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Synthesis and Bioactivity Study of 2-Acylamino-Substituted *N*′-Benzylbenzohydrazide Derivatives

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ABSTRACT: The discovery of new safe and effective pesticides is one of the main means of providing eco-friendly agricultural agents for modern crop protection. To identify new biological molecules based of the anthranilic diamide skeleton of the novel pesticide chlorantraniliprole, which acts on the ryanodine receptor and functional groups in acyl hydrazine insect growth regulators, more than 40 new compounds of 2-acylamino-substituted *N*′-benzylbenzohydrazide derivatives were designed and synthesized. The structures of the new compounds were characterized using ^{1}H nuclear magnetic resonance (NMR), high-resolution mass spectrometry (HRMS), or electron impact mass spectrometry (EI-MS), and their biological activities at a concentration of 600 mg L^{-1} were determined against cotton aphid (*Aphis gossypii* Glover), carmine spider mite (*Tetranychus cinnabarinus*), and diamondback moth (*Plutella xylostella*). The results of a preliminary assay showed that compounds **6a-I-2** and **6d-III-4** maintained the lethal activity of anthranilic diamide against *P. xylostella*; compounds **6c-II-4**, **6d-I-7**, **6d-II-1**, and **6d-III-5** exhibited good lethal activity against *A. gossypii*; and compounds **6a-II-1**, **6a-III-1**, **6b-I-7**, **6c-I-1**, and **6c-III-5** retained promising larvicidal activities against *T. cinnabarinus*. In subsequent further tests against *T. cinnabarinus*, compounds **6a-II-1**, **6a-III-1**, **6c-I-1**, and **6c-III-5** showed an LC_{50} value of <90 mg L^{-1}; especially, the LC_{50} of compound **6a-III-1** was only 27.9 mg L^{-1}. In conclusion, the introduction of the functional fragment-substituted acyl hydrazine improved the acaricidal activity of the anthranilic diamide skeleton, and the halogen atom at X position and the methyl group at R_1 play crucial roles in the biological activities of the compounds.

KEYWORDS: *2-acylamino-substituted N′-benzylbenzohydrazide derivatives, synthesis, insecticide*

INTRODUCTION

Control of pest insects with the application of chemical insecticides is an important aspect in modern agricultural practices. However, broad application of chemicals over the long term induces resistance in the treated pest insects by improving their tolerance[1] and causing changes in their behavior.[2] In addition, there are concerns regarding the impact of residual compounds and environmental issues with the use of chemical insecticides; hence, some existing commercial products might not meet the requirement of pest insect control in the future.

Controlling the development of insecticide resistance in pests and limiting the usage of synthetic chemicals for maintaining an eco-friendly environment necessitates the identification of alternative molecules with new structural features and action modes. For instance, the recently commercialized insecticides flubendiamide (Nihon Nohyaku Co., Ltd., and Bayer Pharmaceutical) and chlorantraniliprole (DuPont Crop Protection), belonging to the new chemical classes phthalic acid diamides and anthranilic diamides, respectively, act on the ryanodine receptor as a new target of insecticides.[3,4] Because of their excellent performance in the field, new mode of action, and low toxicity to mammals, they have been applied broadly worldwide and have been attracting the attention of researchers focusing on insecticide discovery. These compounds were considered as the most important breakthrough in the field of pesticides after the discovery of dibenzoylhydrazine compounds.[5]

In 1988, Rohm & Hass Co. introduced dibenzoylhydrazine compound RH-5849[6,7] as an insect growth inhibitor (ecdysone antagonist) that has high efficacy and a new action mode. Since then, acylhydrazines that act as insect growth regulators have become one of the main classes of pesticides. Subsequently, many new compounds based on the features of RH-5849 have been discovered and widely used over the past 20 years, such as RH-5992,[8] ANS-118,[9] and JS-118.[10]

Functional fragments or skeletons play a crucial role in biologically active chemicals, and modifications in the functional groups could improve or change the biological activity of parent compounds. To improve the biological activity of anthranilic diamide insecticides against aphids or acarids, we incorporated the functional fragment acylhydrazine into anthranilic diamide and synthesized a series of 2-acylamino-substituted *N*′-benzylbenzohydrazide derivatives (Table 3) according to Scheme 1. The biological activities of these compounds against three representative insect species were assessed preliminarily.

MATERIALS AND METHODS

Instruments and Reagents. Reagents and solvents were purchased from Alfa-Aesar, J&K, Beijing Coupling Technology Co.,

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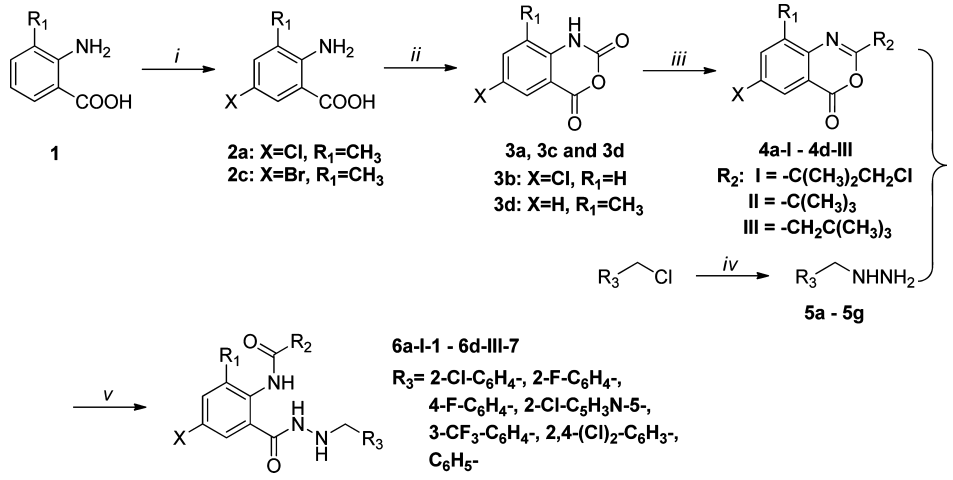
Scheme 1[a]



[a]Reagents and conditions: (*i*) DMF, NCS/NBS, 80 °C, 30 min; (*ii*) 1,4-dioxane, triphosgene, reflux, 6 h; (*iii*) acyl chloride, dry THF, Et_3N, 0 °C, room temperature, 12 h; (*iv*) 85% hydrazine hydrate, ethanol, reflux, 30 min; (*v*) THF, room temperature, 24−48 h.

Ltd., or China Sinopharm Chemical Reagent Co., Ltd., and used directly without further purification. All anhydrous solvents were dried and purified using the standard techniques before use. New compounds were purified using column chromatography filled with silica gel or recrystallized in organic solvents.

^{1}H nuclear magnetic resonance (NMR) spectra were obtained at 300 MHz by using a Bruker spectrometer in $CDCl_3$ or DMSO-d_6 solution with tetramethylsilane as the internal standard. ^{13}C NMR spectra were obtained on a Bruker spectrometer and recorded in $CDCl_3$ solution referenced to its corresponding solvent peaks (δ 77.23). Chemical shift values (δ) were expressed in parts per million (ppm). High-resolution mass spectrometry (HRMS) and MS data were recorded using Agilent G1969A and G1956B spectrometers, respectively, by using the electron impact (EI) ion source. The melting points were determined using a Stuart melting point apparatus and were uncorrected.

Synthesis. *Synthesis of 2-Amino-5-chloro-3-methylbenzoic Acid (**2a**)* .[11] 2-Amino-3-methylbenzoic acid **1** (15.1 g, 0.1 mol) was dissolved in 100 mL of *N*,*N*-dimethylformamide and heated to 70 °C; subsequently, *N*-chlorosuccinimide (14.5 g, 0.11 mol) was gradually added at 70−80 °C. The mixture was stirred at 80 °C for 30 min and poured into a flask containing 500 g of ice. The precipitate was filtered, rinsed with ice water, and dried to afford 18.1 g of **2a** as a pale powder: yield, 97.8%; meting point (mp), 196.2 °C (decomposed); ^{1}H NMR (300 MHz, DMSO) δ 7.61 (d, J = 2.5 Hz, 1H, Ar−H), 7.26 (d, J = 2.5 Hz, 1H, Ar−H), 2.16 (s, 3H, Ar−CH_3); EI-MS (m/z, %) 186 ([M + H]$^+$, 100).

*Synthesis of 2-Amino-5-bromo-3-methylbenzoic Acid (**2c**).* *N*-Bromosuccinimide (19.5 g, 0.11 mol) was reacted with 2-amino-3-methylbenzoic acid **1** (15.1 g, 0.1 mol) according to the procedure described for compound **2a** to afford 21.9 g of **2c** as a brown powder: yield, 95.6%; mp, 218.2−219.7 °C; ^{1}H NMR (300 MHz, DMSO) δ 7.68 (d, J = 2.5 Hz, 1H, Ar−H), 7.32 (d, J = 2.5 Hz, 1H, Ar−H), 2.11 (s, 3H, Ar−CH_3); EI-MS (m/z, %) 230 ([M + H]$^+$, 100).

*Synthesis of 6-Chloro-8-methyl-1H-benzo[d][1,3]oxazine-2,4-dione (**3a**)* .[12] A suspension of **2a** (9.25 g, 50 mmol) in 100 mL of 1,4-dioxane was heated to reflux; subsequently, triphosgene (5.88 g, 20 mmol) in 50 mL of 1,4-dioxane was added dropwise over a period of 30 min. The reaction mixture was kept at reflux for 6 h and then cooled to room temperature in a water bath. The resultant precipitate was filtered, rinsed with petroleum ether, and dried to afford 10.30 g of **3a** as a white powder: yield, 97.6%; mp, 263 °C (decomposed); ^{1}H NMR (300 MHz, DMSO) δ 10.14 (s, 1H, −NH−), 7.85 (d, J = 2.5 Hz, 1H, Ar−H), 7.57 (d, J = 2.5 Hz, 1H, Ar−H), 2.21 (s, 3H, Ar−CH_3); EI-MS (m/z, %) 212 ([M + H]$^+$, 100).

Compound **3b** is commercially available, and intermediates **3c** and **3d** were prepared according to the procedure described for intermediate **3a**. The data for **3c** and **3d** are shown in Table 1.

*Synthesis of 6-Chloro-2-(1-chloro-2-methylpropan-2-yl)-8-methyl-4H-benzo[d][1,3]oxazin-4-one (**4a-I**)* .[13] Intermediate **3a** (1.06 g, 5 mmol) and anhydrous triethylamine (2.02 g, 20 mmol) were added to 20 mL of anhydrous tetrahydrofuran, and the mixture was cooled to 0 °C under stirring. Next, 3-chloro-2,2-dimethylpropanoyl chloride (0.93 g, 6 mmol) was injected slowly into the mixture by using a syringe over a period of 10 min. The mixture was stirred at room temperature for 12 h, and most of the tetrahydrofuran and triethylamine were distilled off using a rotary evaporator. The residue was dissolved in 20 mL of ethyl acetate and then washed once with saturated sodium bicarbonate solution and twice with water. The obtained organic phase was dried and concentrated to afford 1.53 g of **4a-I** as a white solid: yield, 93%; mp, 87.2−87.9 °C; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.95 (d, J = 2.5 Hz, 1H, Ar−H), 7.56 (d, J = 2.5 Hz, 1H, Ar−H), 3.78 (s, 2H, −CH_2−Cl), 2.51 (s, 3H, Ar−CH_3), 1.22 (s, 6H, Me_2−C); EI-MS (m/z, %) 285 (M$^+$, 100).

Intermediates **4a-II**−**4d-III** were prepared according to the procedure used for **4a-I**. The data for **4a-II**−**4d-III** are shown in Table 1.

*Synthesis of 2-Chlorobenzyl Hydrazine (**5a**)* .[14] For this, 50 g (0.85 mol) of hydrazine hydrate (85%) was dissolved in 200 mL of ethanol and heated to reflux. Subsequently, 2-chlorobenzyl chloride (16 g, 0.1 mol) was added dropwise over a period of 1 h, and the reaction mixture was kept at reflux for another 30 min. Most of the ethanol and hydrazine hydrate were distilled off using a rotary evaporator, and the residue was dissolved in ethyl acetate and washed with brine and water. The obtained organic layer was dried and distilled to afford 10.2 g of **5a** as a light yellow liquid (yield, 65%). The other benzyl hydrazines **5b**−**5g** (**5b**, 2-fluorobenzyl hydrazine; **5c**, 4-fluorobenzyl hydrazine; **5d**, 2-chloro-5-(hydrazinylmethyl)pyridine; **5e**, 3-(trifluoromethyl)benzyl hydrazine; **5f**, 2,4-dichlorobenzyl hydrazine; and **5g**, benzyl hydrazine) were prepared by following the same procedure as that used for **5a**, and all of these intermediates were directly reacted with **4a-I**−**4d-III** without purification.

General procedures for the synthesis of target compounds have been described below (**6a-I-1** as an example), and the data for **6a-I-2**−**6d-III-5** are shown in Table 2.

*Synthesis of 3-Chloro-N-(4-chloro-2-(2-(2-chlorobenzyl)-hydrazinecarbonyl)-6-methylphenyl)-2,2-dimethylpropanamide (**6a-I-1**)* .[15] 2-Chlorobenzyl hydrazine **5a** (0.19 g, 1.2 mmol) was added to a solution of compound **4a-I** (0.33 g, 1 mmol) in 5 mL of tetrahydrofuran, and the mixture was stirred for about 24 h at room temperature. When the reaction was completed by using a thin layer chromatography (TLC) monitor, 10 mL of petroleum ether was

Table 1. Structural and Analytical Data for Intermediates 3c, 3d, and 4a-II−4d-III

intermediate	X	R_1	R_2	yield (%)	appearance	mp (°C)	^{1}H NMR and.or ^{13}C NMR	EI-MS (*m*/*z*, %)
3c	Br	CH_3		97	pink powder	284 dec	^{1}H NMR (300 MHz, DMSO) δ 11.18 (s, 1H, −NH−), 7.80 (d, J = 2.3 Hz, 1H, Ar−H), 7.75 (d, J = 2.3 Hz, 1H, Ar−H), 2.31 (s, 3H, Ar−CH_3) ^{13}C NMR (75 MHz, DMSO) δ 159.02, 147.02, 139.89, 139.29, 128.29, 127.63, 114.50, 112.39, 16.83	256 ([M + H]$^+$, 100)
3d	H	CH_3		95	white powder	275.0 dec	^{1}H NMR (300 MHz, DMSO) δ 11.02 (s, 1H, −NH−), 7.76 (dd, J = 7.7, 0.6 Hz, 1H, Ar−H), 7.64 − 7.51 (dd, J = 7.7, 0.6 Hz, 1H, Ar−H), 7.15 (t, J = 7.7 Hz, 1H, Ar−H), 2.33 (s, 3H, Ar−CH_3) ^{13}C NMR (75 MHz, DMSO) δ 160.09, 147.40, 139.81, 138.07, 126.81, 124.58, 123.31, 110.47, 17.10	178 ([M + H]$^+$, 100)
4a-II	Cl	CH_3	$-C(CH_3)_3$	90	white powder	100.2−101.4	^{1}H NMR (300 MHz, $CDCl_3$) δ 7.85 (dd, J = 2.5, 0.6 Hz, 1H, Ar−H), 7.50 (dd, J = 2.5, 0.8 Hz, 1H, Ar−H), 2.51 (s, 3H, Ar−CH_3), 1.42 (s, 9H, Me_3-C) ^{13}C NMR (75 MHz, $CDCl_3$) δ 167.05, 158.92, 143.03, 137.86, 136.44, 132.43, 124.50, 117.35, 37.85, 27.37	251 (M$^+$, 100)
4a-III	Cl	CH_3	$-CH_2-C(CH_3)_3$	88	white crystal	104.3−105.5	^{1}H NMR (300 MHz, $CDCl_3$) δ 7.95 (dd, J = 2.5, 0.6 Hz, 1H, Ar−H), 7.58−7.54 (m, 1H, Ar−H), 2.58 (s, 2H, $-CH_2-$C), 2.52 (s, 3H, Ar−CH_3), 1.11 (s, 9H, Me_3−C) ^{13}C NMR (75 MHz, $CDCl_3$) δ 160.67, 159.19, 143.34, 137.86, 136.89, 132.82, 124.91, 117.71, 48.01, 31.86, 29.56, 16.86	265 (M$^+$, 100)
4b-I	Cl	H	$-C(CH_3)_2CH_2Cl$	79	pale powder	105.5−107.3	^{1}H NMR (300 MHz, $CDCl_3$) δ 8.10 − 8.05 (m, 1H, Ar−H), 7.72 (dd, J = 8.6, 2.4 Hz, 1H, Ar−H), 7.5 −7.53 (m, 1H, Ar−H), 3.82 (s, 2H, $-CH_2-$Cl), 1.50 (s, 6H, Me_2−C) ^{13}C NMR (75 MHz, $CDCl_3$) δ 164.50, 157.97, 144.27, 136.36, 133.70, 128.42, 127.30, 117.66, 51.73, 43.30, 23.50	251 (M$^+$, 100)
4b-II	Cl	H	$-C(CH_3)_3$	81	white powder	106.8−108.0	^{1}H NMR (300 MHz, $CDCl_3$) δ 8.14 (d, J = 2.5 Hz, 1H, Ar−H), 7.76 (dd, J = 8.6, 2.5 Hz, 1H, Ar−H), 7.54 (d, J = 8.6 Hz, 1H, Ar−H), 1.20 (s, 9H, Me_3−C)	237 (M$^+$, 100)
4b-III	Cl	H	$-CH_2-C(CH_3)_3$	74	white crystal	60.0−61.1	^{1}H NMR (300 MHz, $CDCl_3$) δ 8.06 (d, J = 2.5 Hz, 1H, Ar−H), 7.71 (dd, J = 8.6, 2.5 Hz, 1H, Ar−H), 7.53 (d, J = 8.6 Hz, 1H, Ar−H), 2.58 (s, 2H, $-CH_2-$C), 1.12 (s, 9H, Me_3−C) ^{13}C NMR (75 MHz, $CDCl_3$) δ 162.02, 158.16, 144.58, 136.27, 133.33, 128.05, 127.26, 117.65, 47.94, 31.94, 29.47	251 (M$^+$, 100)
4c-I	Br	CH_3	$-C(CH_3)_2CH_2Cl$	92	pale powder	76.3−78.0	^{1}H NMR (300 MHz, $CDCl_3$) δ 8.12 (d, J = 2.3 Hz, 1H, Ar−H), 7.74 (d, J = 2.3 Hz, 1H, Ar−H), 3.82 (s, 2H, $-CH_2-$Cl), 2.53 (s, 3H, Ar−CH_3), 1.49 (s, 6H, Me_2−C) ^{13}C NMR (75 MHz, $CDCl_3$) δ 163.36, 158.20, 142.90, 139.45, 138.13, 127.69, 120.68, 117.61, 51.66, 43.28, 23.40, 16.39	329 (M$^+$, 100)
4c-II	Br	CH_3	$-C(CH_3)_3$	94	needle crystal	99.4−100.5	^{1}H NMR (300 MHz, $CDCl_3$) δ 8.01 (d, J = 2.3 Hz, 1H, Ar−H), 7.65 (d, J = 2.3 Hz, 1H, Ar−H), 2.51 (s, 3H, Ar−CH_3), 1.42 (s, 9H, Me_3−C) ^{13}C NMR (75 MHz, $CDCl_3$) δ 167.22, 158.84, 143.44, 139.34, 138.05, 127.74, 120.29, 117.67, 37.94, 27.43, 16.46	295 (M$^+$, 100)
4c-III	Br	CH_3	$-CH_2-C(CH_3)_3$	89	yellow powde	95.9−97.2	^{1}H NMR (300 MHz, $CDCl_3$) δ 8.09 (d, J = 2.5 Hz, 1H, Ar−H), 7.69 (d, J = 2.5 Hz, 1H, Ar−H), 2.56 (s, 3H, Ar−CH_3), 2.12 (s, 2H, $-CH_2-$C), 1.18 (s, 9H, Me_3−C)	309 (M$^+$, 100)
4d-I	H	CH_3	$-C(CH_3)_2CH_2Cl$	90	pale powder	53.1−53.4	^{1}H NMR (300 MHz, $CDCl_3$) δ 8.04 (dd, J = 7.7, 2.5 Hz, 1H, Ar−H), 7.63 (dd, J = 7.7, 2.5 Hz, 1H, Ar−H), 7.37 (t, J = 7.7 Hz, 1H, Ar−H), 2.51 (s, 3H, Ar−CH_3), 3.80 (s, 2H, $-CH_2-$Cl), 1.20 (s, 6H, Me_2−C)	251 (M$^+$, 100)
4d-II	H	CH_3	$-C(CH_3)_3$	85	white powder	62.2−63.0	^{1}H NMR (300 MHz, $CDCl_3$) δ 7.99 (ddd, J = 7.9, 1.5, 0.6 Hz, 1H, Ar−H), 7.59 (ddd, J = 7.5, 1.5, 0.8 Hz, 1H, Ar−H), 7.33 (t, J = 7.7 Hz, 1H, Ar−H), 2.54 (s, 3H, Ar−CH_3), 1.41 (s, 9H, Me_3−C) ^{13}C NMR (75 MHz, $CDCl_3$) δ 166.74, 160.47, 144.62, 136.87, 135.76, 127.21, 125.61, 116.50, 37.91, 27.56, 16.66	217 (M$^+$, 100)
4d-III	H	CH_3	$-CH_2-C(CH_3)_3$	84	white crystal	77.7−79.0	^{1}H NMR (300 MHz, $CDCl_3$) δ 7.97 (ddd, J = 7.9, 1.5, 0.6 Hz, 1H, Ar−H), 7.58 (ddd, J = 7.5, 1.5, 0.8 Hz, 1H, Ar−H), 7.32 (t, J = 7.7 Hz, 1H, Ar−H), 2.56 (s, 2H, $-CH_2-$C), 2.52 (s, 3H, Ar−CH_3), 1.11 (s, 9H, Me_3−C) ^{13}C NMR (75 MHz, $CDCl_3$) δ 160.03, 159.95, 144.45, 136.75, 135.28, 127.10, 125.44, 116.37, 47.78, 31.54, 29.36, 16.73	231 (M$^+$, 100)

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Table 2. Analytical Data for Target Compounds 6a-I-2−6d-III-5

compd	yield (%)	appearance	mp (°C)	^{1}H NMR	EI-MS or HRMS
6a-I-2	82	yellow powder	200.3−201.4	^{1}H NMR (300 MHz, DMSO) δ 9.87 (d, J = 6.0 Hz, 1H, −NH−), 9.28 (s, 1H, −NH−), 7.55−7.48 (m, 1H, Ar−H), 7.46 (d, J = 2.9 Hz, 1H, Ar−H), 7.36−7.28 (m, 1H, Ar−H), 7.24 (d, J = 2.4 Hz, 1H, Ar−H), 7.21−7.12 (m, 2H), 5.44 (dd, J = 6.0, 4.3 Hz, 1H, −NH−), 4.00 (d, J = 4.3 Hz, 2H, Ar−CH_2−), 3.77 (s, 2H, −CH_2−Cl), 2.17 (s, 3H, Ar−CH_3), 1.28 (s, 6H, Me_2−C)	EI-MS (m/z, %) 426 ([M + H]$^+$, 100)
6a-I-3	87	white powder	186.5−186.7	^{1}H NMR (300 MHz, DMSO) δ 9.81 (d, J = 5.1 Hz, 1H, −NH−), 9.28 (s, 1H, −NH−), 7.46 (d, J = 2.5 Hz, 1H, Ar−H), 7.43−7.37 (m, 2H, Ar−H), 7.22 (d, J = 2.5 Hz, 1H, Ar−H), 7.18−7.10 (m, 2H, Ar−H), 5.40 (dd, J = 5.1, 3.8 Hz, 1H, −NH−), 3.92 (d, J = 3.8 Hz, 2H, Ar−CH_2−), 3.78 (s, 2H, −CH_2−Cl), 2.16 (s, 3H, Ar−CH_3), 1.28 (s, 6H, Me_2−C)	HRMS (EI) calcd for $C_{20}H_{23}Cl_2FN_3O_2$ [M + H]$^+$ 426.1151, found 426.1148
6a-I-4	79	white powder	183.7−185.3	^{1}H NMR (300 MHz, DMSO) δ 9.77 (d, J = 5.1 Hz, 1H, −NH−), 9.25 (s, 1H, −NH−), 8.39 (d, J = 2.0 Hz, 1H, Ar−H), 7.86 (dd, J = 5.9, 2.5 Hz, 1H, Ar−H), 7.47 (dd, J = 5.9, 2.0 Hz, 1H, Ar−H), 7.46 (s, 1H, Ar−H), 7.20 (d, J = 2.5 Hz, 1H, Ar−H), 5.63 (dd, J = 5.1, 2.8 Hz, 1H, −NH−), 3.95 (d, J = 2.8 Hz, 2H, Ar−CH_2−), 3.78 (s, 2H, −CH_2−Cl), 2.17 (s, 3H, Ar−CH_3), 1.28 (s, 6H, Me_2−C)	HRMS (EI) calcd for $C_{19}H_{22}Cl_3N_4O_2$ [M + H]$^+$ 443.0808, found 443.0815
6a-I-5	84	white powder	192.4−192.9	^{1}H NMR (300 MHz, DMSO) δ 9.82 (d, J = 6.3 Hz, 1H, −NH−), 9.27 (s, 1H, −NH−), 7.74 (s, 1H, Ar−H), 7.70−7.66 (m, 1H, Ar−H), 7.64−7.52 (m, 2H, Ar−H), 7.46 (dd, J = 2.5, 0.6 Hz, 1H, Ar−H), 7.18 (dd, J = 2.5, 0.6 Hz, 1H, Ar−H), 5.61 (dd, J = 6.3, 4.7 Hz, 1H, −NH−), 4.03 (d, J = 4.7 Hz, 2H, Ar−CH_2−), 3.78 (s, 2H, −CH_2−Cl), 2.16 (s, 3H, Ar−CH_3), 1.28 (s, 6H, Me_2−C)	EI-MS (m/z, %) 476 ([M + H]$^+$, 100)
6a-I-6	89	white powder	187.7−190.8	^{1}H NMR (300 MHz, DMSO) δ 9.86 (d, J = 6.0 Hz, 1H, −NH−), 9.26 (s, 1H, −NH−), 7.65 (d, J = 8.3 Hz, 1H, Ar−H), 7.58 (d, J = 2.5 Hz, 1H, Ar−H), 7.46 (d, J = 2.2 Hz, 1H, Ar−H), 7.41 (dd, J = 8.3, 2.2 Hz, 1H, Ar−H), 7.25 (d, J = 2.5 Hz, 1H, Ar−H), 5.59 (dd, J = 6.0, 3.9 Hz, 1H, −NH−), 4.03 (d, J = 3.9 Hz, 2H, Ar−CH_2−), 3.77 (s, 2H, −CH_2−Cl), 2.16 (s, 3H, Ar−CH_3), 1.27 (s, 6H, Me_2−C)	EI-MS (m/z, %) 476 ([M + H]$^+$, 100)
6a-I-7	85	white powder	170.1−171.3	^{1}H NMR (300 MHz, DMSO) δ 9.86 (d, J = 6.1 Hz, 1H, −NH−), 9.30 (s, 1H, −NH−), 7.46 (dd, J = 2.5, 0.6 Hz, 1H, Ar−H), 7.39−7.31 (m, 4H, Ar−H), 7.29−7.24 (m, 1H, Ar−H), 7.23 (dd, J = 2.5, 0.6 Hz, 1H, Ar−H), 5.37 (dd, J = 6.1, 4.5 Hz, 1H, −NH−), 3.94 (d, J = 4.5 Hz, 2H, Ar−CH_2−), 3.78 (s, 2H, −CH_2−Cl), 2.17 (s, 3H, Ar−CH_3), 1.29 (s, 6H, Me_2−C)	EI-MS (m/z, %) 408 ([M + H]$^+$, 100)
6a-II-1	83	white powder	198.3−199.3	^{1}H NMR (300 MHz, DMSO) δ 9.87 (d, J = 5.5 Hz, 1H, −NH−), 9.14 (s, 1H, −NH−), 7.61 (m, 1H, Ar−H), 7.46−7.39 (m, 2H, Ar−H), 7.36−7.27 (m, 2H, Ar−H), 7.26 (d, J = 2.4 Hz, 1H, Ar−H), 5.58 (dd, J = 5.5, 4.1 Hz, 1H, −NH−), 4.07 (d, J = 4.1 Hz, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.20 (s, 9H, Me_3−C)	HRMS (EI) calcd for $C_{20}H_{24}Cl_2N_3O_2$ [M + H]$^+$ 408.1246, found 408.1237
6a-II-2	88	faint yellow powder	150.2−151.2	^{1}H NMR (300 MHz, DMSO) δ 9.86 (d, J = 5.3 Hz, 1H, −NH−), 9.14 (s, 1H, −NH−), 7.56−7.49 (m, 1H, Ar−H), 7.44 (d, J = 2.4 Hz, 1H, Ar−H), 7.34−7.24 (m, 2H, Ar−H), 7.24−7.09 (m, 2H, Ar−H), 5.49 (dd, J = 5.3, 2.8 Hz, 1H, −NH−), 4.02 (d, J = 2.8 Hz, 2H, Ar−CH_2−), 2.14 (s, 3H, Ar−CH_3), 1.20 (s, 9H, Me_3−C)	EI-MS (m/z, %) 392 ([M + H]$^+$, 100)
6a-II-3	84	white powder	196.0−196.2	^{1}H NMR (300 MHz, DMSO) δ 9.78 (d, J = 6.5 Hz, 1H, −NH−), 9.12 (s, 1H, −NH−), 7.44 (d, J = 2.4 Hz, 1H, Ar−H), 7.43−7.36 (m, 2H, Ar−H), 7.22 (d, J = 2.4 Hz, 1H, Ar−H), 7.18−7.09 (m, 2H, Ar−H), 5.45 (dd, J = 6.5, 5.0 Hz, 1H, −NH−), 3.92 (d, J = 5.0 Hz, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.20 (s, 9H, Me_3−C)	EI-MS (m/z, %) 392 ([M + H]$^+$, 100)
6a-II-4	71	white powder	191.4−192.8	^{1}H NMR (300 MHz, DMSO) δ 9.73 (d, J = 6.3 Hz, 1H, −NH−), 9.08 (s, 1H, −NH−), 8.39 (d, J = 2.3 Hz, 1H, Ar−H), 7.86 (dd, J = 8.2, 2.5 Hz, 1H, Ar−H), 7.49−7.42 (m, 2H, Ar−H), 7.20 (d, J = 2.5 Hz, 1H, Ar−H), 5.66 (dd, J = 6.3, 4.5 Hz, 1H, −NH−), 3.95 (d, J = 4.5 Hz, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.19 (s, 9H, Me_3−C)	EI-MS (m/z, %) 409 ([M + H]$^+$, 100)
6a-II-5	75	white powder	157.5−158.1	^{1}H NMR (300 MHz, DMSO) δ9.72 (d, J = 6.3 Hz, 1H, −NH−), 9.17 (s, 1H, −NH−), 7.64 (s, 1H, Ar−H), 7.43−7.57 (m, 3H, Ar−H), 7.35 (m, 1H, Ar−H), 7.07−7.08 (m, 1H, Ar−H), 5.48−5.53 (dd, J = 6.3, 4.6 Hz, 1H, −NH−), 3.93 (d, J = 4.6 Hz, 2H, Ar−CH_2−), 2.06 (s, 3H, Ar−CH_3), 1.18 (s, 9H, Me_3−C)	EI-MS (m/z, %) 442 ([M + H]$^+$, 100)
6a-II-6	87	white powder	206.2−207.5	^{1}H NMR (300 MHz, DMSO) δ 9.83 (s, 1H, −NH−), 9.10 (s, 1H, −NH−), 7.65 (d, J = 8.3 Hz, 1H, Ar−H), 7.57 (d, J = 2.5 Hz, 1H, Ar−H), 7.44 (d, J = 2.2 Hz, 1H, Ar−H), 7.40 (dd, J = 8.3, 2.2 Hz, 1H, Ar−H), 7.25 (d, J = 2.5 Hz, 1H, Ar−H), 5.63 (s, 1H, −NH−), 4.04 (s, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.18 (s, 9H, Me_3−C)	EI-MS (m/z, %): 442 ([M + H]$^+$, 100)
6a-II-7	91	white powder	172.5−173.3	^{1}H NMR (300 MHz, DMSO) δ 9.80 (d, J = 4.9 Hz, 1H, −NH−), 9.13 (s, 1H, −NH−), 7.44 (d, J = 2.1 Hz, 1H, Ar−H), 7.39−7.29 (m, 4H, Ar−H), 7.29−7.18 (m, 2H, Ar−H), 5.40 (d, J = 4.9 Hz, 1H, −NH−), 3.94 (s, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.23 (s, 9H, Me_3-C)	EI-MS (m/z, %) 374 ([M + H]$^+$, 100)
6a-III-1	75	white powder	152.9−153.7	^{1}H NMR (300 MHz, DMSO) δ 9.84 (s, 1H, −NH−), 9.28 (s, 1H, −NH−), 7.62 (m, 1H, Ar−H), 7.45−7.40 (m, 2H, Ar−H), 7.34−7.27 (m, 2H, Ar−H), 7.20 (d, J = 2.5 Hz, 1H, Ar−H), 5.47 (s, 1H, −NH−), 4.06 (s, 2H, Ar−CH_2−), 2.17 (s, 3H, Ar−CH_3), 2.08 (s, 2H, −CH_2−C), 1.00 (s, 9H, Me_3−C)	HRMS (EI) calcd for $C_{21}H_{26}Cl_2N_3O_2$ [M + H]$^+$ 422.1402, found 422.1387
6a-III-5	86	white powder	165.6−165.7	^{1}H NMR (300 MHz, DMSO) δ 9.30 (s, 1H, −NH−), 7.73−7.59 (m, 4H, Ar−H), 7.36 (d, J = 2.4 Hz, 1H, Ar−H), 7.26 (d, J = 2.4 Hz, 1H, Ar−H), 4.73 (s, 2H, Ar−CH_2−), 2.17 (s, 3H, Ar−CH_3), 1.99 (s, 2H, −CH_2−C), 0.97 (s, 9H, Me_3−C)	EI-MS (m/z, %) 456 ([M + H]$^+$, 100)
6a-III-6	75	white powder	177.1−177.5	^{1}H NMR (300 MHz, DMSO) δ 9.78 (s, 1H, −NH−), 9.25 (s, 1H, −NH−), 7.67 (d, J = 8.3 Hz, 1H, Ar−H), 7.57 (d, J = 2.2 Hz, 1H, Ar−H), 7.43 (d, J = 2.4 Hz, 1H, Ar−H), 7.40 (dd, J = 8.3, 2.2 Hz, 1H, Ar−H), 7.19 (d, J = 2.4 Hz, 1H, Ar−H), 5.53 (s, 1H, −NH−), 4.03 (s, 2H, Ar−CH_2−), 2.17 (s, 3H, Ar−CH_3), 2.04 (s, 2H, −CH_2−C), 1.00 (s, 9H, Me_3−C)	EI-MS (m/z, %) 456 ([M + H]$^+$, 100)
6a-III-7	75	white powder	179.1−183.0	^{1}H NMR (300 MHz, DMSO) δ 9.77 (s, 1H, −NH−), 9.26 (s, 1H, −NH−), 7.47−7.41 (m, 1H, Ar−H), 7.41−7.29 (m, 4H, Ar−H), 7.29−7.22 (m, 1H, Ar−H), 7.21−7.12 (m, 1H, Ar−H), 5.27 (s, 1H, −NH−), 3.94 (s, 2H, Ar−CH_2−), 2.17 (s, 3H, Ar−CH_3), 2.12 (s, 2H, −CH_2−C), 1.02 (s, 9H, Me_3−C)	HRMS (EI) calcd for $C_{21}H_{27}ClN_3O_2$ [M + H]$^+$ 388.1792, found 388.1784

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Table 2. continued

compd	yield (%)	appearance	mp (°C)	^{1}H NMR	EI-MS or HRMS
6b-II-3	87	white powder	143.7−144.9	^{1}H NMR (300 MHz, DMSO) δ 11.25 (s, 1H, −NH−), 10.33 (d, J = 6.3 Hz, 1H, −NH−), 8.42 (d, J = 9.0 Hz, 1H, Ar−H), 7.58 (d, J = 2.4 Hz, 1H, Ar−H), 7.47 (dd, J = 9.0, 2.4 Hz, 1H, Ar−H), 7.35 (dd, J = 8.4, 5.8 Hz, 2H, Ar−H), 7.07 (t, J = 8.4 Hz, 2H, Ar−H), 5.71 (dd, J = 6.3, 5.1 Hz, 1H, −NH−), 3.90 (d, J = 5.1 Hz, 2H, Ar−CH_2−), 1.13 (s, 9H, Me_3−C)	EI-MS (m/z, %) 378 ([M + H]$^+$, 100)
6b-II-5	83	white crystal	152.4−154.8	^{1}H NMR (300 MHz, DMSO) δ 11.29 (s, 1H, −NH−), 10.38 (s, 1H, −NH−), 8.49 (d, J = 9.0 Hz, 1H, Ar−H), 7.76 (s, 1H, Ar−H), 7.71−7.51 (m, 5H, Ar−H), 5.94 (s, 1H, −NH−), 4.09 (s, 2H, Ar−CH_2−), 1.20 (s, 9H, Me_3−C)	EI-MS (m/z, %) 428 ([M + H]$^+$, 100)
6b-III-2	71	white powder	147.9−149.1	^{1}H NMR (300 MHz, DMSO) δ 10.74 (s, 1H, −NH−), 10.35 (s, 1H, −NH−), 8.35 (d, J = 8.9 Hz, 1H, Ar−H), 7.61 (d, J = 2.5 Hz, 1H, Ar−H), 7.57−7.46 (m, 2H, Ar−H), 7.40−7.10 (m, 4H, Ar−H), 5.67 (s, 1H, −NH−), 4.04 (s, 2H, Ar−CH_2−), 2.17 (s, 2H, −CH_2−C), 1.00 (s, 9H, Me_3−C)	EI-MS (m/z, %) 392 ([M + H]$^+$, 100)
6c-I-1	77	faint yellow powder	178.8−179.5	^{1}H NMR (300 MHz, DMSO) δ 9.91 (s, 1H, −NH−), 9.29 (s, 1H, −NH−), 7.64−7.57 (m, 2H, Ar−H), 7.45−7.37 (m, 2H, Ar−H), 7.36−7.26 (m, 2H, Ar−H), 5.53 (s, 1H, −NH−), 4.07 (s, 2H, Ar−CH_2−), 3.77 (s, 2H, −CH_2−Cl), 2.15 (s, 3H, Ar−CH_3), 1.27 (s, 6H, Me_2−C)	EI-MS (m/z, %) 486 ([M + H]$^+$, 100)
6c-I-4	72	white powder	190.4−190.5	^{1}H NMR (300 MHz, DMSO) δ 9.77 (d, J = 6.3 Hz, 1H, −NH−), 9.25 (s, 1H, −NH−), 8.39 (d, J = 2.3 Hz, 1H, Ar−H), 7.86 (dd, J = 8.2, 2.5 Hz, 1H, Ar−H), 7.59 (dd, J = 2.3, 0.6 Hz, 1H, Ar−H), 7.47 (dd, J = 8.2, 0.6 Hz, 1H, Ar−H), 7.36−7.30 (m, 1H, Ar−H), 5.62 (dd, J = 6.3, 4.5 Hz, 1H, −NH−), 3.95 (d, J = 4.5 Hz, 2H, Ar−CH_2−), 3.78 (s, 2H, −CH_2−Cl), 2.16 (s, 3H, Ar−CH_3), 1.27 (s, 6H, Me_2−C)	HRMS (EI) calcd for $C_{19}H_{22}BrCl_2N_4O_2$ [M + H]$^+$ 487.0303, found 487.0299
6c-I-5	73	white powder	181.8−183.3	^{1}H NMR (300 MHz, DMSO) δ 9.81 (d, J = 5.7 Hz, 1H, −NH−), 9.25 (s, 1H, −NH−), 7.73 (s, 1H, Ar−H), 7.70−7.65 (m, 1H, Ar−H), 7.63−7.53 (m, 3H, Ar−H), 7.30 (d, J = 2.1 Hz, 1H, Ar−H), 5.59 (dd, J = 5.7, 3.8 Hz, 1H, −NH−), 4.03 (d, J = 3.8 Hz, 2H, Ar−CH_2−), 3.77 (s, 2H, −CH_2−Cl), 2.16 (s, 3H, Ar−CH_3), 1.27 (s, 6H, Me_2−C)	EI-MS (m/z, %) 520 ([M + H]$^+$, 100)
6c-I-6	79	white powder	186.1−186.4	^{1}H NMR (300 MHz, DMSO) δ 9.88 (s, 1H, −NH−), 9.25 (s, 1H, −NH−), 7.62 (d, J = 8.3 Hz, 1H, Ar−H), 7.57 (dd, J = 2.4, 0.6 Hz, 1H, Ar−H), 7.55 (d, J = 2.2 Hz, 1H, Ar−H), 7.39 (dd, J = 8.3, 2.2 Hz, 1H, Ar−H), 7.35 (dd, J = 2.4, 0.6 Hz, 1H, Ar−H), 5.55 (s, 1H, −NH−), 4.01 (s, 2H, Ar−CH_2−), 3.74 (s, 2H, −CH_2−Cl), 2.14 (s, 3H, Ar−CH_3), 1.24 (s, 6H, Me_2−C)	EI-MS (m/z, %) 520 ([M + H]$^+$, 100)
6c-I-7	82	faint yellow powder	207.1−207.9	^{1}H NMR (300 MHz, DMSO) δ 9.85 (s, 1H, −NH−), 9.29 (s, 1H, −NH−), 7.58 (d, J = 2.2 Hz, 1H, Ar−H), 7.39−7.29 (m, 5H, Ar−H), 7.29−7.23 (m, 1H, Ar−H), 5.35 (s, 1H, −NH−), 3.94 (s, 2H, Ar−CH_2−), 3.78 (s, 2H, −CH_2−Cl), 2.16 (s, 3H, Ar−CH_3), 1.27 (s, 6H, Me_2−C)	EI-MS (m/z, %) 452 ([M + H]$^+$, 100)
6c-II-1	86	white powder	177.5−178.7	^{1}H NMR (300 MHz, DMSO) δ 9.85 (s, 1H, −NH−), 9.11 (s, 1H, −NH−), 7.64−7.55 (m, 2H, Ar−H), 7.44−7.36 (m, 2H, Ar−H), 7.35−7.25 (m, 2H, Ar−H), 5.56 (s, 1H, −NH−), 4.07 (s, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.19 (s, 9H, Me_3−C)	HRMS (EI) calcd for $C_{20}H_{24}BrClN_3O_2$ [M + H]$^+$ 452.0740, found 452.0731
6c-II-4	81	white powder	187.7−187.9	^{1}H NMR (300 MHz, DMSO) δ 9.74 (d, J = 6.3 Hz, 1H, −NH−), 9.07 (s, 1H, −NH−), 8.39 (d, J = 2.5 Hz, 1H, Ar−H), 7.86 (dd, J = 8.2, 2.5 Hz, 1H, Ar−H), 7.58 (d, J = 2.2 Hz, 1H, Ar−H), 7.46 (d, J = 8.2 Hz, 1H, Ar−H), 7.32 (d, J = 2.2 Hz, 1H, Ar−H), 5.66 (dd, J = 6.3, 4.5 Hz, 1H, −NH−), 3.95 (d, J = 4.5 Hz, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.18 (s, 9H, Me_3−C)	HRMS (EI) calcd for $C_{19}H_{23}BrClN_4O_2$ [M + H]$^+$ 453.0693, found 453.0702
6c-II-5	79	white powder	204.0−205.2	^{1}H NMR (300 MHz, DMSO) δ 9.77 (d, J = 5.5 Hz, 1H, −NH−), 9.08 (s, 1H, −NH−), 7.74 (s, 1H, Ar−H), 7.70−7.65 (m, 1H, Ar−H), 7.63−7.53 (m, 3H, Ar−H), 7.30 (d, J = 2.2 Hz, 1H, Ar−H), 5.63 (dd, J = 5.5, 3.5 Hz, 1H, −NH−), 4.04 (d, J = 3.5 Hz, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.19 (s, 9H, Me_3−C)	EI-MS (m/z, %) 486 ([M + H]$^+$, 100)
6c-II-6	76	white powder	210.7−212.6	^{1}H NMR (300 MHz, DMSO) δ 9.82 (d, J = 6.1 Hz, 1H, −NH−), 9.09 (s, 1H, −NH−), 7.64 (d, J = 8.3 Hz, 1H, Ar−H), 7.60−7.56 (m, 2H, Ar−H), 7.40 (dd, J = 8.3, 2.2 Hz, 1H, Ar−H), 7.37 (dd, J = 2.3, 0.4 Hz, 1H, Ar−H), 5.62 (dd, J = 6.1, 4.7 Hz, 1H, −NH−), 4.03 (d, J = 4.7 Hz, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.18 (s, 9H, Me_3−C)	HRMS (EI) calcd for $C_{20}H_{23}BrCl_2N_3O_2$ [M + H]$^+$486.0351, found 486.0334
6c-II-7	88	white powder	168.3−169.6	^{1}H NMR (300 MHz, DMSO) δ 9.80 (d, J = 4.6 Hz, 1H, −NH−), 9.13 (s, 1H, −NH−), 7.57 (d, J = 2.3 Hz, 1H, Ar−H), 7.40−7.33 (m, 4H, Ar−H), 7.33−7.31 (m, 1H, Ar−H), 7.30−7.25 (m, 1H, Ar−H), 5.40 (d, J = 4.6 Hz, 1H, −NH−), 3.94 (s, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.20 (s, 9H, Me_3−C)	HRMS (EI) calcd for $C_{20}H_{25}BrN_3O_2$ [M + H]$^+$ 418.1130, found 418.1129
6c-III-1	76	yellow powder	181.4−181.7	^{1}H NMR (300 MHz, DMSO) δ 9.85 (s, 1H, −NH−), 9.28 (s, 1H, −NH−), 7.62 (dd, J = 7.1, 2.2 Hz, 1H, Ar−H), 7.56 (d, J = 1.8 Hz, 1H, Ar−H), 7.44−7.40 (m, 1H, Ar−H), 7.34−7.27 (m, 3H, Ar−H), 5.43 (w, 1H, −NH−), 4.07 (s, 2H, Ar−CH_2−), 2.17 (s, 3H, Ar−CH_3), 2.09 (s, 2H, −CH_2−C), 1.00 (s, 9H, Me_3−C)	EI-MS (m/z, %) 466 ([M + H]$^+$, 100)
6c-III-4	75	yellow powder	178.9−180.1	^{1}H NMR (300 MHz, DMSO) δ 9.24 (s, 1H, −NH−), 8.41 (d, J = 2.2 Hz, 1H, Ar−H), 7.85 (dd, J = 8.2, 2.5 Hz, 1H, Ar−H), 7.49 (m, 2H, Ar−H), 7.38 (d, J = 2.2 Hz, 1H, Ar−H), 4.65 (s, 2H, Ar−CH_2−), 2.16 (s, 3H, Ar−CH_3), 1.98 (s, 2H, −CH_2−C), 0.97 (s, 9H, Me_3−C)	EI-MS (m/z, %) 467 ([M + H]$^+$, 100)
6c-III-5	76	white powder	200.9−202.1	^{1}H NMR (300 MHz, DMSO) δ 9.74 (d, J = 4.2 Hz, 1H, −NH−), 9.24 (s, 1H, −NH−), 7.74 (s, 1H, Ar−H), 7.71−7.66 (m, 1H, Ar−H), 7.64−7.54 (m, 3H, Ar−H), 7.24 (d, J = 2.0 Hz, 1H, Ar−H), 5.51 (d, J = 4.2 Hz, 1H, −NH−), 4.03 (s, 2H, Ar−CH_2−), 2.16 (s, 3H, Ar−CH_3), 2.09 (s, 2H, −CH_2−C), 1.01 (s, 9H, Me_3−C)	EI-MS (m/z, %) 500 ([M + H]$^+$, 100)
6c-III-7	79	white powder	211.9−212.9	^{1}H NMR (300 MHz, DMSO) δ 9.77 (d, J = 4.7 Hz, 1H, −NH−), 9.26 (s, 1H, −NH−), 7.56 (dd, J = 2.3, 0.6 Hz, 1H, Ar−H), 7.40−7.30 (m, 4H, Ar−H), 7.30−7.22 (m, 2H, Ar−H), 5.26 (d, J = 4.7 Hz, 1H, −NH−), 3.94 (s, 2H, Ar−CH_2−), 2.18 (s, 3H, Ar−CH_3), 2.13 (s, 2H, −CH_2−C), 1.01 (s, 9H, Me_3-C)	HRMS (EI) calcd for $C_{21}H_{27}BrN_3O_2$ [M + H]$^+$ 432.1287, found 432.1283

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Table 2. continued

compd	yield (%)	appearance	mp (°C)	^{1}H NMR	EI-MS or HRMS
6d-I-1	83	white powder	189.7−192.8	^{1}H NMR (300 MHz, DMSO) δ 9.79 (s, 1H, −NH−), 9.32 (s, 1H, −NH−), 7.61 (dd, J = 7.3, 2.0 Hz, 1H, Ar−H), 7.44−7.40 (m, 1H, Ar−H), 7.40−7.29 (m, 3H, Ar−H), 7.29−7.14 (m, 3H, Ar−H), 5.53 (s, 1H, −NH−), 4.08 (s, 2H, Ar−CH_2−), 3.77 (s, 2H, −CH_2−Cl), 2.16 (s, 3H, Ar−CH_3), 1.29 (s, 6H, Me_2−C)	EI-MS (m/z, %) 408 ([M + H]$^+$, 100)
6d-I-7	74	white powder	144.8−144.9	^{1}H NMR (300 MHz, DMSO) δ 9.76 (s, 1H, −NH−), 9.37 (s, 1H, −NH−), 7.40−7.29 (m, 4H, Ar−H), 7.29−7.16 (m, 4H, Ar−H), 5.68 (s, 1H, −NH−), 3.95 (s, 2H, Ar−CH_2−), 3.78 (s, 2H, −CH_2−Cl), 2.17 (s, 3H, Ar−CH_3), 1.30 (s, 6H, Me_2−C)	HRMS (EI) calcd for $C_{20}H_{25}ClN_3O_2$ [M + H]$^+$ 374.1635, found 374.1634
6d-II-1	71	white powder	207.9−208.5	^{1}H NMR (300 MHz, DMSO) δ 9.74 (s, 1H, −NH−), 9.18 (s, 1H, −NH−), 7.64−7.57 (m, 1H, Ar−H), 7.45−7.39 (m, 1H, Ar−H), 7.35−7.27 (m, 3H, Ar−H), 7.27−7.15 (m, 2H, Ar−H), 5.59 (s, 1H, −NH−), 4.08 (s, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.20 (s, 9H, Me_3−C)	HRMS (EI) calcd for $C_{20}H_{25}ClN_3O_2$ [M + H]$^+$ 374.1635, found 374.1640
6d-II-5	85	white powder	158.7−163.1	^{1}H NMR (300 MHz, DMSO) δ 9.66 (d, J = 6.2 Hz, 1H, −NH−), 9.14 (s, 1H, −NH−), 7.74 (s, 1H, Ar−H), 7.69−7.65 (m, 1H, Ar−H), 7.63−7.53 (m, 2H, Ar−H), 7.36−7.30 (m, 1H, Ar−H), 7.22−7.15 (m, 2H, Ar−H), 5.62 (dd, J = 6.2, 4.7 Hz, 1H, −NH−), 4.05 (d, J = 4.7 Hz, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.20 (s, 9H, Me_3−C)	EI-MS (m/z, %) 408 ([M + H]$^+$, 100)
6d-II-6	83	white powder	179.8−185.7	^{1}H NMR (300 MHz, DMSO) δ 9.71 (d, J = 6.1 Hz, 1H, −NH−), 9.15 (s, 1H, −NH−), 7.64 (d, J = 8.3 Hz, 1H, Ar−H), 7.57 (d, J = 2.1 Hz, 1H, Ar−H), 7.40 (dd, J = 8.3, 2.2 Hz, 1H, Ar−H), 7.33 (dd, J = 7.1, 2.1 Hz, 1H, Ar−H), 7.25−7.16 (m, 2H, Ar−H), 5.63 (dd, J = 6.1, 4.8 Hz, 1H, −NH−), 4.05 (d, J = 4.8 Hz, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.19 (s, 9H, Me_3−C)	EI-MS (m/z, %) 408 ([M + H]$^+$, 100)
6d-II-7	86	white powder	157.0−158.1	^{1}H NMR (300 MHz, DMSO) δ 9.70 (d, J = 6.1 Hz, 1H, −NH−), 9.20 (s, 1H, −NH−), 7.40−7.33 (m, 3H, Ar−H), 7.33−7.28 (m, 2H, Ar−H), 7.28−7.21 (m, 2H, Ar−H), 7.21−7.14 (m, 1H, Ar−H), 5.39 (dd, J = 6.1, 4.9 Hz, 1H, −NH−), 3.95 (d, J = 4.9 Hz, 2H, Ar−CH_2−), 2.13 (s, 3H, Ar−CH_3), 1.21 (s, 9H, Me_3−C)	EI-MS (m/z, %) 340 ([M + H]$^+$, 100)
6d-III-4	73	white powder	135.1−136.0	^{1}H NMR (300 MHz, DMSO) δ 8.38 (s, 1H, Ar−H), 7.98 (dd, J = 8.0, 0.9 Hz, 1H, Ar−H), 7.88 (dd, J = 8.2, 2.5 Hz, 1H, Ar−H), 7.72−7.64 (m, 1H, Ar−H), 7.54 (d, J = 8.2 Hz, 1H, Ar−H), 7.43−7.35 (m, 1H, Ar−H), 6.81 (t, J = 5.5 Hz, 1H, −NH−), 4.16 (d, J = 5.5 Hz, 2H, Ar−CH_2−), 2.81 (s, 2H, −CH_2−C), 2.53 (s, 3H, Ar−CH_3), 1.03 (s, 9H, Me_3−C)	EI-MS (m/z, %) 389 ([M + H]$^+$, 100)
6d-III-5	87	white powder	184.3−185.8	^{1}H NMR (300 MHz, DMSO) δ 9.62 (s, 1H, −NH−), 9.21 (s, 1H, −NH−), 7.75 (s, 1H, Ar−H), 7.71−7.66 (m, 1H, Ar−H), 7.64−7.54 (m, 2H, Ar−H), 7.34−7.28 (m, 1H, Ar−H), 7.20−7.10 (m, 2H, Ar−H), 5.51 (s, 1H, −NH−), 4.05 (s, 2H, Ar−CH_2−), 2.17 (s, 3H, Ar−CH_3), 2.08 (s, 2H, −CH_2−C), 1.02 (s, 9H, Me_3−C)	HRMS (EI) calcd for $C_{22}H_{27}F_3N_3O_2$ [M + H]$^+$ 422.2055, found 422.2046

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Table 3. Structure and Preliminary Biological Activity of the New Compounds

					% mortality		
compd	X	R_1	R_2	R_3	against *A. gossypii*	against *T. cinnabarinus*	against *P. xylostella*
6a-I-1	−Cl	$-CH_3$	$-C(CH_3)_2CH_2Cl$	2-Cl−C_6H_4−	14.5	11	64
6a-I-2	−Cl	$-CH_3$	$-C(CH_3)_2CH_2Cl$	2-F−C_6H_4−	70.2	61.1	100
6a-I-3	−Cl	$-CH_3$	$-C(CH_3)_2CH_2Cl$	4-F−C_6H_4−	81.5	67	64
6a-I-4	−Cl	$-CH_3$	$-C(CH_3)_2CH_2Cl$	2-Cl−C_5H_3N-5−	30.6	47.1	16
6a-I-5	−Cl	$-CH_3$	$-C(CH_3)_2CH_2Cl$	3-CF_3−C_6H_4−	5.1	61.8	52
6a-I-6	−Cl	$-CH_3$	$-C(CH_3)_2CH_2Cl$	2,4-Cl−C_6H_3−	42.3	73.8	
6a-I-7	−Cl	$-CH_3$	$-C(CH_3)_2CH_2Cl$	C_6H_5−	68.5	50.9	52
6a-II-1	−Cl	$-CH_3$	$-C(CH_3)_3$	2-Cl−C_6H_4−	51.7	100	76
6a-II-2	−Cl	$-CH_3$	$-C(CH_3)_3$	2-F−C_6H_4−	38.9	80.5	76
6a-II-3	-Cl	$-CH_3$	$-C(CH_3)_3$	4-F−C_6H_4−	41.5	73.8	88
6a-II-4	−Cl	$-CH_3$	$-C(CH_3)_3$	2−Cl-C_5H_3N-5-	24.2	81.3	50
6a-II-5	−Cl	$-CH_3$	$-C(CH_3)_3$	3-CF_3−C_6H_4−	68.3	8.6	40
6a-II-6	−Cl	$-CH_3$	$-C(CH_3)_3$	2,4-Cl−C_6H_3−	77.8	69	64
6a-II-7	−Cl	$-CH_3$	$-C(CH_3)_3$	C_6H_5−	52.4	92.5	34
6a-III-1	−Cl	$-CH_3$	$-CH_2-C(CH_3)_3$	2-Cl−C_6H_4−	65.5	100	52
6a-III-5	−Cl	$-CH_3$	$-CH_2-C(CH_3)_3$	3-CF_3−C_6H_4−	58.3	77.2	76
6a-III-6	−Cl	$-CH_3$	$-CH_2-C(CH_3)_3$	2,4-Cl−C_6H_3−	62	88.8	88
6a-III-7	−Cl	$-CH_3$	$-CH_2-C(CH_3)_3$	C_6H_5−	70.4	70.1	94
6b-II-3	−Cl	−H	$-C(CH_3)_3$	4-F−C_6H_4−	91.3	73.8	
6b-II-5	−Cl	−H	$-C(CH_3)_3$	3-CF_3−C_6H_4−	28.9	73.8	28
6b-III-2	−Cl	−H	$-CH_2-C(CH_3)_3$	2-F−C_6H_4−	1.8	77.6	28
6c-I-1	−Br	$-CH_3$	$-C(CH_3)_2CH_2Cl$	2-Cl−C_6H_4−	8.5	95.7	76
6c-I-4	−Br	$-CH_3$	$-C(CH_3)_2CH_2Cl$	2-Cl−C_5H_3N-5−	33.3	55.8	52
6c-I-5	−Br	$-CH_3$	$-C(CH_3)_2CH_2Cl$	3-CF_3−C_6H_4−	78.6	72.7	52
6c-I-6	−Br	$-CH_3$	$-C(CH_3)_2CH_2Cl$	2,4-Cl−C_6H_3−	3.8	82.5	70
6c-I-7	−Br	$-CH_3$	$-C(CH_3)_2CH_2Cl$	C_6H_5−	84.6	20.3	52
6c-II-1	−Br	$-CH_3$	$-C(CH_3)_3$	2-Cl−C_6H_4−	49.1	40.1	64
6c-II-4	−Br	$-CH_3$	$-C(CH_3)_3$	2-Cl-C_5H_3N-5−	96.2	19.3	64
6c-II-5	−Br	$-CH_3$	$-C(CH_3)_3$	3-CF_3−C_6H_4−	54.2	19.8	16
6c-II-6	−Br	$-CH_3$	$-C(CH_3)_3$	2,4-Cl−C_6H_3−	90.2	77	34
6c-II-7	−Br	$-CH_3$	$-C(CH_3)_3$	C_6H_5−	86.1	64.9	50
6c-III-1	−Br	$-CH_3$	$-CH_2-C(CH_3)_3$	2-Cl−C_6H_4−	88	11.6	76
6c-III-4	−Br	$-CH_3$	$-CH_2-C(CH_3)_3$	2-Cl−C_5H_3N-5−	25.9	23.4	40
6c-III-5	−Br	$-CH_3$	$-CH_2-C(CH_3)_3$	3-CF_3−C_6H_4−	18.7	100	40
6c-III-7	−Br	$-CH_3$	$-CH_2-C(CH_3)_3$	C_6H_5−	96.6	78.1	22
6d-I-1	−H	$-CH_3$	$-C(CH_3)_2CH_2Cl$	2-Cl−C_6H_4−	68.8	46.7	22
6d-I-7	−H	$-CH_3$	$-C(CH_3)_2CH_2Cl$	C_6H_5−	95.6	79.3	52
6d-II-1	−H	$-CH_3$	$-C(CH_3)_3$	2-Cl−C_6H_4−	92.2	5.9	64
6d-II-5	−H	$-CH_3$	$-C(CH_3)_3$	3-CF_3−C_6H_4−	75	46.8	64
6d-II-6	−H	$-CH_3$	$-C(CH_3)_3$	2,4-Cl−C_6H_3−	89.4	52.9	50
6d-II-7	−H	$-CH_3$	$-C(CH_3)_3$	C_6H_5−	62.3	16.7	52
6d-III-4	−H	$-CH_3$	$-CH_2-C(CH_3)_3$	2-Cl−C_5H_3N-5−	65.5	19.3	100
6d-III-5	−H	$-CH_3$	$-CH_2-C(CH_3)_3$	3-CF_3−C_6H_4−	94.2	57.9	40
imidacloprid					100[a]		
chlorantraniliprole					91.3[b]	29.9[b]	100[c]
pyridaben						100[d]	

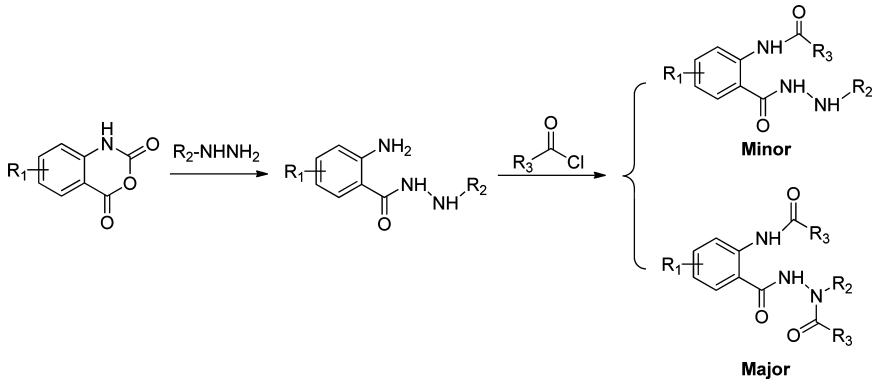
[a]Concentration of imidacloprid was 2 mg L^{-1}. [b]Concentration of chlorantraniliprole was 200 mg L^{-1}. [c]Concentration of chlorantraniliprole was 0.2 mg L^{-1}. [d]Concentration of pyridaben was 50 mg L^{-1}.

added. The white solid precipitates were collected by filtration, washed with petroleum ether (5 mL), and dried to afford 0.39 g of **6a-I-1** as a white powder: yield, 73%; mp, 188.2−190.0 °C; ^{1}H NMR (300 MHz, DMSO) δ 9.91 (s, 1H, −NH−), 9.29 (s, 1H, −NH−), 7.66−7.58 (m, 1H, Ar−H), 7.49−7.40 (m, 2H, Ar−H), 7.38−7.28 (m, 2H, Ar−H), 7.26 (d, J = 2.5 Hz, 1H, Ar−H), 5.54 (s, 1H, −NH−), 4.07 (s, 2H, Ar−CH_2−), 3.77 (s, 2H, $-CH_2-$Cl), 2.16 (s, 3H, Ar−CH_3), 1.28 (s, 6H, Me_2−C); EI-MS (m/z, %) 442 ([M + H]$^+$, 100).

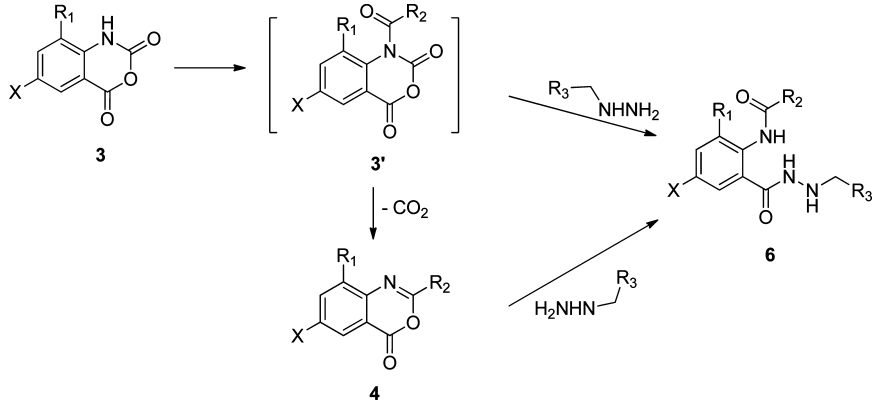
Biological Assay. All bioassays were performed on representative test organisms reared in the laboratory. Each bioassay was repeated at 25 ± 1 °C according to statistical requirements. Assessments were made on a dead/alive basis, and mortality rates were corrected using Abbott's formula.[16] Evaluations were made on the basis of a percentage scale of 0−100, in which 0 = no activity and 100 = total mortality. There was a deviation of ±5% in the values. First, 12 mg of each tested sample was dissolved completely in 2 mL of acetone/methanol (v/v, 1:1) to obtain a solution having a concentration of 6 mg mL^{-1}. Next, the solution was diluted to 600 mg L^{-1} with 18 mL of distilled water containing Triton X-100 (0.05%).

Lethal Activity against Cotton Aphid (Aphis gossypii Glover). The lethal activities of the new compounds against cotton aphid were evaluated according to the reported procedure.[17,18] More than 30

Scheme 2



Scheme 3



healthy apterous third-instar nymphae on tender *Hibiscus syriacus* leaves were dipped into the diluted solutions of the compounds for 5 s; the superfluous fluid on the leaves was then removed. The treated insects were kept in an illumination box at 25 ± 1 °C, and mortality was recorded after 24 h.

Lethal Activity against Carmine Spider Mite (Tetranychus cinnabarinus). The larvicidal activities of the new compounds against carmine spider mites were assessed according to the reported procedure.[19,20] More than 20 third-instar mite larvae on *Phaseolus vulgaris* leaves were dipped in the diluted solutions of related chemicals for 5 s; the superfluous liquor on the leaves was then removed. The larvae were kept in an illumination box at 25 ± 1 °C, and mortality was recorded 48 h after treatment.

Lethal Activity against Diamondback Moth (Plutella xylostella). The larvicidal activities of the new compounds against the diamondback moth were tested according to the reported procedure.[18,21] More than 20 second-instar moth larvae on cruciferous vegetables leaves were dipped in the diluted solutions of the compounds for 5 s; the superfluous liquor was then removed. The larvae were kept in an illumination box at 25 ± 1 °C, and mortality was recorded 48 h after treatment.

Commercial pesticides chlorantraniliprole 95% (0.2 mg L^{-1}), imidacloprid 95% (2 mg L^{-1}), and pyridaben 97.1% (50 mg L^{-1}) were used as positive controls against *P. xylostella*, *A. gossypii*, and *T. cinnabarinus*, respectively, and were treated under the same conditions with acetone, methanol, and water containing Triton X-100 (0.05%). Each test was performed in triplicate. The biological activities from the preliminary assay are summarized in Table 3.

RESULTS AND DISCUSSION

Synthesis of Compounds. For the synthesis of new anthranilic diamide compounds, the substituted amines were reacted with isatoic anhydrides,[3] followed by acylation with acyl chloride (Scheme 2). This method yielded many disubstituted byproducts.

To avoid the formation of disubstituted byproducts, another synthetic pathway (Scheme 1) was designed, in which isatoic anhydrides were first treated with acyl chlorides to form intermediate *N*-acyl-substituted-1*H*-benzo[*d*][1,3]oxazine-2,4-diones (**3′**) and then treated with benzylhydrazines to afford the target compounds. Target compounds **6** were obtained readily and efficiently without the formation of disubstituted byproducts. When intermediates **3′** and final compounds **6** were analyzed using mass spectrum, the masses of the final compounds **6** were confirmed to be accurate. However, the mass of **3′** compounds was 44 less than that expected. Characterization with ^{13}C NMR and corresponding analysis revealed that the intermediate **3′** formed during the reaction was not stable and could undergo an internal molecular transformation to form compounds **4** after releasing one molecule of carbon dioxide as shown in Scheme 3.

The acyl chlorides were first reacted with 6- and/or 8-substituted 1*H*-benzo[*d*][1,3]oxazine-2,4-dione to afford **4a-I−4d-III**; the structures of these intermediates were confirmed by ^{1}H NMR, ^{13}C NMR, and EI-MS. To our knowledge, these have been reported for the first time. Next, the compounds were reacted with substituted benzylhydrazine to efficiently form the corresponding new compounds **6a-I-1−6c-III-7**.

Biological Assay. The results from the assay on the three representative organisms showed that all of the new compounds exhibited biological activities to a certain extent at a concentration of 600 mg L^{-1}, and the substituted groups at positions X, R_1, R_2, and R_3 had various influences on mortality. The data presented in Table 3 suggest that the relationship

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Table 4. Inhibition Activity against Carmine Spider Mite of Four Synthetic Compounds at Five Concentrations Compared with Chlorantraniliprole and Pyridaben

	lethal activity (%) against *T. cinnabarinus*					
compd	400 mg L^{-1}	200 mg L^{-1}	100 mg L^{-1}	50 mg L^{-1}	25 mg L^{-1}	LC_{50} (mg L^{-1})
6a-II-1	97.8	82.2	60.8	62.5	30.9	43.8
6a-III-1	92.2	86.4	80.4	74.1	38.4	27.9
6c-I-1	89.6	83.6	62.6	44.9	36.5	51.7
6c-III-5	93.3	64.4	43.3	38.4	26.0	89.8
chlorantraniliprole	40.7	29.9	17.2	13.2	7.4	632.9
pyridaben	100	100	100	100	97.7	5.5

between the substituted groups and biological activities could be summarized as below.

Lethal Activity against Carmine Diamondback Moth (*P. xylostella*). Of the compounds produced by the structure modification via the introduction of benzylhydrazine, only compounds **6a-I-2** and **6d-III-4** showed 100% mortality against *P. xylostella* at 600 mg L^{-1}; the other compounds showed markedly weaker lethal activity than that exerted by the positive control 95% chlorantraniliprole at 0.2 mg L^{-1}. This suggested that the replacement of the methyl group by a hydrogen atom at position R_1 remarkably reduced the mortality. Moreover, when the test concentration was lowered to 200 mg L^{-1}, all new compounds showed very weak lethal activity.

The tested moths began to die 24 h after treatment, and only a small part of the corpses changed to yellow or black. This death pattern resembled that when moths are poisoned by chlorantraniliprole.

Lethal Activity against Cotton Aphid (*A. gossypii*). The replacement of the hydrogen atom with chlorine or bromine atom at X position on anthranilic diamides reduced the lethal activity. For example, the lethal activities of compounds **6d** were higher than those of the corresponding compounds **6a** and **6c**; in particular, the mortality rate after treatment with **6d-III-5** reached 94.2%, but those after treatment with **6c-III-5** and **6a-III-5** were only 18.7 and 58.3%, respectively. Of the three different groups, *tert*-butyl, chlorinated *tert*-butyl (2-chloro-1,1-dimethylethyl), and neopentyl at position R_2, the chlorinated *tert*-butyl group remarkably decreased the lethal activities of **6a-I-1**, **6c-I-1**, **6a-I-5**, and **6c-I-6**. The replacement of neopentyl with *tert*-butyl did not improve the lethal activity of the compounds; for instance, the lethal activity against *A. gossypii* of **6b-III-2**, **6c-III-4**, and **6c-III-5** was apparently decreased. At position R_3, the introduction of a chlorine atom, fluorine atom, and trifluoromethyl group on the aromatic groups could slightly improve the lethal activity.

Overall, when X was a hydrogen or bromine atom, most of the compounds had good lethal activity, and especially, the mortality rates for **6d-II-1**, **6c-II-4**, **6d-III-5**, **6c-II-6**, **6c-III-7**, and **6d-I-7** reached >90%. Thus, the anthranilic diamides containing benzylhydrazine derivatives with no substitution at position X and a phenyl group at R_3 showed markedly improved lethal activity against aphids.

Lethal Activity against Carmine Spider Mite (*T. cinnabarinus*). Chlorination and bromination at position X have crucial effects on the acaricidal activity; introduction of a chlorine or bromine atom at X position on the skeleton structure of anthranilic diamides could remarkably improve the acaricidal activity. For instance, when the hydrogen atom at position X of compounds **6d-II-1** and **6d-I-1** was only replaced by chlorine and bromine atoms, the mortality rates for **6a-II-1** and **6c-I-1** against *T. cinnabarinus* were increased to 100 and 95.7% from 5.9 and 46.7%, respectively. The substituted groups at position R_2 had impacts on the acaricidal activity depending on the atoms that were substituted at position X. When a chlorine atom was substituted at position X, compounds **6a-II-1**, **6a-III-1**, **6a-II-2**, and **6a-III-6** with *tert*-butyl or neopentyl group showed stronger lethal activities against *T. cinnabarinus* than compounds **6a-I-1**, **6a-I-2**, and **6a-I-6** with a 2-chloro-1,1-dimethylethyl group. On the other hand, when a bromine atom was located at position X, compounds containing 2-chloro-1,1-dimethylethyl exhibited better acaricidal activity. Among the various substituted aromatic groups at position R_3, only 2-chlorophenyl or 3-trifluoromethylphenyl could slightly improve the lethal activity. Hence, modifying the R_2 and R_3 groups and retaining the chlorine and bromine atoms at position X might effectively improve the acaricidal activity of the new class of compounds.

The acaricidal activity of the four compounds **6a-I-1**, **6a-III-1**, **6c-I-1**, and **6c-III-5**, which exhibited good lethal activity against *T. cinnabarinus* at a concentration of 600 mg L^{-1}, was further investigated by conducting the assay at concentrations of 400, 200, 100, 50, and 25 mg L^{-1}. The lethal activity levels and calculated LC_{50} values are shown in Table 4. The results showed that the four compounds exhibited promising acaricidal activity even at lower concentrations; for instance, **6a-II-1** and **6a-III-1** showed 62.5 and 74.1% mortality at 50 mg L^{-1}, respectively, and the LC_{50} of compound **6a-III-1** was only 27.9 mg L^{-1}.

In all of the tests, the mite larvae stopped moving after 1 h of treatment, and many larvae died within 24 h after treatment. The corpses changed to black after 48 h of treatment. This suggests that the mode of action of these new compounds against *T. cinnabarinus* could be different from that of acylhydrazine compounds.

In conclusion, more than 40 2-acylamino-substituted *N*′-benzylbenzohydrazide derivatives were synthesized and characterized in this study. Some new compounds exhibited good lethal activities against *A. gossypii*, *P. xylostella*, and *T. cinnabarinus*. In particular, four compounds, **6a-II-1**, **6a-III-1**, **6c-I-1**, and **6c-III-5**, exhibited promising lethal activity against *T. cinnabarinus*, and their LC_{50} values were below 90 mg L^{-1}. Compound **6a-III-1**(X = Cl, R_1 = CH_3, R_2 = *tert*-butyl, R_2 = 2-chlorophenyl) possessed the best LC_{50} value of 27.9 mg L^{-1}, which is not close to that of a commercial acracidal agent pyridaben (5.5 mg L^{-1}), but it is much better than that of chlorantraniliprole (632.9 mg L^{-1}). This finding indicates that the incorporation of the acylhydrazine fragment could improve the acaricidal activity of the anthranilic diamide compounds as expected and warrant further studies for designing new molecules with good biological activity.

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Notes

The authors declare no competing financial interest.

ABBREVIATIONS USED

HRMS, high-resolution mass spectrometry; MS, mass spectrometry; DMF, *N*,*N*-dimethylformamide; NCS, *N*-chlorosuccinimide; NBS, *N*-bromosuccinimide; THF, tetrahydrofuran; TLC, thin layer chromatography; NMR, nuclear magnetic resonance.

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