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Design, Multicomponent Synthesis, and Bioactivities of Novel Neonicotinoid Analogues with 1,4-Dihydropyridine Scaffold†

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Novel neonicotinoid analogues bearing a 1,4-dihydropridine scaffold were designed and synthesized by multicomponent reactions (MCRs) to enhance $\pi-\pi$ stacking. The synthesized compounds were identified by ^{1}H NMR, ^{13}C NMR, high-resolution mass spectroscopy, and elemental analysis. Bioassay tests showed that some of them exhibited high insecticidal activities against pea aphid (*Aphis craccivora*).

KEYWORDS: 1,4-Dihydropridine; MCRs; biological activity

INTRODUCTION

Acting on nicotinic acetylcholine receptor (nAChR) insect neuronal systems (*1*−*4*), imidacloprid (**1a**) (**Figure 1**) started the era of neonicotinoid insecticides, which have become a major insecticide class with high activities widely used for crop protection and veterinary pest control (*5*). As is well-known, 6-Cl-PMNI ((2-chloro-5-((2-(nitromethylene)imidazolidin-1-yl)methyl)pyridine, **1b**) (**Figure 1**) was obtained before imidacloprid and had higher binding affinity and activity than imidacloprid. Although its application was limited by its photoinstability (*6*) and inferior hydrophobicity (*7*), it is still an attractive molecule as lead compound for chemists worldwide (*8*, *9*).

In our previous work, numerous **1b** derivatives were synthesized. Compounds **2** (**Figure 2**) were obtained by introducing a tetrahydropyridine ring. The photostability of **2** was improved as desired (*10*). Afterward, compounds **3** (**Figure 2**) were reported, which had a bulky group conjugated system introduced by five-membered heterocycles. Some **3** compounds had higher activities than imidacloprid (*11*). The results encouraged us to consider further structure derivation of **1b**.

The research of binding model is always beneficial for molecular design. In the development of neonicotinoid insecticides, three kinds of action models were proposed by Yamamoto, Kagabu, and Casida successively (*12*−*14*). In 2007, Qian et al. suggested a new binding model of $\pi-\pi$ interaction induced by the hydrogen bond based on theory calculation (*15*). Then two kinds of AChBP−imidacloprid complex crystal structures were achieved by Casida and Sattelle (*16*, *17*), respectively. After that, Casida et al. designed compounds with extended N-substituted imine substituents and presumed the significance of the π-stacking formed by the amidine plane (*18*). Also, Kagabu et al.

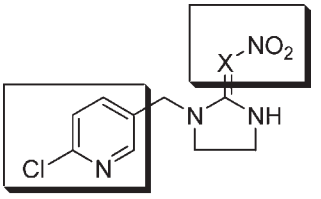

Figure 1. Structures of imidacloprid (**1a**) and **1b**.

proposed that a water bridge was formed between the neonicotinoid and the relevant amino acid at the ligand binding pocket. Novel compounds with methyl ketone, trifluoromethyl ketone, and epoxycyclopentylmethyl substituents were obtained and shown to enhance the binding affinity (*19*).

On the basis of the theory above, the influence factor of higher activities of compounds **3** was thought to be over. It was presumed that $\pi-\pi$ stacking was enhanced by the big conjugated system in compounds **3**, which might influence the binding model and improve the activities. Therefore, we try to further enhance $\pi-\pi$ stacking by introducing an aromatic bicyclic moiety.

It is well-known that the heteroaromatic moiety and nitro pharmacophore were very important in the structure of neonicotinoids, which were also included in **1b** (**Figure 3**). According to this analysis, reaction sites a and b (**Figure 3**) were to be modified, building target compound **6** (**Figure 2**) bearing a 1,4-dihydropyridine scaffold. The 1,4-dihydropyridine moiety was frequently used as a subunit of antiatherosclerotic, anticancer, and antidiabetic drugs (*20*−*22*). It can be achieved conveniently by multicomponent reactions (MCRs) using **1b**, aryl carbonyl compounds, and malononitrile as starting materials (*23*).

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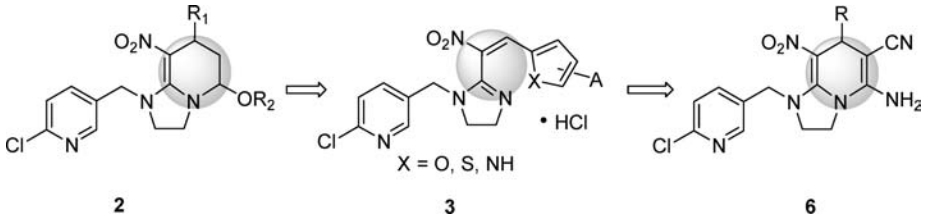


Figure 2. Development of novel neonicotinoid analogues in our group.

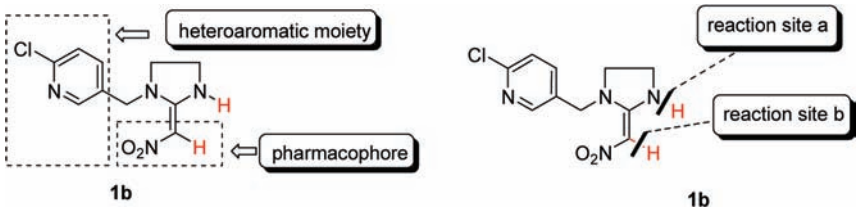


Figure 3. Pharmacophore and reaction sites at **1b**.

MATERIALS AND METHODS

Instruments. All melting points (mp) were obtained with a Büchi Melting Point B540 and are uncorrected. NMR spectra were recorded in DMSO-d_6 (^{1}H at 400 MHz and ^{13}C at 100 MHz) using TMS as the internal standard on a Bruker WP-400SY (400 MHz) spectrometer. Chemical shifts are reported in δ (parts per million) values. High-resolution mass spectra were recorded under electron impact (70 eV) condition using a MicroMass GCT CA 055 instrument. Combustion analyses for elemental composition were made with an Elementar vario EL III. Analytical thin-layer chromatography (TLC) was carried out on precoated plates (silica gel 60 F254), and spots were visualized with ultraviolet (UV) light. All other solvents and reagents were used as obtained from commercial sources without further purification.

General Procedure for the Preparation of Compounds 6a−p. A solution of malononitrile (15 mmol) in anhydrous alcohol (15 mL) was added dropwise to a solution of aryl aldehyde (15 mmol) in anhydrous alcohol (15 mL) at room temperature. After 5 min of stirring at room temperature, piperidine (0.1 mmol) used as catalyst was added dropwise. The resulting mixture was stirred for another 2 h, then **1b** (10 mmol) was added to the reaction mixture, refluxed for 15−20 h, and cooled to room temperature. Solid crystal products was filtered, washed with CH_2Cl_2, and dried to give desired products.

*Data for **6a**:* yield, 82%; mp 223.2−223.7 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 3.93−4.01 (m, 3H), 4.08−4.17 (m, 1H), 4.72 (s, 1H), 4.74 (s, 2H), 6.54 (s, 2H, NH_2), 6.98 (dd, J_1 = 1.4 Hz, J_2 = 7.8 Hz, 1H), 7.15−7.23 (m, 3H), 7.28 (d, J = 8.0 Hz, 1H), 7.64 (dd, J_1 = 2.6 Hz, J_2 = 8.2 Hz, 1H), 8.28 (d, J = 2.0 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 41.5, 43.9, 51.1, 51.5, 60.1, 106.2, 121.2, 124.4, 126.7, 127.0, 128.7, 131.4, 139.5, 144.8, 149.5, 149.6, 149.9, 152.9. Anal. Calcd for $C_{20}H_{17}ClN_6O_2$: C, 58.75; H, 4.19; N, 20.56. Found: C, 58.77; H, 3.89; N, 20.40.

*Data for **6b**:* yield, 78%; mp 253.5−253.9 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 4.02−4.15 (m, 4H), 4.70 (d, J = 15.6 Hz, 1H), 4.77 (d, J = 16.0 Hz, 1H), 4.81 (s, 1H), 6.70 (s, 2H, NH_2), 7.21 (d, J = 8.0 Hz, 1H), 7.22 (d, J = 8.8 Hz, 2H), 7.60 (dd, J_1 = 1.6 Hz, J_2 = 8.0 Hz, 1H), 8.05 (d, J = 8.4 Hz, 2H), 8.17 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 41.6, 43.9, 51.3, 51.8, 58.5, 105.6, 120.8, 124.0, 124.3, 128.1, 131.3, 139.5, 146.6, 149.5, 149.9, 150.0, 152.1, 152.2. Anal. Calcd for $C_{20}H_{16}ClN_7O_4$: C, 52.93; H, 3.55; N, 21.60. Found: C, 52.90; H, 3.39; N, 21.48.

*Data for **6c**:* yield, 59%; mp 244.3−245.1 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 4.01−4.15 (m, 4H), 4.69 (d, J = 16.0 Hz, 1H), 4.74 (d, J = 16.0 Hz, 1H), 4.87 (s, 1H), 6.73 (s, 2H, NH_2), 7.17 (d, J = 8.4 Hz, 1H), 7.44 (d, J = 7.6 Hz, 1H), 7.51 (t, J = 7.8 Hz, 1H), 7.60 (dd, J_1 = 2.4 Hz, J_2 = 8.4 Hz, 1H), 7.88 (t, J = 1.8 Hz, 1H), 8.07 (dd, J_1 = 1.2 Hz, J_2 = 8.0 Hz, 1H), 8.17 (d, J = 2.4 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 41.3, 44.0, 51.4, 51.5, 58.5, 105.8, 120.9, 121.4, 122.2, 124.1, 130.4, 131.2, 133.7, 139.4, 146.9, 148.1, 149.5, 149.8, 150.2, 152.3. Anal. Calcd for $C_{20}H_{16}ClN_7O_4$: C, 52.93; H, 3.55; N, 21.60. Found: C, 52.82; H, 3.22; N, 21.38.

*Data for **6d**:* yield, 41%; mp 224.6−245.8 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 4.00−4.13 (m, 4H), 4.73 (dd, J_1 = 16.0 Hz, J_2 = 20.0 Hz, 2H), 4.75 (s, 1H), 6.67 (s, 2H, NH_2), 7.16 (d, J = 8.4 Hz, 1H), 7.24 (d, J = 8.0 Hz, 2H), 7.63 (dd, J_1 = 2.4 Hz, J_2 = 8.2 Hz, 1H), 7.64 (d, J = 8.4 Hz, 2H), 8.18 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 41.8, 43.9, 51.4, 51.7, 58.6, 105.7, 109.9, 119.4, 120.9, 124.3, 127.9, 131.3, 132.7, 139.4, 149.5, 149.9, 149.9, 150.1, 152.3. Anal. Calcd for $C_{21}H_{16}ClN_7O_2$: C, 58.14; H, 3.72; N, 22.60. Found: C, 58.00; H, 3.50; N, 22.49.

*Data for **6e**:* yield, 65%; mp 246.4−247.5 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 3.99−4.14 (m, 4H), 4.73 (s, 2H), 4.78 (s, 1H), 6.65 (s, 2H, NH_2), 7.24 (d, J = 8.0 Hz, 1H), 7.36 (d, J = 7.2 Hz, 1H), 7.43 (t, J = 7.2 Hz, 1H), 7.50 (s, 1H), 7.62 (s, 2H), 8.22 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 41.6, 43.9, 51.5, 51.6, 58.9, 105.7, 111.8, 119.4, 120.9, 124.2, 129.9, 130.5, 131.0, 131.3, 132.2, 139.4, 146.3, 149.5, 149.8, 149.9, 152.4. HRMS (ESI) calcd for $C_{21}H_{16}ClN_7O_2$ (M + Na), 456.0952; found, 456.0970.

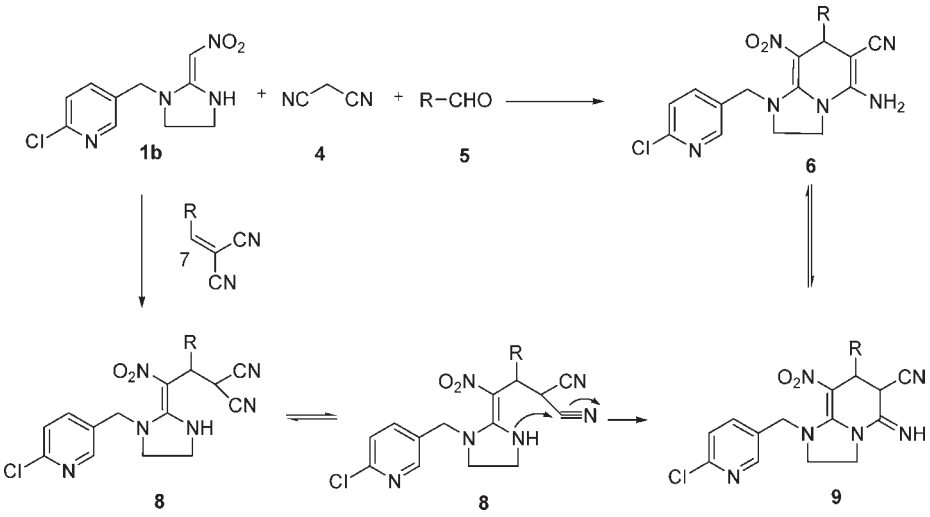
*Data for **6f**:* yield, 61%; mp 191.5−192.0 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 3.97−4.16 (m, 4H), 4.74 (s, 3H), 6.60 (s, 2H, NH_2), 7.10 (d, J = 8.8 Hz, 2H), 7.18 (d, J = 8.0 Hz, 2H), 7.28 (d, J = 8.4 Hz, 1H), 7.65 (dd, J_1 = 2.6 Hz, J_2 = 8.2 Hz, 1H), 8.26 (d, J = 2.0 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 41.1, 43.9, 51.4, 51.5, 59.5, 105.9, 119.3, 121.1, 121.2, 121.8, 124.3, 128.6, 131.4, 139.5, 144.1, 147.4, 149.6, 149.7, 149.9, 152.6. HRMS (ESI) calcd for $C_{21}H_{16}ClF_3N_6O_3$ (M + Na), 515.0822; found, 515.0832. Anal. Calcd for $C_{21}H_{16}ClF_3N_6O_3$: C, 51.18; H, 3.27; N, 17.05. Found: C, 50.43; H, 3.08; N, 16.59.

*Data for **6g**:* yield, 64%; mp 237.7−238.2 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 3.91−4.15 (m, 4H), 4.73 (s, 3H), 6.62 (s, 2H, NH_2), 6.97 (d, J = 7.6 Hz, 1H), 7.17 (t, J = 7.8 Hz, 1H), 7.26 (t, J = 1.8 Hz, 1H), 7.28 (d, J = 8.0 Hz, 1H), 7.37−7.39 (m, 1H), 7.65 (dd, J_1 = 2.4 Hz, J_2 = 8.4 Hz, 1H), 8.25 (d, J = 2.0 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 41.4, 43.9, 51.1, 51.5, 59.2, 105.8, 121.0, 122.1, 124.3, 126.0, 129.6, 130.0, 131.0, 131.3, 139.4, 147.5, 149.5, 149.8, 149.9, 152.7. Anal. Calcd for $C_{20}H_{16}BrClN_6O_2$: C, 49.25; H, 3.31; N, 17.23. Found: C, 49.19; H, 3.01; N, 17.05.

*Data for **6h**:* yield, 49%; mp 261.7−262.1 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 4.01−4.14 (m, 4H), 4.72 (d, J = 15.6 Hz, 1H), 4.78 (d, J = 16.0 Hz, 1H), 5.14 (s, 1H), 6.53 (s, 2H, NH_2), 6.98 (d, J = 8.4 Hz, 1H), 7.20 (dd, J_1 = 2.0 Hz, J_2 = 8.0 Hz, 1H), 7.42 (d, J = 8.0 Hz, 1H), 7.46 (d, J = 2.0 Hz, 1H), 7.77 (dd, J_1 = 2.2 Hz, J_2 = 8.2 Hz, 1H), 8.31 (d, J = 1.6 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 39.7, 43.9, 51.3, 51.8, 59.1, 105.5, 120.4, 124.5, 127.9, 129.1, 131.4, 131.6, 132.1, 133.0, 139.8, 141.5, 149.5, 149.7, 150.0, 152.7. Anal. Calcd for $C_{20}H_{15}Cl_3N_6O_2$: C, 50.28; H, 3.16; N, 17.59. Found: C, 50.36; H, 2.90; N, 17.38.

*Data for **6i**:* yield, 65%; mp 252.1−253.0 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 3.96−4.18 (m, 4H), 4.77 (t, J = 17.0 Hz, 2H), 4.89 (s, 1H), 6.56 (s, 2H, NH_2), 7.01 (t, J = 8.2 Hz, 1H), 7.09 (d, J = 8.0 Hz, 1H), 7.23 (d, J = 10.4 Hz, 1H), 7.36 (d, J = 8.0 Hz, 1H), 7.72 (d, J = 8.0 Hz, 1H), 8.26 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 43.9, 51.4, 51.5, 56.5,

Scheme 1. Target Compound Synthesis via Aryl Aldehyde-Based Muliticomponet Reactions



58.9, 104.9, 116.3, 116.5, 120.6, 124.4, 124.9, 124.9, 130.6, 130. 8, 131.2, 131.2, 131.3, 132.3, 132.4, 139.6, 149.5, 149.7, 150.1, 152.5, 158.8, 160.3. HRMS (ESI) calcd for $C_{20}H_{15}Cl_2FN_6O_2$ (M + Na), 483.0515; found, 483.0523.

*Data for **6j**:* yield, 72%; mp 265.4−265.8 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 3.98−4.05 (m, 3H), 4.09−4.12 (m, 1H), 4.71 (d, J = 15.6 Hz, 1H), 4.76 (d, J = 15.6 Hz, 1H), 5.12 (s, 1H), 6.53 (s, 2H, NH_2), 6.89 (d, J = 8.4 Hz, 1H), 7.31 (dd, J_1 = 2.0 Hz, J_2 = 8.0 Hz, 1H), 7.42 (d, J = 8.0 Hz, 1H), 7.56 (d, J = 2.0 Hz, 1H), 7.76 (dd, J_1 = 2.4 Hz, J_2 = 8.4 Hz, 1H), 8.30 (d, J = 2.0 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 40.1, 43.9, 51.3, 51.8, 59.0, 105.5, 120.3, 120.4, 124.5, 130.8, 131.4, 131.8, 131.9, 133.2, 139.8, 141.9, 149.5, 149.7, 150.0, 152.7. HRMS (ESI) calcd for $C_{20}H_{15}BrCl_2N_6O_2$ (M + Na), 542.9715; found, 542.9741.

*Data for **6k**:* yield, 40%; mp 202.6−203.2 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 2.23 (s, 3H), 3.90−4.01 (m, 3H), 4.14−4.20 (m, 1H), 4.73 (d, J = 16.0 Hz, 1H), 4.78 (d, J = 15.6 Hz, 1H), 4.84 (s, 1H), 6.49 (s, 2H, NH_2), 6.82 (dd, J_1 = 2.0 Hz, J_2 = 7.6 Hz, 1H), 6.90 (dd, J_1 = 8.6 Hz, J_2 = 10.6 Hz, 1H), 6.99−7.02 (m, 1H), 7.35 (d, J = 8.4 Hz, 1H), 7.70 (dd, J_1 = 2.2 Hz, J_2 = 8.2 Hz, 1H), 8.29 (d, J = 2.4 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 20.7, 37.6, 44.0, 50.8, 51.5, 59.7, 105.0, 115.5, 115.7, 120.7, 124.4, 129.2, 129.3, 130.0, 130.1, 131.0, 131.1, 131.3, 133.6, 133.6, 139.6, 149.3, 149.7, 149.9, 152.9, 157.5, 159.9. Anal. Calcd for $C_{21}H_{18}ClFN_6O_2$: C, 57.21; H, 4.12; N, 19.06. Found: C, 56.98; H, 3.78; N, 18.79.

*Data for **6l**:* yield, 32%; mp 257.4−258.4 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 2.39 (s, 3H), 3.97−4.09 (m, 4H), 4.72 (d, J = 16.0 Hz, 1H), 4.76 (d, J = 16.8 Hz, 1H), 4.93 (d, J = 1.6 Hz, 1H), 6.51 (s, 2H, NH_2), 6.75 (d, J = 9.6 Hz, 1H), 7.37 (d, J = 8.0 Hz, 1H), 7.41 (d, J = 7.2 Hz, 1H), 7.81 (dd, J_1 = 2.6 Hz, J_2 = 8.2 Hz, 1H), 8.27 (d, J = 2.0 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 18.3, 38.1, 43.9, 51.4, 52.0, 59.9, 105.5, 105.7, 106.4, 115.4, 115.6, 120.8, 124.3, 131.5, 132.9, 133.0, 134.3, 139.7, 146.4, 146.5, 149.0, 149.6, 150.0, 152.6, 156.4, 158.8. Anal. Calcd for $C_{21}H_{17}BrFN_6O_2$: C, 48.53; H, 3.30; N, 16.17. Found: C, 48.28; H, 2.98; N, 16.02.

*Data for **6m**:* yield, 63%; mp 236.2−236.9 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 2.41 (s, 3H), 3.96−4.14 (m, 4H), 4.74 (d, J = 16.0 Hz, 1H), 4.79 (d, J = 15.6 Hz, 1H), 4.99 (d, J = 2.0 Hz, 1H), 6.48 (s, 2H, NH_2), 6.59 (dd, J_1 = 2.4 Hz, J_2 = 10.0 Hz, 1H), 6.85−6.90 (m, 1H), 7.09−7.12 (m, 1H), 7.39 (d, J = 8.4 Hz, 1H), 7.82 (dd, J_1 = 2.4 Hz, J_2 = 8.0 Hz, 1H), 8.31 (d, J = 2.0 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 18.7, 38.3, 43.9, 51.1, 52.0, 60.6, 106.6, 113.4, 113.7, 113.8, 114.0, 120.9, 124.4, 130.7, 130.7, 131.5, 131.7, 131.8, 139.6, 146.5, 146.6, 149.0, 149.6, 149.9, 152.9, 160.4, 162.8. HRMS (ESI) calcd for $C_{21}H_{18}ClFN_6O_2$ (M + Na), 463.1061; found, 463.1056.

*Data for **6n**:* yield, 26%; mp 236.1−236.8 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 4.07 (m, 4H), 4.75 (s, 2H), 5.03 (s, 1H), 6.54 (s, 2H, NH_2), 7.03 (d, J = 6.0 Hz, 1H), 7.42−7.46 (m, 2H), 7.62 (d, J = 1.6 Hz, 1H), 7.77 (dd, J_1 = 2.4 Hz, J_2 = 8.0 Hz, 1H), 8.30 (d, J = 1.6 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 37.9, 43.9, 51.7, 51.8, 60.3, 106.7, 120.1, 122.5, 124.5, 125.5, 125.6, 131.5, 131.9, 132.2, 133.2, 139.8, 144.4, 149.7, 149.7, 150.1, 152.3. HRMS (ESI) calcd for $C_{21}H_{15}Cl_2F_3N_6O_2$ (M + H), 511.0664; found, 511.0677.

*Data for **6o**:* yield, 10%; mp 217.1−217.3 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 3.73 (s, 3H), 3.89−4.41 (m, 4H), 4.49 (s, 1H), 4.66 (s, 1H), 4.75 (d, J = 2.4 Hz, 1H), 6.51 (s, 2H, NH_2), 6.75−6.77 (m, 2H), 6.87−6.90 (m, 1H), 7.30 (d, J = 8.0 Hz, 1H), 7.64 (dd, J_1 = 2.8 Hz, J_2 = 8.0 Hz, 1H), 8.27 (d, J = 2.4 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 42.9, 43.9, 45.7, 48.4, 51.1, 51.4, 55.5, 60.5, 106.6, 114.1, 124.4, 124.8, 127.7, 131.5, 136.8, 139.5, 139.7, 149.4, 149.6, 149.9, 158.4. HRMS (ESI) calcd for $C_{21}H_{19}ClN_6O_3$ (M + H), 439.1285; found, 439.1274.

*Data for **6p**:* yield, 19%; mp 225.1−226.2 °C; ^{1}H NMR (400 MHz, DMSO-d_6), δ 2.32 (d, J = 1.6 Hz, 3H), 3.98−4.15 (m, 4H), 4.76 (s, 2H), 5.02 (s, 1H), 6.48 (s, 2H, NH_2), 6.59 (d, J = 7.6 Hz, 1H), 6.95−6.99 (m, 1H), 7.01−7.04 (m, 1H), 7.40 (d, J = 8.4 Hz, 1H), 7.76 (dd, J_1 = 2.4 Hz, J_2 = 8.0 Hz, 1H), 8.32 (d, J = 2 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6), δ 10.1, 10.2, 37.7, 43.9, 51.2, 51.8, 60.7, 106.8, 112.9, 113.2, 120.9, 121.7, 121.8, 123.3, 124.5, 127.6, 127.6, 131.5, 139.7, 148.9, 149.7, 149.9, 152.8, 159.2, 161.6. HRMS (ESI) calcd for $C_{21}H_{18}ClFN_6O_2$ (M + H), 441.1242; found, 441.1227.

Biological Assay. All compounds were dissolved in acetone and diluted with water containing Triton X-100 (0.1 mg/L) to obtain series concentrations of 500.0, 50.0, and 25.0 mg/L and others for bioassays.

As previously tested (*10*), cowpea aphids (*Aphis craccivora*) were dipped according to a slightly modified FAO dip test (*24*). Tender shoots of soybean with 40−60 healthy apterous adults were dipped in diluted solutions of the chemicals containing Triton X-100 (0.1 mg/L) for 5 s, the superfluous fluid was removed, and the shoots were placed in the conditioned room (25 ± 1 °C, 50% RH). Water containing Triton X-100 (0.1 mg L^{-1}) was used as control. Mortality was assessed after 24 h; the control mortality was 4.3%. Each treatment had three repetitions, and the data were corrected and subjected to probit analysis using SPSS software.

RESULTS AND DISCUSSION

Synthesis. The neonicotinoid analogues bearing a 1,4-dihydropyridine scaffold (**6a**−**p**) were synthesized as shown in **Scheme 1**. The assembly of **6** can be explained via the initial Michael addition of **7** to the ylidenic bond in **1b**, leading to the formation of an acyclic intermediate **8**, which cyclized into the intermediate **9** via nucleophilic attack of an NH group on a cyano carbon, followed by tautomerization to the final product **6**. The formation of **7** is via Knoevenagel condensation reaction of malononitrile **4** and appropriate aromatic aldehyde **5**.

The search for one-pot condensation reaction parameters started with nitromethylene (**1b**), malononitrile (**4**), and *p*-nitrobenzaldehyde (**5a**) in molar ratios from 1:1:1 to 1:2:2 in boiling ethanolic piperidine from 2 to 17 h; desirable product could be obtained up to 78% yield. However, no substantial gain in the

Table 1. Insecticidal Activities of Neonicotinoid Analogues **6a**−**p** Bearing a 1,4-Dihydropyridine Scaffold against Pea Aphid (*Aphis craccivora*)

compd	R	mortality (%) in vivo at 1 day (500 mg L^{-1})	LC_{50} (mmol L^{-1})
6a	Ph	83.1	nt[a]
6b	4-NO_2−C_6H_4	77	nt
6c	3-NO_2−C_6H_4	91.9	0.07903
6d	4-CN−C_6H_4	93.7	0.1746
6e	3-CN−C_6H_4	80	nt
6f	4-CF_3O−C_6H_4	100	0.09797
6g	3-Br−C_6H_4	91.8	0.19702
6h	2,4-DiCl−C_6H_3	93.4	0.1326
6i	2-F−4-Cl−C_6H_3	88	nt
6j	2-Cl−4-Br−C_6H_3	85	nt
6k	2-F−4-Me−C_6H_3	72.9	nt
6l	2-Br−4-F−5-Me−C_6H_2	75.9	nt
6m	2-CH_3−5-F−C_6H_3	100	0.09614
6n	2-CF_3−4-Cl−C_6H_3	100	0.06159
6o	4-OCH_3−C_6H_4	100	0.00975
6p	2-CH_3−3-F−C_6H_3	100	0.00345
imidacloprid		100[b]	0.03502[b]

[a] Not tested. [b] Data from Shao et al. (*11*).

product yield was observed with increasing **4** and **5a** loading from 150 to 200 mol %. Therefore, the optimum usage was fixed at 1:1.5:1.5 (**1b**/**2**/**3a**) for all subsequent experiments. To evaluate the novel analogues, compounds **6b**−**p** (**Table 1**) were achieved from 15 additional aryl aldehydes (**5b**−**p**) using the optimized reaction conditions. Unfortunately, no desirable product could be obtained from alkyl aldehydes such as butyl aldehyde or heterocyclic aldehydes such as pyridine aldehyde, furaldehyde, and thienyl aldehyde. The structures of the title compounds were well characterized by ^{1}H NMR, ^{13}C NMR, HRMS, and elemental analysis.

Biological Activities. Compounds **6c**, **6d**, **6f**, **6g**, **6h**, **6m**, **6n**, **6o**, and **6p** exhibited good insecticidal activity against pea aphid (**Table 1**) and had >90% mortality at 500 mg L^{-1}. The LC_{50} values of those compounds were 0.07903, 0.1746, 0.09797, 0.19702, 0.1326, 0.09614, 0.06159, 0.00975, and 0.00345 mmol L^{-1}, respectively, whereas that of imidacloprid was 0.03502 mmol L^{-1}. It was concluded that compounds **6o** and **6p** showed 3.6- and 10.2-fold potency compared with imidacloprid. For the effect of substituents at the phenyl group, it was observed that compounds demonstrated good activities with either electron-withdrawing or electron-donating groups. Nevertheless, an electron-donating group is favorable for high activities from the data analysis present. Further study is underway.

In conclusion, various neonicotinoid analogues bearing a 1,4-dihydropridine scaffold were synthesized successfully via multicomponent reactions. Most of compounds showed good insecticidal activities at 500 mg L^{-1}, which implied a new method of molecular design, which is to obtain highly active compounds by enhancing $\pi-\pi$ stacking.

Supporting Information Available: ^{1}H NMR and ^{13}C NMR spectra of compounds **6**. This material is available free of charge via the Internet at http://pubs.acs.org.

LITERATURE CITED

(1) Liu, M.; Casida, J. E. High affinity binding of [^{3}H] imidacloprid in the insect acetylcholine receptor. *Pestic. Biochem. Physiol.* **1993**, *46*, 40–46.

(2) Mori, K.; Okumoto, T.; Kawahara, N.; Ozoe, Y. Interaction of dinotefuran and its analogues with nicotinic acetylcholine receptors of cockroach nerve cords. *Pest Manage. Sci.* **2001**, *58*, 190–196.

(3) Tomizawa, M.; Casida, J. E. Neonicotinoid insecticide toxicology: mechanisms of selective action. *Annu. Rev. Pharmacol. Toxicol.* **2005**, *45*, 247–268.

(4) Matsuda, K.; Shimomura, M.; Ihara, M.; Akamatsu, M.; Sattelle, D. B. Neonicotinoids show selective and diverse actions on their nicotinic receptor targets: electrophysiology, molecular biology, and receptor modeling studies. *Biosci., Biotechnol., Biochem.* **2005**, *69*, 1442–1452.

(5) Kagabu, S. Chloronicotinyl insecticides−discovery, application and future perspective. *Rev. Toxicol.* **1997**, *1*, 75–129.

(6) Okazawa, A.; Akamatsu, M.; Ohoka, A.; Nishiwaki, H.; Cho, W. J.; Nakagawa, Y.; Nishimura, K.; Ueno1, T. Prediction of the binding mode of imidacloprid and related compounds to housefly head acetylcholine receptors using three-dimensional QSAR analysis. *Pestic. Sci.* **1998**, *54*, 134–144.

(7) Nishiwaki, H.; Nakagawa, Y.; Ueno, T.; Kagabu, S.; Nishimura, K. Insecticidal and binding activities of N3-substituted imidacloprid derivatives against the housefly *Musca domestica* and the α-bungarotoxin binding sites of nicotinic acetylcholine receptors. *Pest Manage. Sci.* **2001**, *57*, 810–814.

(8) Kagabu, S.; Medej, S. Chloronicotinyl insecticides. Part VI. Stability comparison of imidacloprid and related compounds under simulated sunlight, hydrolysis conditions, and to oxygen. *Biosci., Biotechnol., Biochem.* **1995**, *59*, 980–985.

(9) Yamamoto, I.; Tomizawa, M.; Satio, T.; Miyamoto, T.; Walcott, E. C.; Sumikawa, W. Structural factors contributing to insecticidal and selective actions of neonicotinoids. *Arch. Insect. Biochem. Physiol.* **1998**, *37*, 24–32.

(10) Tian, Z. Z.; Shao, X. S.; Li, Z.; Qian, X. H.; Huang, Q. C. Synthesis, insecticidal activity and QSAR of novel nitromethylene neonicotinoids with tetrahydropyridine fixed *cis* configuration and exo-ring ether modification. *J. Agric. Food Chem.* **2007**, *55*, 2288–2292.

(11) Shao, X. S.; Li, Z.; Qian, X. H.; Xu, X. Y. Design, synthesis, and insecticidal activities of novel analogues of neonicotinoids: replacement of nitromethylene with nitroconjugated system. *J. Agric. Food Chem.* **2009**, *57*, 951–957.

(12) Tomizawa, M.; Yamamoto, I. Structure−activity relationships of nicotinoids and imidacloprid analogs. *Nihon Noyaku Gakkaishi* **1993**, *18*, 91–98.

(13) Kagabu, S.; Matsuno, H. Chloronicotinyl insecticides. 8. Crystal and molecular structures of imidacloprid and analogous compounds. *J. Agric. Food Chem.* **1997**, *45*, 276–281.

(14) Tomizawa, M.; Lee, D. L.; Casida, J. E. Neonicotinoid insecticides: molecular features conferring selectivity for insect versus mammalian nicotinic receptors. *J. Agric. Food Chem.* **2000**, *48*, 6016–6024.

(15) Wang, Y. L.; Cheng, J. G.; Qian, X. H.; Li, Z. Actions between neonicotinoids and key residues of insect nAChR based on an ab initio quantum chemistry study: hydrogen bonding and cooperative $\pi-\pi$ interaction. *Bioorg. Med. Chem.* **2007**, *15*, 2624–2630.

(16) Tomizawa, M.; Talley, T. T.; Maltby, D.; Durkin, K. A.; Medzihradszky, K. F.; Burlingame, A. L.; Taylor, P.; Casida, J. E. Mapping the

elusive neonicotinoid binding site. *Proc. Natl. Acad. Sci. U.S.A.* **2007**, *104*, 9075–9080.
(17) Ihara, M.; Okajima, T.; Yamashita, A.; Oda, T.; Hirata, K.; Nishiwaki, H.; Morimoto, T.; Akamatsu, M.; Ashikawa, Y.; Kuroda, S.; Mega, R.; Kuramitsu, S.; Sattelle, D. B.; Matsuda, K. Crystal structures of *Lymnaea stagnalis* AChBP in complex with neonicotinoid insecticides imidacloprid and clothianidin. *Invert. Neurosci.* **2008**, *8*, 71–81.
(18) Tomizawa, M.; Kagabu, S.; Ohno, I.; Durkin, K. A.; Casida, J. E. Potency and selectivity of trifluoroacetylimino and pyrazinoylimino nicotinic insecticides and their fit at a unique binding site niche. *J. Med. Chem.* **2008**, *51*, 4213–4218.
(19) Ohno, I.; Tomizawa, M.; Durkin, K. A.; Casida, J. E.; Kagabu, S. Neonicotinoid substituents forming a water bridge at the nicotinic acetylcholine receptor. *J. Agric. Food Chem.* **2009**, *57*, 2436–2440.
(20) Sausins, A.; Duburs, G. Synthesis of 1,4-dihydropyridines by cyclocondensation reactions. *Heterocycles* **1988**, *27*, 269–289.
(21) Mager, P. P.; Coburn, R. A.; Solo, A. J.; Triggle, D. J.; Rothe, H. QSAR, diagnostic statistics and molecular modeling of 1,4-dihydropyridine calcium antagonists: a difficult road ahead. *Drug Design Discov.* **1992**, *8*, 273–289.
(22) Manhold, R.; Jablonka, B.; Voigdt, W.; Schoenfinger, K. F.; Schravan, K. Calcium- and calmodulin-antagonism of elnadipine derivatives: a comparison of structure−activity relationships. *Eur. J. Med. Chem.* **1992**, *27*, 229–235.
(23) Sharanin, Y. A.; Baskakov, Y. A.; Abramenko, Y. T.; Putsykin, Y. G.; Vasil'ev, A. F.; Nazarova, E. B. Cyclization of nitriles. III. Synthesis of *o*-cyanoanilines using the Thorpe reaction. *Zh. Org. Khim.* **1980**, *16*, 2192–2204.
(24) 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*, 6−9.

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