
8. Chang, J.-Y.; Lu, K.-H.; Hsieh, T.-T. *Plant Prot. Bull.* **2007**, *49*, 187.
9. Bretschneider, T.; Fischer, R.; Nauen, R. In *Modern Crop Protection Compounds*; Krämer, W., Schirmer, U. Eds.; Wiley-VCH Verlag GmbH & Co: Weinheim, Germany, pp 909-925, 2007.
10. Nauen, R.; Bretschneider, T.; Elbert, A.; Fisher, R.; Tiemann, R. *Pestic. Outlook* **2003**, *12*, 243.
11. Dekeyser, M. A. *Pest Manag. Sci.* **2005**, *61*, 103.
12. Bretschneider, T.; Benet-Buchholz, J.; Fischer, R.; Nauen, R. *Chimia* **2003**, *57*, 697.
13. Bretschneider, T.; Fischer, R.; Benet-Buchholz, J. *Pflanzenschutz-Nachrichten Bayer* **2005**, *58*, 307.
14. Nauen, R.; Bretschneider, T.; Brueck, E.; Elbert, A.; Reckmann, U.; Wachendorff, U.; Tiemann, R. *Proc. Brighton Crop Protection Conference - Pests & Diseases* **2002**, *2A-3*, 39.
15. Liu, T.-X. *Crop Prot.* **2004**, *23*, 505.
16. Bielza, P.; Fernández, E.; Grávalos, C.; Izquierdo, J. *BioControl* **2009**, *54*, 229.
17. Fischer, R.; Himmler, T.; Nauen, R.; Reckmann, U.; Schmitt, W. *Int. Plant Prot. Congress* (IPPC), Glasgow, 2007; p 100.
18. Van Waetermeulen, X.; Brück, E.; Elbert, A.; Fischer, R.; Krueger, S.; Kühnhold, J.; Nauen, R.; Niebes, J. F.; Reckmann, U.; Schnorbach, H. J.; Steffens, R. *Int. Plant Prot. Congress* (IPPC), Glasgow, 2007; p 60.
19. Nauen, R.; Bretschneider, T.; Elbert, A.; Fischer, R.; Reckmann, U.; van Waetermeulen, X. *11th IUPAC International Congress of Pesticide Chemistry*, Aug 6-11. Kobe, Japan, 2006; p 109.
20. Fischer, R.; Wei, H.-C. *Bayer CropScience J.* **2008**, *61*, 127.
21. Brück, E.; Elbert, A.; Fischer, R.; Krueger, S.; Kühnhold, J.; Klueken, A. M.; Nauen, R.; Niebes, J. F.; Reckmann, U.; Schnorbach, H. J.; Steffens, R.; van Waetermeulen, X. *Crop Prot.* **2009**, *28*, 838.
22. Van Pottelberge, S.; Van Leeuwen, T.; Khajehali, J.; Tirry, L. *Pest Manag. Sci.* **2009**, *65*, 358.
23. Nauen, R.; Stumpf, N.; Elbert, A. *Proc. Brighton Crop Protection Conference - Pests & Diseases* **2000**, *4D-9*, 453.
24. Rauch, N.; Nauen, R. *Pestic. Biochem. Phys.* **2003**, *74*, 91.
25. Nauen, R. *J. Pestic. Sci.* **2005**, *30*, 272.
26. Van Pottelberge, S.; Khajehali, J.; Van Leeuwen, T.; Tirry, L. *Exp. Appl. Acarol.* **2009**, *47*, 301.
27. Mallinger, A.; Le Gall, T.; Mioskowski, C. *Synlett* **2008**, 386.
28. Mallinger, A.; Le Gall, T.; Mioskowski, C. *J. Org. Chem.* **2009**, *74*, 1124.
29. Fischer, R.; Bretschneider, T.; Krüger, B. W.; Bachmann, J.; Erdelen, C.; Wachendorff-Neumann, U.; Santel, H. J.; Lürssen, K.; Schmidt, R. R. *US 5262383*, 1993.
30. Fischer, R.; Lehr, S.; Feucht, D.; Lösel, P.; Malsam, O.; Bojack, G.; Auler, T.; Hills, M. J.; Kehne, H.; Rosinger, C. H. *WO 2005066125*, 2005.
31. Fischer, R.; Lehr, S.; Feucht, D.; Lösel, P.; Malsam, O.; Bojack, G.; Auler, T.; Hills, M. J.; Kehne, H.; Rosinger, C. H. *US 20070244007*, 2007.
32. Lu, Y.; Xu, X.; Sun, N.; Zhao, J. *Fine Chem. Intermed.* **2009**, *39*, 19.
33. Zhao, J.; Ji, M.; Xu, X.; Cheng, J.; Zhu, G. *Chin. Chem. Lett.* **2009**, *20*, 1307.
34. Zhao, J.; Zhou, Y.; Xu, X.; Cheng, J.; Zhu, G. *Chin. J. Struct. Chem.* **2009**, *28*, 837.
35. FAO. Recommended methods for the detection and measurement of resistance of agricultural pests to pesticides: method for adult aphids; FAO method 17. *FAO Plant Prot. Bull.* **1979**, *18*, 6.
36. Xue, S.; Ke, S.; Yan, L.; Cai, Z.; Wei, Y. *J. Inorg. Biochem.* **2005**, *99*, 2276.