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Design, Synthesis, and Antiviral, Fungicidal, and Insecticidal Activities of Tetrahydro-β-carboline-3-carbohydrazide Derivatives

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ABSTRACT: According to our previous research on the antiviral activity of β-carboline and tetrahydro-β-carboline derivatives, using (1*S*,3*S*)-1-methyl-2,3,4,9-tetrahydro-1*H*-pyrido[3,4-*b*]indole-3-carbohydrazide (**1**) as a lead compound, series of novel tetrahydro-β-carboline derivatives containing acylhydrazone moiety were designed, synthesized, and first evaluated for their biological activities. Most of these compounds exhibited excellent antiviral activity both *in vitro* and *in vivo*. The *in vivo* inactivation, curative, and protection activities of compounds **8**, **9**, **12**, **16**, **28**, **29**, and **30** were much higher than that of ribavirin (37.6%, 39.4%, and 37.9% at 500 μg/mL) and the lead compound (40.0%, 42.3%, and 39.6% at 500 μg/mL). Especially, the *in vitro* and *in vivo* activities of compound **16** (36.9%, 33.6%, 30.2%, and 35.8%) at 100 μg/mL, which were very close to that of ribavirin (40.0% for *in vitro* activity) at 500 μg/mL. Compounds **9** and **29** were chosen for the field trials of antiviral efficacy against TMV (tobacco mosaic virus); the results exhibited that both compounds, especially compound **29**, showed better activities than control plant virus inhibitors. At the same time, the fungicidal results showed that compounds **6**, **9**, and **11** exhibited good fungicidal activities against 14 kinds of phytopathogens. Additionally, compounds **3** and **23** exhibited moderate insecticidal activity against the four tested species of insects.

KEYWORDS: *tetrahydro-β-carboline, acylhydrazone, antiviral activity, antifungal activity, insecticidal activity, field trials*

INTRODUCTION

Botanical pesticides refer to the pesticides that are developed using plant resources. They may be the exact active ingredients extracted from plants or the compounds deried from the active compounds. Because most of them are safe to untargetted beings and they could be rapidly degraded to reduce the environment risk of residues, botanical pesticides have long been touted as attractive alternatives and have become one of the preferred green biopesticides.[1] However, some deficiencies, such as being too complex to synthesize and active ingredients being easy to decompose, inevitably limit the widespread use of botanical pesticides.[2,3] Based on the above reasons, our group was committed to the research on developing novel effective pesticides using active substances of plant origin as precursors to solve these problems.

β-Carboline alkaloids are widely distributed in nature. Due to their broad spectrum of biological activities,[4−9] β-carboline alkaloids have attracted great attention of pharmaceutical chemists. In our previous work, we found that tetrahydro-β-carboline-3-carbohydrazide **1** exhibited excellent anti-TMV activity. The antiviral activity against tobacco mosaic virus (TMV) in vitro and inactivation, curative, and protection activities in vivo were 40.4%, 40.0%, 42.3%, and 39.6%, respectively, at 500 μg/mL.

The biological activities of acylhydrazone compounds had always been the hot spots on pharmacology research.[10−13] Acylhydrazone compounds (Figure 1) containing active fragment (−CONHN=CH−) widely exist in drug molecules, which show good insecticidal or herbicidal activities such as pymetrozine,[14] metaflumizone,[15] and diflufenzopyr.[16] In this work, by adopting the tactics of active fragment stitching and using compound **1** as the lead compound, we designed and synthesized a series of novel tetrahydro-β-carboline derivatives containing acylhydrazone moiety (Figure 2). Meanwhile, their anti-TMV, fungicidal, and insecticidal activities were evaluated for the first time. Based on the comprehensive analysis of bioactivity, synthetic procedure, physical properties, and so on, compounds **9** and **29** were chosen for the field trials of antiviral efficacy against TMV. Both compounds exhibited better activities than control plant virus inhibitors, which indicated that **9** and **29** exhibited a promising application prospect as new candicates for controlling TMV in the field.

MATERIALS AND METHODS

Instruments. ^{1}H NMR spectra were obtained at 400 MHz using a Bruker AV400 spectrometer in $CDCl_3$ or DMSO-d_6 solution with tetramethylsilane as the internal standard. HRMS data were obtained on an FTICR-MS instrument (Ionspec 7.0 T). The melting points were determined on an X-4 binocular microscope melting point apparatus and are uncorrected.

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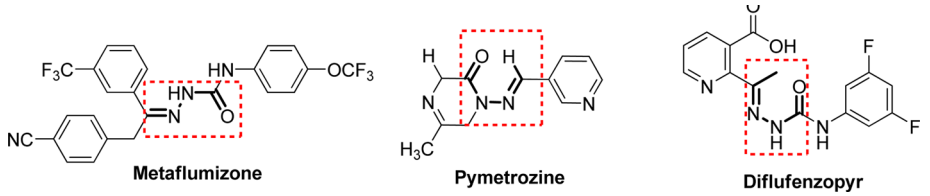

Figure 1. Acylhydrazone compounds.

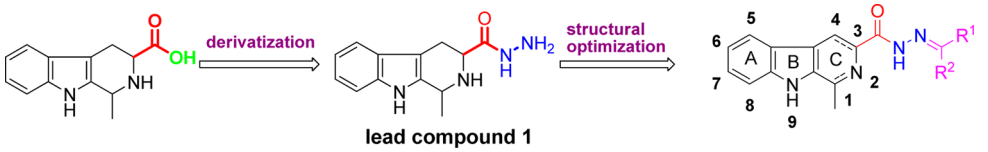

Figure 2. Design of target compounds.

General Synthesis. All anhydrous solvents were dried and purified by standard techniques. The synthetic routes are given in Scheme 2.

Synthesis of (3*S*)-1-Methyl-2,3,4,9-tetrahydro-1*H*-pyrido-[3,4-*b*]indole-3-carbohydrazide (1). This compound was synthesized by the method[17] reported in 99% yield: mp =100−103 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.15 (s, 1H, NH), 7.90 (s, 1H, NHCO), 7.50 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.32 (d, $^3J_{HH}$ = 8.0 Hz, 1H, Ar−H), 7.17 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 7.11 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 4.18−4.23 (m, 1H, C**H**CH$_3$), 3.93 (br, 2H, NH$_2$), 3.65 (q, $^3J_{HH}$ = 8.4 Hz, 1H, C**H**CO), 3.26−3.32 (m, 1H, C**H**$_2$), 2.71−2.78 (m, 1H, C**H**$_2$), 1.48 (d, $^3J_{HH}$ = 6.8 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 172.0, 137.9, 135.8, 126.9, 120.5, 118.3, 117.4, 110.9, 106.2, 56.1, 48.2, 25.4, 20.0; HRMS (ESI) calcd for $C_{13}H_{17}N_4O$ [M + H]$^+$ 245.1397, found 245.1398.

Synthesis of 1-Methyl-9*H*-carbazole-3-carboxylic Acid Hydrazide (2). A mixture of ethyl 1-methyl-9*H*-pyrido[3,4-*b*]indole-3-carboxylate[18] (2.00 g, 7.86 mmol) and hydrazine hydrate (80%) (3.15 g, 5.04 mmol) in a mixed solvent of alcohol (80 mL) and DMF (25 mL) was heated at reflux for 24 h; then the mixture was concentrated in vacuo. Solid was precipitated out and collected by filtration. The cake was extracted with boiling alcohol (each 30 mL) for several times until the filter cake did not dissolve in hot alcohol any more. The combined extract was concentrated to give **2** as a light-pink solid (0.90 g, 48%): mp =242−244 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 11.92 (s, 1H, NH), 9.51 (s, 1H, O=C-NH), 8.64 (s, 1H, Ar−H), 8.35 (d, $^3J_{HH}$ = 8.1 Hz, 1H, Ar−H)), 7.64 (d, $^3J_{HH}$ = 8.1 Hz, 1H, Ar−H), 7.57 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 7.28 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 4.55 (s, 2H, NH$_2$), 2.82 (s, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 164.1, 141.0, 140.7, 138.7, 135.7, 128.3, 127.3, 122.2, 121.4, 119.9, 112.2, 111.9, 20.4; HRMS (ESI) calcd for $C_{13}H_{13}N_4O$ [M + H]$^+$ 241.1084, found 241.1084.

Synthesis of *N*′-Benzylidene-(3*S*)-1-methyl-2,3,4,9-tetrahydro-1*H*-pyrido[3,4-*b*]indole-3-carbohydrazide (3). To a slurry of compound **1** (0.50 g, 2.05 mmol) in toluene (35 mL) was added dropwise benzaldehyde (0.44 g, 4.10 mmol) at room temperature, Then the reaction mixture was heated to reflux. After 5 h, TLC analysis showed the complete consumption of compound **1**. The reaction solution was cooled to room temperature, and solid was precipitated out. The solid was filtrated and washed with toluene and then stirred in absolute ether for 1.5 h to remove the remaining toluene. Filtered once again to give **3** as a yellow solid (0.50 g, 74%): mp = 200−204 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.48 and 11.35 (s, 1H, NH), 10.84 and 10.81 (s, 1H, O=C-NH), 8.34 and 8.06 (s, 1H, N=CH), 7.71 and 7.61 (dd, $^3J_{HH}$ = 7.6 Hz, $^4J_{HH}$ = 1.6 Hz, 1H, Ph-H), 7.49−7.42 (m, 2H, Ph-H), 7.42−7.33 (m, 2H, Ph-H and Ar−H), 7.31 and 7.30 (d, $^3J_{HH}$ = 8.0 Hz, 1H, Ar−H), 7.03 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 6.95 and 6.92 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 4.48 and 3.61 (dd, $^3J_{HH}$ = 10.8 Hz, 3.6 Hz, 1H, C**H**), 4.15 (q, $^3J_{HH}$ = 8.0 Hz, 1H, C**H**), 2.98−2.89 (m, 1H, C**H**$_2$), 2.71 and 2.62 (ddd, $^2J_{HH}$ = 14.4 Hz, $^3J_{HH}$ = 11.6 Hz, $^4J_{HH}$ = 2.0 Hz, 1H, C**H**$_2$), 1.45 (d, $^3J_{HH}$ = 6.8 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.6 and 169.1, 147.0 and 143.4, 137.9, 135.8, 134.4 and 134.1, 130.0 and 129.8, 128.9 and 128.8, 127.1 and 126.7, 126.9 and 126.9, 120.6, 118.4, 117.5, 111.0, 106.2 and 106.1, 56.7 and 53.3, 48.4 and 47.7, 25.5 and 25.4, 20.0; HRMS (ESI) calcd for $C_{20}H_{21}N_4O$ [M + H]$^+$ 333.1710, found 333.1715.

Compounds **4**−**26** were synthesized using the similar procedure as compound **3**.

*Data for 1-Methyl-9H-b-carboline-3-carboxylic Acid Benzylidene-hydrazide (**4**).* This compound was obtained as a yellow solid in 91% yield: mp = 303−306 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 12.05 (s, 1H, NH), 11.85 (s, 1H, O=C-NH), 8.82 (s, 1H, N=CH), 8.69 (s, 1H, Ar−H), 8.41 (d, $^3J_{HH}$ = 7.8 Hz, 1H, Ar−H), 7.77 (d, $^3J_{HH}$ = 6.3 Hz, 2H, Ph-H), 7.67 (d, $^3J_{HH}$ = 8.1 Hz, 1H, Ar−H), 7.60 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 7.55−7.41 (m, 3H, Ph-H), 7.31 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 2.92 (s, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 161.2, 147.9, 141.1, 140.8, 138.2, 136.1, 134.6, 129.9, 128.9, 128.4, 127.5, 127.1, 122.3, 121.4, 120.1, 113.2, 112.3, 20.4; HRMS (ESI) calcd for $C_{20}H_{17}N_4O$ [M + H]$^+$ 329.1397, found 329.1402.

*Data for N′-Benzylidene-(3R)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**5**).* This compound was obtained as a light yellow solid in 76% yield: mp = 197−200 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.67 and 11.47 (s, 1H, NH), 10.98 and 10.86 (s, 1H, O=C−NH), 8.35 and 8.09 (s, 1H, N=CH), 7.75−7.69 (m, 1H, Ph-H), 7.66−7.60 (m, 1H, Ph-H), 7.50−7.41 (m, 2H, Ph-H), 7.41−7.36 (m, 2H, Ph-H and Ar−H), 7.33 and 7.31 (d, $^3J_{HH}$ = 8.8 Hz, 1H, Ph-H), 7.06 and 7.04 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 7.00−6.92 (m, 1H, Ar−H), 4.70−4.61 and 3.69 (dd, $^3J_{HH}$ = 10.4 Hz, 3.6 Hz, 1H, C**H**), 4.42−4.14 (m, 1H, C**H**), 3.08 and 2.98 (dd, $^2J_{HH}$ = 14.8 Hz, $^3J_{HH}$ = 2.8 Hz, 1H, C**H**), 2.79−2.67 (m, 1H, C**H**$_2$), 1.53 and 1.48 (d, $^3J_{HH}$ = 6.8 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 173.4 and 168.8, 147.2 and 144.1, 137.2, 136.0 and 135.9, 134.3 and 134.0, 130.0 and 130.0, 128.8, 127.1, 126.8 and 126.6, 120.9 and 120.7, 118.6 and 118.5, 117.7 and 117.5, 111.1 and 111.1, 106.0 and 105.7, 56.6 and 53.6, 48.5 and 48.1, 25.3 and 24.7, 19.7 and 19.0; HRMS (ESI) calcd for $C_{20}H_{21}N_4O$ [M + H]$^+$ 333.1710, found 333.1708.

*Data for N′-(4-tert-Butylbenzylidene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**6**).* This compound was obtained as a yellow solid in 72% yield: mp = 139−143 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.47 and 11.32 (s, 1H, NH), 10.87 and 10.82 (s, 1H, O=C-NH), 8.31 and 8.03 (s, 1H, N=CH), 7.88 and 7.70−7.27 (m, 6H, Ph-H and Ar−H), 7.04 (t, $^3J_{HH}$ = 6.8 Hz, 1H, Ar−H), 6.96 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 4.48 and 3.62 (dd, $^3J_{HH}$ = 10.4 Hz, 7.2 Hz, 1H, C**H**), 4.27−4.08 (m, 1H, C**H**), 3.02−2.88 (m, 1H, C**H**$_2$), 2.77−2.59 (m, 1H, C**H**$_2$), 1.52−1.35 (m, 3H, C**H**$_3$), 1.30 and 1.24 (s, 9H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.3 and 168.9, 152.8 and 152.6, 147.0 and 143.3, 137.8, 135.8, 131.6 and 131.4, 129.2, 126.9 and 126.5, 125.7 and 125.6, 120.6 and 120.6, 118.4, 117.5 and 117.5, 111.0, 106.2 and 106.0, 56.7 and 53.5, 48.4 and 47.7, 34.6 and 34.5, 31.0 and 30.9, 25.5 and 25.2, 19.9 and 19.9; HRMS (ESI) calcd for $C_{24}H_{29}N_4O$ [M + H]$^+$ 389.2336, found 389.2338.

*Data for N′-(4-(Dimethylamino)benzylidene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**7**).*

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This compound was obtained as a yellow solid in 73% yield: mp = 215−220 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.20 and 11.05 (s, 1H, NH), 10.84 and 10.80 (s, 1H, O═C-NH), 8.16 and 7.92 (s, 1H, N═CH), 7.52 and 7.36 (d, $^3J_{HH}$ = 7.6 Hz, 2H, Ph-H), 7.44−7.38 (m, 1H, Ar−H), 7.30 (d, $^3J_{HH}$ = 6.4 Hz, 1H, Ar−H), 7.03 (t, $^3J_{HH}$ = 6.8 Hz, 1H, Ar−H), 6.95 (t, $^3J_{HH}$ = 6.8 Hz, 1H, Ar−H), 6.75 and 6.67 (d, $^3J_{HH}$ = 7.6 Hz, 2H, Ph-H), 4.44 and 3.57 (d, $^3J_{HH}$ = 8.8 Hz, 1H, C**H**), 4.23−4.08 (m, 1H, C**H**), 3.07−2.86 (m, 7H, N−C**H**$_3$ and C**H**$_2$), 2.74−2.56 (m, 1H, C**H**$_2$), 1.52−1.38 (m, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 173.8 and 168.5, 151.5 and 151.3, 147.8 and 144.2, 137.8, 135.8, 128.4 and 127.9, 126.9, 121.6 and 121.5, 120.6 and 120.5, 118.4, 117.5, 111.9 and 111.8, 111.0, 106.2 and 106.1, 56.6 and 53.5, 48.4 and 47.7, 39.8, 25.6 and 25.2, 19.9; HRMS (ESI) calcd for $C_{22}H_{26}N_5O$ $[M + H]^+$ 376.2132, found 376.2137.

*Data for N′-(4-Nitrobenzylidene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**8**).* This compound was obtained as a yellow solid in 74% yield: mp = 222−227 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.81 and 11.68 (s, 1H, NH), 10.87 and 10.82 (s, 1H, O═C-NH), 8.46 and 8.17 (s, 1H, N═CH), 8.31 and 8.22 (d, $^3J_{HH}$ = 8.4 Hz, 2H, Ph-H), 7.98 and 7.88 (d, $^3J_{HH}$ = 8.4 Hz, 2H, Ph-H), 7.41 and 7.36 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.31 (d, $^3J_{HH}$ = 6.4 Hz, 1H, Ar−H), 7.04 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 6.96 (t, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 4.55 and 3.67 (d, $^3J_{HH}$ = 8.8 Hz, 1H, C**H**), 4.25−4.10 (m, 1H, C**H**), 2.96 (d, $^2J_{HH}$ = 13.6 Hz, 1H, C**H**$_2$), 2.79−2.60 (m, 1H, C**H**$_2$), 1.47 (d, $^3J_{HH}$ = 6.4 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.8 and 169.6, 147.8 and 147.7, 144.6, 140.7 and 140.4, 137.8 and 137.5, 135.9, 128.0 and 127.7, 126.9, 124.1, 120.7 and 120.6, 118.5, 117.6 and 117.5, 111.0, 106.1 and 106.0, 56.8 and 53.2, 48.5 and 47.8, 25.4 and 25.4, 19.9 and 19.8; HRMS (ESI) calcd for $C_{20}H_{20}N_5O_3$ $[M + H]^+$ 378.1561, found 378.1563.

*Data for N′-(4-Chlorobenzylidene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**9**).* This compound was obtained as a yellow solid in 81% yield: mp = 140−145 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.62 and 11.51 (s, 1H, NH), 10.91 and 10.85 (s, 1H, O═C-NH), 8.35 and 8.07 (s, 1H, N═CH), 7.74 and 7.64 (d, $^3J_{HH}$ = 7.2 Hz, 2H, Ph-H), 7.58−7.35 (m, 3H, Ph-H and Ar−H), 7.35−7.28 (m, 1H, Ar−H), 7.10−7.00 (m, 1H, Ar−H), 7.00−6.90 (m, 1H, Ar−H), 4.56 and 3.66 (d, $^3J_{HH}$ = 9.6 Hz, 1H, C**H**), 4.30−4.12 (m, 1H, C**H**), 3.03−2.90 (m, 1H, C**H**$_2$), 2.79−2.62 (m, 1H, C**H**$_2$), 1.54−1.39 (m, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.2 and 169.1, 145.8 and 142.4, 137.6 and 135.9, 134.5 and 134.3, 133.3 and 133.0, 129.0, 128.7, 128.4, 126.9 and 126.8, 120.7 and 120.6, 118.5 and 118.4, 117.6 and 117.5, 111.0, 106.1 and 105.9, 56.7 and 53.3, 48.5 and 47.8, 25.4 and 25.1, 19.8 and 19.6; HRMS (ESI) calcd for $C_{20}H_{20}N_4OCl$ $[M + H]^+$ 367.1320, found 367.1323.

*Data for N′-(2,4-Dichlorobenzylidene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**10**).* This compound was obtained as a light yellow solid in 85% yield: mp = 211−213 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.82 and 11.77 (s, 1H, NH), 10.91 and 10.84 (s, 1H, O═C-NH), 8.71 and 8.42 (s, 1H, N═CH), 8.00 and 7.85 (d, $^3J_{HH}$ = 8.4 Hz, 1H, Ph-H), 7.73 and 7.71 (d, $^4J_{HH}$ = 2.0 Hz, 1H, Ph-H), 7.53 and 7.43−7.35 (dd, $^3J_{HH}$ = 8.4 Hz, $^4J_{HH}$ = 2.0 Hz, 1H, Ph-H), 7.41 and 7.37 (d, $^3J_{HH}$ = 8.0 Hz, 1H, Ar−H), 7.32 and 7.31 (d, $^3J_{HH}$ = 8.0 Hz, 1H, Ar−H), 7.04 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 6.96 and 6.94 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 4.65−4.56 and 3.66 (dd, $^3J_{HH}$ = 10.4 Hz, 3.6 Hz, 1H, C**H**), 4.35−4.13 (m, 1H, C**H**), 3.06−2.91 (m, 1H, C**H**$_2$), 2.78−2.64 (m, 1H, C**H**$_2$), 1.54−1.43 (m, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.0 and 169.2, 142.0 and 139.0, 137.5 and 137.5, 135.9, 135.0 and 134.9, 133.8 and 133.7, 130.8 and 130.4, 129.4 and 129.4, 128.1 and 128.1, 128.1 and 127.8, 126.8 and 126.7, 120.8 and 120.6, 118.5 and 118.5, 117.7 and 117.5, 111.1 and 111.0, 106.1 and 105.8, 56.7 and 53.4, 48.5 and 48.0, 25.3 and 25.0, 19.8 and 19.3; HRMS (ESI) calcd for $C_{20}H_{19}N_4OCl_2$ $[M + H]^+$ 401.0931, found 401.0929.

*Data for N′-(3,4-Dichlorobenzylidene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**11**).* This compound was obtained as a light yellow solid in 79% yield: mp = 189−193 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.84 and 11.67 (s, 1H, NH), 10.98 and 10.86 (s, 1H, O═C-NH), 8.34 and 8.07 (s, 1H, N═CH), 7.94 and 7.88 (s, 1H, Ph-H), 7.72 and 7.64 (s, 2H, Ph-H), 7.41 and 7.39 (d, $^3J_{HH}$ = 8.4 Hz, 1H, Ar−H), 7.33 and 7.31 (d, $^3J_{HH}$ = 8.4 Hz, 1H, Ar−H), 7.06 and 7.04 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 6.96 and 6.96 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 4.68 and 3.70 (dd, $^3J_{HH}$ = 10.4 Hz, 4.4 Hz, 1H, C**H**), 4.38 and 4.19 (q, $^3J_{HH}$ = 6.4 Hz, 1H, C**H**), 3.06 and 2.98 (dd, $^2J_{HH}$ = 14.4 Hz, $^3J_{HH}$ = 3.6 Hz, 1H, C**H**$_2$), 2.80−2.66 (m, 1H, C**H**$_2$), 1.53 and 1.48 (d, $^3J_{HH}$ = 6.4 Hz, 3H, CH$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 173.5 and 169.0, 144.5 and 141.6, 137.2, 135.9 and 135.9, 135.2 and 134.8, 132.2 and 132.1, 131.7, 131.1, 128.6 and 128.5, 126.9 and 126.8, 126.6 and 126.4, 120.9 and 120.7, 118.6 and 118.5, 117.7 and 117.5, 111.1 and 111.0, 105.9 and 105.6, 56.6 and 53.5, 48.5 and 48.1, 25.3 and 24.7, 19.6 and 18.9; HRMS (ESI) calcd for $C_{20}H_{19}N_4OCl_2$ $[M + H]^+$ 401.0931, found 401.0934.

*Data for N′-(4-Methoxybenzylidene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**12**).* This compound was obtained as a yellow solid in 69% yield: mp = 138−143 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.35 and 11.22 (s, 1H, NH), 10.84 and 10.81 (s, 1H, O═C-NH), 8.27 and 8.00 (s, 1H, N═CH), 7.65 and 7.55 (d, $^3J_{HH}$ = 8.0 Hz, 2H, Ph-H), 7.40 and 7.36 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.30 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.08−6.89 (m, 4H, Ar−H and Ph-H), 4.45 and 3.59 (d, $^3J_{HH}$ = 8.8 Hz, 1H, C**H**), 4.20−4.08 (m, 1H, C**H**), 3.81 and 3.74 (s, 3H, O−C**H**$_3$), 2.93 (d, $^2J_{HH}$ = 14.4 Hz, 1H, C**H**$_2$), 2.76−2.56 (m, 1H, C**H**$_2$), 1.45 (d, $^3J_{HH}$ = 6.0 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.3 and 168.8, 160.8 and 160.6, 146.9 and 143.2, 137.9, 135.8 and 135.8, 128.6 and 128.2, 127.0 and 126.9, 126.7, 120.5, 118.4, 117.5, 114.3, 111.0, 106.2 and 106.1, 56.7 and 53.3, 55.3 and 55.2, 48.4 and 47.6, 25.5 and 25.4, 20.0 and 19.9; HRMS (ESI) calcd for $C_{21}H_{23}N_4O_2$ $[M + H]^+$ 363.1816, found 363.1819.

*Data for N′-(3-Methoxybenzylidene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**13**).* This compound was obtained as yellow solid in 63% yield: mp = 186−190 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.54 and 11.39 (s, 1H, NH), 10.87 and 10.83 (s, 1H, O═C-NH), 8.32 and 8.03 (s, 1H, N═CH), 7.45−7.11 (m, 5H, Ph-H and Ar−H), 7.08−6.89 (m, 3H, Ar−H and Ph-H), 4.48 and 3.63 (d, $^3J_{HH}$ = 8.0 Hz, 1H, C**H**), 4.25−4.10 (m, 1H, C**H**), 3.81 and 3.69 (s, 3H, O−C**H**$_3$), 3.02−2.89 (m, 1H, C**H**$_2$), 2.77−2.60 (m, 1H, C**H**$_2$), 1.46 (d, $^3J_{HH}$ = 5.2 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.3 and 169.2, 159.6, 147.1 and 143.4, 137.8 and 137.5, 135.9 and 135.8, 135.6, 130.0 and 130.0, 126.9, 120.7 and 120.6, 120.0 and 119.1, 118.4, 117.5, 116.2 and 115.4, 112.0 and 111.3, 111.0, 106.2 and 106.0, 56.7 and 53.6, 55.2 and 55.1, 48.5 and 47.8, 25.5 and 25.2, 19.9 and 19.8; HRMS (ESI) calcd for $C_{21}H_{23}N_4O_2$ $[M + H]^+$ 363.1816, found 363.1818.

*Data for N′-(2-Methoxybenzylidene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**14**).* This compound was obtained as yellow solid in 82% yield: mp = 180−183 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.52 and 11.46 (s, 1H, NH), 10.90 and 10.84 (s, 1H, O═C-NH), 8.66 and 8.41 (s, 1H, N═CH), 7.84 and 7.68 (d, $^3J_{HH}$ = 7.2 Hz, 1H, Ph-H), 7.41 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.37 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.31 (dd, $^3J_{HH}$ = 8.0 Hz, 4.0 Hz, 1H, Ph-H), 7.14−6.86 (m, 4H, Ph-H and Ar−H), 4.54 and 3.61 (dd, $^3J_{HH}$ = 10.0 Hz, 2.8 Hz, 1H, C**H**), 4.30−4.10 (m, 1H, C**H**), 3.86 and 3.84 (s, 3H, O−C**H**$_3$), 3.04−2.89 (m, 1H, C**H**$_2$), 2.77−2.60 (m, 1H, C**H**$_2$), 1.55−1.37 (m, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 173.9 and 168.9, 157.7 and 157.6, 142.5 and 139.3, 137.6, 135.8, 131.5 and 131.4, 126.9 and 126.8, 125.6 and 125.2, 122.3 and 122.1, 120.7 and 120.7, 120.6, 118.5 and 118.4, 117.6 and 117.5, 111.8, 111.0, 106.1 and 105.9, 56.6 and 53.5, 55.7, 48.4 and 47.8, 25.5 and 25.1, 19.9 and 19.6; HRMS (ESI) calcd for $C_{21}H_{23}N_4O_2$ $[M + H]^+$ 363.1816, found 363.1823.

*Data for N′-(3,4-Dimethoxybenzylidene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**15**).* This compound was obtained as light yellow solid in 91% yield: mp = 203−206 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.47 and 11.27 (s, 1H, NH), 10.89 and 10.83 (s, 1H, O═C-NH), 8.25 and 7.99 (s, 1H, N═CH), 7.43−7.36 (m, 1H, Ar−H), 7.34−7.27 and 7.22−7.15 (m, 3H, Ar−H and Ph-H), 7.07−6.91 (m, 3H, Ar−H and Ph-H), 4.49 and 3.67−3.59 (dd, $^3J_{HH}$ = 11.2 Hz, 3.6 Hz, 1H, C**H**), 4.33−4.11 (m, 1H, C**H**), 3.82 and 3.75 (s, 3H, O−C**H**$_3$), 3.81 and 3.64 (s, 3H, O−C**H**$_3$), 3.03 and 2.94 (dd, $^2J_{HH}$ = 14.8 Hz, $^3J_{HH}$ = 2.8 Hz, 1H, C**H**$_2$), 2.76−

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2.65 (m, 1H, C**H**$_2$), 1.48 and 1.46 (d, $^3J_{HH}$ = 6.8 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 173.7 and 168.8, 150.7 and 150.5, 149.1 and 148.9, 147.4 and 143.7, 137.7, 135.9, 127.0, 126.9 and 126.9, 121.8 and 120.7, 120.6 and 120.4, 118.4, 117.5, 111.7 and 111.5, 111.0, 109.2 and 108.3, 106.1 and 106.0, 56.6 and 53.8, 55.6 and 55.5, 55.5 and 55.3, 48.4 and 48.0, 25.5 and 24.9, 19.9 and 19.6; HRMS (ESI) calcd for $C_{22}H_{25}N_4O_3$ [M + H]$^+$ 393.1921, found 393.1918.

*Data for N′-(Benzo[d][1,3]dioxol-5-ylmethylene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**16**).* This compound was obtained as yellow solid in 83% yield: mp = 199−203 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.41 and 11.26 (s, 1H, NH), 10.86 and 10.81 (s, 1H, O═C-NH), 8.24 and 7.96 (s, 1H, N═CH), 7.40 and 7.36 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.31 and 7.30 (d, $^3J_{HH}$ = 8.0 Hz, 1H, Ar−H), 7.28 and 7.17 (s, 1H, Ph-H), 7.15 and 7.09 (d, $^3J_{HH}$ = 8.0 Hz, 1H, Ph-H), 7.06−6.96 (m, 2H, Ar−H and Ph-H), 6.94 (t, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 6.10 (s, 1H, O−C**H**$_2$), 6.02 (d, $^2J_{HH}$ = 4.8 Hz, 1H, O−C**H**$_2$), 4.49 and 3.60 (dd, $^3J_{HH}$ = 10.8 Hz, 3.6 Hz, 1H, C**H**), 4.22 and 4.13 (m, 1H, C**H**), 2.99−2.87 (m, 1H, C**H**$_2$), 2.75−2.57 (m, 1H, C**H**$_2$), 1.45 (d, $^3J_{HH}$ = 6.4 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.2 and 168.9, 149.1 and 148.9, 148.0 and 147.9, 146.9 and 143.2, 137.8, 135.8, 128.8 and 128.6, 126.9, 123.3 and 122.8, 120.6 and 120.6, 118.4, 117.5, 111.0, 108.5, 106.2 and 106.0, 105.1 and 105.0, 101.6 and 101.5, 56.7 and 53.3, 48.4 and 47.7, 25.5 and 25.2, 19.9 and 19.8; HRMS (ESI) calcd for $C_{21}H_{21}N_4O_3$ [M + H]$^+$ 377.1608, found 377.1615.

*Data for N′-((2,3-Dihydrobenzo[b][1,4]dioxin-6-yl)methylene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**17**).* This compound was obtained as yellow solid in 81% yield: mp = 204−207 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.44 and 11.26 (s, 1H, NH), 10.89 and 10.82 (s, 1H, O═C-NH), 8.21 and 7.94 (s, 1H, N═CH), 7.40 and 7.36 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.32 and 7.30 (d, $^3J_{HH}$ = 8.0 Hz, 1H, Ar−H), 7.20 and 7.10 (s, 1H, Ph-H), 7.19 and 7.09 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ph-H), 7.07−7.01 (m, 1H, Ar−H), 6.99−6.83 (m, 2H, Ar−H and Ph-H), 4.51 and 3.61 (dd, $^3J_{HH}$ = 10.8 Hz, 3.6 Hz, 1H, C**H**), 4.35−4.09 (m, 5H, C**H** and O−C**H**$_2$-C**H**$_2$-O), 3.03−2.89 (m, 1H, C**H**$_2$), 2.76−2.60 (m, 1H, C**H**$_2$), 1.53−1.43 (m, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 173.8 and 168.8, 146.8, 145.2 and 145.1, 143.6 and 143.6, 143.2, 137.7 and 137.1, 135.9 and 135.8, 127.7 and 127.5, 126.9 and 126.8, 120.7 and 120.7, 120.6 and 120.3, 118.5 and 118.4, 117.6 and 117.5, 115.3 and 114.9, 111.1 and 111.0, 106.2 and 105.9, 64.3 and 64.3, 64.1 and 64.0, 56.6 and 53.6, 48.5 and 47.9, 25.5 and 25.0, 19.9 and 19.6; HRMS (ESI) calcd for $C_{22}H_{23}N_4O_3$ [M + H]$^+$ 391.1765, found 391.1763.

*Data for N′-((6-Hydroxynaphthalen-2-yl)methylene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**18**).* This compound needed recrystallization from methanol to removed the remaining aldehyde. This compound was obtained as yellow solid in 70% yield: mp = 275−278 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.46 and 11.35 (s, 1H, NH), 10.85 and 10.81 (s, 1H, O═C-NH), 9.98 (s, 1H, OH), 8.42 and 8.16 (s, 1H, N═CH), 7.99 and 7.91 (s, 1H, Naphthalene-H), 7.85 (d, $^3J_{HH}$ = 9.2 Hz, 1H, Naphthalene-H), 7.80 and 7.61 (d, $^3J_{HH}$ = 8.8 Hz, 1H, Naphthalene-H), 7.74 and 7.70 (d, $^3J_{HH}$ = 8.8 Hz, 1H, Naphthalene-H), 7.42 and 7.37 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.16 and 7.08 (s, 1H, Naphthalene-H), 7.15−7.09 (m, 1H, Naphthalene-H), 7.04 (t, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.00−6.90 (m, 1H, Ar−H), 4.52 and 3.63 (d, $^3J_{HH}$ = 8.4 Hz, 1H, C**H**), 4.25−4.10 (m, 1H, C**H**), 3.02−2.90 (m, 1H, C**H**$_2$), 2.73 and 2.65 (t, $^2J_{HH}$ = 12.8 Hz, 1H, C**H**$_2$), 1.47 (d, $^3J_{HH}$ = 6.0 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.5 and 169.0, 156.5 and 156.5, 147.4 and 144.0, 137.9, 135.8 and 135.8, 135.6 and 135.4, 130.1 and 130.0, 129.0, 128.7 and 128.7, 127.3 and 127.3, 127.0 and 126.9, 126.9 and 126.8, 123.0 and 122.3, 120.6, 119.2 and 119.2, 118.4, 117.5, 111.0, 109.1, 106.1 and 106.0, 56.7 and 53.3, 48.4 and 47.7, 25.6 and 25.4, 20.0 and 20.0; HRMS (ESI) calcd for $C_{24}H_{23}N_4O_2$ [M + H]$^+$ 399.1816, found 399.1822.

*Data for N′-(Pyridin-4-ylmethylene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**19**).* This compound was obtained as yellow solid in 79% yield: mp = 235−239 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.84 and 11.69 (s, 1H, NH), 10.90 and 10.84 (s, 1H, O═C-NH), 8.65 and 8.57 (d, $^3J_{HH}$ = 5.6 Hz, 2H, Py-H), 8.35 and 8.05 (s, 1H, N═CH), 7.65 and 7.57 (d, $^3J_{HH}$ = 5.6 Hz, 2H, Py-H), 7.41 and 7.37 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ph-H), 7.34−7.29 (m, 1H, Ar−H), 7.04 (t, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 6.95 (t, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 4.58 and 3.67 (dd, $^3J_{HH}$ = 10.8 Hz, 3.6 Hz, 1H, C**H**), 4.26 and 4.16 (q, $^3J_{HH}$ = 6.4 Hz, 1H, C**H**), 3.05−2.91 (m, 1H, C**H**$_2$), 2.80−2.62 (m, 1H, C**H**$_2$), 1.48 (d, $^3J_{HH}$ = 6.4 Hz, 3H, CH$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.5 and 169.4, 150.3, 144.7, 141.6, 141.3 and 141.2, 137.7 and 135.8, 126.9 and 126.8, 121.0, 120.7 and 120.6, 118.5 and 118.4, 117.6 and 117.5, 111.0, 106.0 and 105.9, 56.7 and 53.3, 48.5 and 47.9, 25.4 and 25.2, 19.8 and 19.6; HRMS (ESI) calcd for $C_{19}H_{20}N_5O$ [M + H]$^+$ 334.1662, found 334.1663.

*Data for N′-(pyridin-3-ylmethylene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**20**).* This compound was obtained as yellow solid in 72% yield: mp = 205−209 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.74 and 11.58 (s, 1H, NH), 10.91 and 10.84 (s, 1H, O═C-NH), 8.84 and 8.79 (s, 1H, Py-H), 8.62 and 8.55 (d, $^3J_{HH}$ = 4.0 Hz, 1H, Py-H), 8.41 and 8.12 (s, 1H, N═CH), 8.12 and 8.03 (d, $^3J_{HH}$ = 4.0 Hz, 1H, Py-H), 7.49 and 7.44−7.35 (m, 2H, Py-H and Ar−H), 7.33 and 7.31 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ph-H), 7.04 (t, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 6.96 (t, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 4.59 and 3.66 (dd, $^3J_{HH}$ = 10.8 Hz, 3.6 Hz, 1H, C**H**), 4.32 and 4.12 (m, 1H, C**H**), 3.05−2.92 (m, 1H, C**H**$_2$), 2.78−2.63 (m, 1H, C**H**$_2$), 1.53−1.39 (m, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 169.1, 150.7 and 150.5, 148.7 and 148.5, 144.4 and 140.9, 137.6 and 135.8, 133.4 and 133.2, 130.3, 130.0, 126.9 and 126.8, 124.0 and 124.0, 120.7 and 120.6, 118.5 and 118.4, 117.6 and 117.5, 111.0, 106.0 and 105.9, 56.7 and 53.4, 48.5 and 47.9, 25.4 and 25.1, 19.8 and 19.5; HRMS (ESI) calcd for $C_{19}H_{20}N_5O$ [M + H]$^+$ 334.1662, found 334.1664.

*Data for N′-(Pyridin-2-ylmethylene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**21**).* This compound was obtained as yellow solid in 85% yield: mp = 245−249 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.69 and 11.65 (s, 1H, NH), 10.85 and 10.82 (s, 1H, O═C-NH), 8.62 and 8.57 (d, $^3J_{HH}$ = 4.8 Hz, 1H, Py-H), 8.36 and 8.10 (s, 1H, N═CH), 7.95 and 7.80 (d, $^3J_{HH}$ = 8.0 Hz, 1H, Py-H), 7.88 and 7.75 (td, $^3J_{HH}$ = 7.6 Hz, $^4J_{HH}$ = 1.2 Hz, 1H, Py-H), 7.44−7.33 (m, 2H, Py-H and Ar−H), 7.31 and 7.30 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.03 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 6.95 and 6.92 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 4.50 and 3.63 (dd, $^3J_{HH}$ = 10.8 Hz, 4.0 Hz, 1H, C**H**), 4.16 (q, $^3J_{HH}$ = 8.0 Hz, 1H, C**H**), 2.94 (ddd, $^2J_{HH}$ = 14.8 Hz, $^3J_{HH}$ = 4.4 Hz, $^4J_{HH}$ = 1.6 Hz, 1H, C**H**$_2$), 2.72 and 2.63 (ddd, $^2J_{HH}$ = 14.8 Hz, $^3J_{HH}$ = 10.8 Hz, $^4J_{HH}$ = 2.0 Hz, 1H, C**H**$_2$), 1.46 and 1.45 (d, $^3J_{HH}$ = 6.8 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.8 and 169.5, 153.4 and 153.0, 149.5, 147.2 and 143.9, 137.8 and 137.8, 136.9 and 136.9, 135.8 and 135.8, 126.9 and 126.9, 124.4 and 124.2, 120.6, 119.9 and 119.6, 118.4, 117.5 and 117.5, 111.0, 106.1 and 106.0, 56.8 and 53.2, 48.5 and 47.7, 25.5, 19.9; HRMS (ESI) calcd for $C_{19}H_{20}N_5O$ [M + H]$^+$ 334.1662, found 334.1666.

*Data for N′-(Furan-2-ylmethylene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**22**).* The crude product was chromatographed on silica gel (DCM/MeOH = 40:1 → 30:1 → 20:1) to give two compounds. Compound **22a** was obtained as yellow solid in 55% yield: mp = 144−148 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 10.33 and 10.20 (s, 1H, NH), 8.26 and 8.11 (s, 1H, O═C-NH), 8.04 and 7.72 (s, 1H, N═CH), 7.51−7.38 (m, 2H, Ar−H and furan-H), 7.34 and 7.31 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.14 (t, $^3J_{HH}$ = 6.8 Hz, 1H, Ar−H), 7.08 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 6.77 and 6.62 (s, 1H, furan-H), 6.45 and 6.42 (s, 1H, furan-H), 4.65 and 3.69 (d, $^3J_{HH}$ = 7.6 Hz, 1H, C**H**), 4.32−4.11 (m, 1H, C**H**), 3.30 and 3.19 (d, $^2J_{HH}$ = 13.6 Hz, 1H, C**H**$_2$), 2.79 (t, $^2J_{HH}$ = 13.2 Hz, 1H, C**H**$_2$), 1.97 (br, 1H, NH), 1.52 and 1.46 (d, $^3J_{HH}$ = 6.4 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.4 and 169.1, 149.5 and 149.1, 145.1 and 144.9, 137.8 and 137.7, 136.9, 135.8, 126.9 and 126.9, 120.6, 118.4, 117.5, 113.6 and 113.3, 112.2 and 112.1, 111.0, 106.2 and 106.1, 56.7 and 53.2, 48.5 and 47.7, 25.5 and 25.4, 19.9; HRMS (ESI) calcd for $C_{18}H_{19}N_4O_2$ [M + H]$^+$ 323.1503, found 323.1505.

Compound **22b** was obtained as yellow solid in 3% yield: mp = 154−157 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 12.00 and 10.44 (s, 1H, NH), 8.00 and 7.94 (s, 1H, O═C-NH), 7.64 and 7.60 (s, 1H, N═CH), 7.52 and 7.46 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.40−7.30 (m, 2H, Ar−H and furan-H), 7.16 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 7.11 (t, $^3J_{HH}$

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= 7.2 Hz, 1H, Ar−H), 6.78 and 6.70 (s, 1H, furan-H), 6.60 and 6.55 (s, 1H, furan-H), 4.67 and 3.82 (d, $^3J_{HH}$ = 8.8 Hz, 1H, C**H**), 4.36−4.25 (m, 1H, C**H**), 3.30 and 3.20 (d, $^2J_{HH}$ = 15.2 Hz, 1H, C**H**$_2$), 2.92−2.75 (m, 1H, C**H**$_2$), 1.72 (br, 1H, NH), 1.61−1.49 (m, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 169.4, 147.8, 145.6, 138.2, 135.8, 130.8, 127.0, 120.6, 118.4, 117.6, 116.2, 112.5, 111.0, 106.8, 56.9, 48.8, 24.4, 19.9; HRMS (ESI) calcd for $C_{18}H_{19}N_4O_2$ [M + H]$^+$ 323.1503, found 323.1509.

*Data for N′-((1H-Pyrrol-2-yl)methylene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**23**).* This compound was obtained as red solid in 75% yield: mp = 207−209 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.52 and 11.31 (s, 1H, NH), 11.16 and 11.05 (s, 1H, Pyrrole-NH), 10.83 and 10.80 (s, 1H, O═C-NH), 8.15 and 7.88 (s, 1H, N═CH), 7.45−7.35 (m, 1H, Ar−H), 7.30 (d, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 7.03 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 6.95 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 6.90 and 6.83 (s, 1H, Pyrrole-H), 6.45 and 6.39 (s, 1H, Pyrrole-H), 6.13 and 6.08 (s, 1H, Pyrrole-H), 4.60 and 3.58 (d, $^3J_{HH}$ = 8.8 Hz, 1H, C**H**), 4.24−4.08 (m, 1H, C**H**), 2.98−2.88 (m, 1H, C**H**$_2$), 2.70−2.59 (m, 1H, C**H**$_2$), 1.50−1.45 (m, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 173.9 and 168.4, 140.2, 137.8, 136.4, 135.8, 127.0 and 126.9, 121.9, 120.6 and 120.6, 118.4, 117.7 and 117.5, 113.3 and 112.8, 111.0, 109.2 and 109.1, 106.4 and 106.2, 56.6 and 52.6, 48.4 and 47.7, 25.6 and 25.1, 19.9 and 19.7; HRMS (ESI) calcd for $C_{18}H_{20}N_5O$ [M + H]$^+$ 322.1662, found 322.1668.

*Data for N′-(Thiophen-2-ylmethylene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**24**).* This compound was obtained as yellow solid in 76% yield: mp = 139−141 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.55 and 11.39 (s, 1H, NH), 10.90 and 10.83 (s, 1H, O═C-NH), 8.56 and 8.24 (s, 1H, N═CH), 7.67 and 7.54 (d, $^3J_{HH}$ = 4.8 Hz, 1H, Thiophene-H), 7.47−7.35 (m, 2H, Thiophene-H and Ar−H), 7.35−7.28 (m, 1H, Ar−H), 7.14 and 7.09 (t, $^3J_{HH}$ = 4.4 Hz, 1H, Thiophene-H), 7.04 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 7.00−6.90 (m, 1H, Ar−H), 4.38 and 3.62 (dd, $^3J_{HH}$ = 8.8 Hz, 4.0 Hz, 1H, C**H**), 4.29−4.10 (m, 1H, C**H**), 3.04−2.89 (m, 1H, C**H**$_2$), 2.77−2.61 (m, 1H, C**H**$_2$), 1.54−1.38 (m, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 173.6 and 168.9, 142.3, 139.1 and 138.9, 138.6 and 137.6, 135.9 and 135.8, 130.8 and 130.4, 128.8 and 128.4, 128.0 and 127.9, 126.9 and 126.8, 120.7 and 120.6, 118.5 and 118.4, 117.5, 111.1 and 111.0, 106.1 and 105.9, 56.7 and 53.7, 48.5 and 47.9, 25.4 and 25.0, 19.8 and 19.6; HRMS (ESI) calcd for $C_{18}H_{19}N_4OS$ [M + H]$^+$ 339.1274, found 339.1278.

*Data for N′-((1H-imidazol-2-yl)methylene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**25**).* This compound was obtained as green solid in 81% yield: mp = 188−190 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 14.10, 13.33, 12.81, and 12.57 (s, 1H, Imidzole-NH), 12.93 and 11.47 (s, 1H, NH), 10.84 and 10.81 (s, 1H, O═C-NH), 8.45, 8.26, 7.95, and 7.44 (s, 1H, N═CH), 7.42−6.88 (m, 6H, Ar−H and Imidzole-H), 4.65, 4.50, 3.69, and 3.63 (m, 1H, C**H**), 4.37 and 4.17 (m, 1H, C**H**), 3.03 and 2.95 (d, $^2J_{HH}$ = 14.0 Hz, 1H, C**H**$_2$), 2.78−2.56 (m, 1H, C**H**$_2$), 1.52−1.42 (m, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.9, 174.7, 170.4 and 169.2, 142.6, 142.5, 141.1 and 141.0, 139.0, 138.2, 137.8 and 137.7, 135.8 and 135.2, 129.6 and 129.3, 127.0, 126.9, 126.9 and 126.8, 120.6 and 120.6, 118.4, 117.7 and 117.5, 111.0 and 111.0, 106.5, 106.4, 106.2 and 106.0, 57.2, 56.8, 53.1 and 52.5, 48.7, 48.5, 47.9 and 47.7, 25.6, 25.3, 25.3 and 25.1, 20.0, 20.0, 19.8 and 19.8; HRMS (ESI) calcd for $C_{17}H_{19}N_6O$ [M + H]$^+$ 323.1615, found 323.1620.

*Data for (N′-((E)-But-2-enylidene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**26**).* The crude produt was chromatographed on silica gel to give **26** as a yellow solid in 79% yield: mp = 145−148 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 10.10 and 9.63 (s, 1H, NH), 8.23 and 8.15 (s, 1H, O═C-NH), 7.72 and 7.37 (d, $^3J_{HH}$ = 8.4 Hz, 1H, N═CH), 7.43 (d, $^3J_{HH}$ = 6.8 Hz, 1H, Ar−H), 7.35−7.27 (m, 1H, Ar−H), 7.20−7.00 (m, 2H, Ar−H), 6.42−5.94 (m, 2H, C**H**C**H**), 4.57−4.48 and 3.70−3.58 (m, 1H, C**H**), 4.31−4.06 (m, 1H, C**H**), 3.27 and 3.12 (d, $^2J_{HH}$ = 14.4 Hz, 1H, C**H**$_2$), 2.85−2.66 (m, 1H, C**H**$_2$), 2.09 (br, 1H, NH), 1.92−1.75 (m, 3H, C**H**$_3$), 1.50 and 1.43 (d, $^3J_{HH}$ = 5.6 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.5 and 169.2, 150.3 and 147.2, 139.7 and 138.9, 136.8, 136.0 and 135.9, 128.5 and 128.1, 127.3 and 127.2, 121.8 and 121.7, 119.6 and 119.5, 118.3 and 118.1, 111.1, 107.9 and 107.7, 57.2 and 54.1, 49.4 and 48.0, 25.7 and 25.2, 20.2 and 20.2, 18.7 and 18.6; HRMS (ESI) calcd for $C_{18}H_{20}N_5O$ [M + H]$^+$ 297.1710, found 297.1714.

Synthesis of *N′*-(Butylidene)-(3*S*)-1-methyl-2,3,4,9-tetrahydro-1*H*-pyrido[3,4-*b*]indole-3-carbohydrazide (27).[19] To a solution of compound **1** (0.50 g, 2.05 mmol) in THF (35 mL) was added dropwise *n*-butanal (0.30 g, 4.10 mmol) at room temperature, After 3 h, TLC analysis showed the complete consumption of compound **1**. Then the mixture was concentrated in vacuo, and the crude product was purified by column chromatography on silica gel eluted with DCM/MeOH (25:1 → 10:1) to give **27** as a yellow solid in 71% yield: mp = 113−117 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.04 and 10.97 (s, 1H, NH), 10.82 and 10.80 (s, 1H, O═C-NH), 7.59 and 7.42−7.33 (m, 2H, N═CH and Ar−H), 7.30 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ph-H), 7.03 (t, $^3J_{HH}$ = 6.8 Hz, 1H, Ar−H), 6.95 (t, $^3J_{HH}$ = 6.8 Hz, 1H, Ar−H), 4.28 and 3.51 (d, $^3J_{HH}$ = 9.6 Hz, 1H, C**H**), 4.19−4.04 (m, 1H, C**H**), 2.89 (d, $^2J_{HH}$ = 14.0 Hz, 1H, C**H**$_2$), 2.67 and 2.57 (t, $^2J_{HH}$ = 12.8 Hz, 1H, C**H**$_2$), 2.25−2.10 (m, 2H, C**H**$_2$), 1.55−1.40 (m, 5H, C**H**$_3$ and C**H**$_2$), 0.92 and 0.86 (t, $^3J_{HH}$ = 6.8 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 173.9 and 168.7, 151.3 and 147.8, 137.8 and 137.7, 135.9, 126.9, 120.6, 118.4, 117.5 and 117.5, 111.0, 106.2 and 106.1, 56.5 and 53.3, 48.4 and 47.7, 33.9 and 33.7, 25.5 and 25.2, 20.0, 19.5 and 19.2, 13.7 and 13.5; HRMS (ESI) calcd for $C_{17}H_{23}N_4O$ [M + H]$^+$ 299.1866, found 299.1870.

Compounds **28** and **29** were synthesized using a similar procedure as compound **27**.

*Data for N′-(Octylidene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**28**).* The crude product was purified by column chromatography on silica gel eluted with DCM/MeOH (40:1 → 30:1 → 10:1) to give **28** as a yellow solid in 78% yield: mp = 68−71 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.94 and 9.02 (s, 1H, NH), 7.90 and 7.50 (s, 1H, N═CH), 7.50 and 7.46 (d, $^3J_{HH}$ = 8.0 Hz, 1H, Ar−H), 7.34 and 7.32 (d, $^3J_{HH}$ = 8.4 Hz, 1H, Ar−H), 7.17 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 7.11 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 4.53 and 3.70 (dd, $^3J_{HH}$ = 11.2 Hz, 4.4 Hz, 1H, C**H**), 4.33−4.17 (m, 1H, C**H**), 3.37 and 3.13 (dd, $^2J_{HH}$ = 14.4 Hz, $^3J_{HH}$ = 2.8 Hz, 1H, C**H**$_2$), 2.83−2.72 (m, 1H, C**H**$_2$), 2.38 and 2.22 (q, $^3J_{HH}$ = 7.2 Hz, 2H, C**H**$_2$), 1.96 (br, 1H, NH), 1.58−1.43 (m, 4H, C**H**$_2$ and C**H**$_3$), 1.42−1.18 (m, 9H, C**H**$_2$), 0.95−0.81 (m, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.2 and 168.6, 152.8 and 148.8, 136.7 and 136.6, 135.9 and 135.9, 127.3 and 127.2, 122.0 and 121.8, 119.7 and 119.5, 118.4 and 118.1, 111.0, 108.4 and 107.9, 57.3 and 54.1, 49.5 and 47.9, 32.4 and 32.1, 31.7 and 31.7, 29.3, 29.1 and 29.0, 26.7 and 26.3, 25.6 and 25.1, 22.6 and 22.6, 20.2, 14.1 and 14.1; HRMS (ESI) calcd for $C_{21}H_{31}N_4O$ [M + H]$^+$ 355.2493, found 355.2492.

*Data for N′-(Cyclohexylmethylene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**29**).* The crude product was purified by column chromatography on silica gel eluted with DCM/MeOH (100:1 → 40:1 → 20:1) to give **29** as a yellow solid in 92% yield: mp = 123−126 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.88 and 9.21 (s, 1H, NH), 7.98 and 7.96 (s, 1H, N═CH), 7.49 and 7.47 (d, $^3J_{HH}$ = 8.0 Hz, 1H, Ar−H), 7.38−7.29 and 7.05 (m, 2H, Ar−H and O═C-NH), 7.16 and 7.16 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 7.10 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 4.52 and 3.66 (dd, $^3J_{HH}$ = 10.8 Hz, 4.4 Hz, 1H, C**H**), 4.28 and 4.18 (q, $^3J_{HH}$ = 6.8 Hz, 1H, C**H**), 3.35 and 3.12 (dd, $^2J_{HH}$ = 15.6 Hz, $^3J_{HH}$ = 2.8 Hz, 1H, C**H**$_2$), 2.85−2.72 (m, 1H, C**H**$_2$), 2.47−2.35 and 2.23−2.13 (m, 1H, C**H**), 1.94 (br, 1H, NH), 1.87−1.58 (m, 4H, C**H**$_2$), 1.53 and 1.48 (d, $^3J_{HH}$ = 10.8 Hz, 3H, C**H**$_3$), 1.38−1.11 (m, 6H, C**H**$_2$), 0.95−0.81 (m, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, $CDCl_3$) δ 175.3 and 168.6, 156.3 and 152.2, 136.9 and 136.6, 135.9 and 135.8, 127.4 and 127.3, 122.0 and 121.7, 119.8 and 119.5, 118.4 and 118.1, 110.9, 108.5 and 108.0, 57.4 and 54.2, 49.5 and 47.9, 40.8 and 40.4, 30.2, 30.0 and 30.0, 25.9 and 25.8, 25.6 and 25.4, 25.3 and 25.1, 20.3 and 20.3; HRMS (ESI) calcd for $C_{20}H_{27}N_4O$ [M + H]$^+$ 339.2180, found 339.2179.

Synthesis of *N′*-(2,2-Dimethylpropylidene)-(3*S*)-1-methyl-2,3,4,9-tetrahydro-1*H*-pyrido[3,4-*b*]indole-3-carbohydrazide (30).[19] To a solution of compound **1** (0.38 g, 1.56 mmol) in THF (35 mL) was added dropwise trimethylacetaldehyde (0.27 g, 3.11 mmol)

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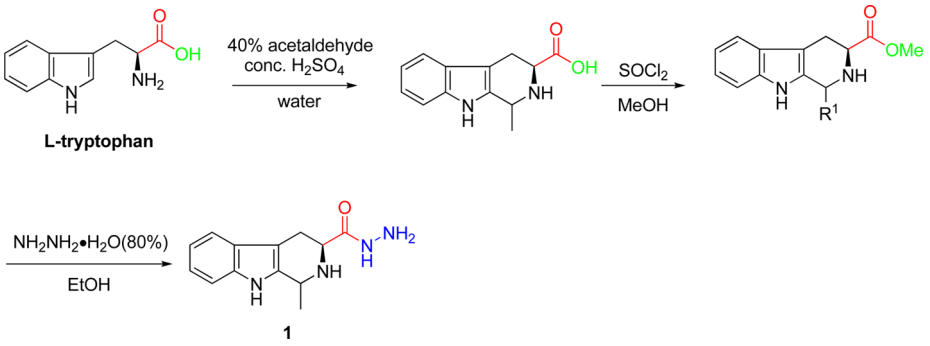
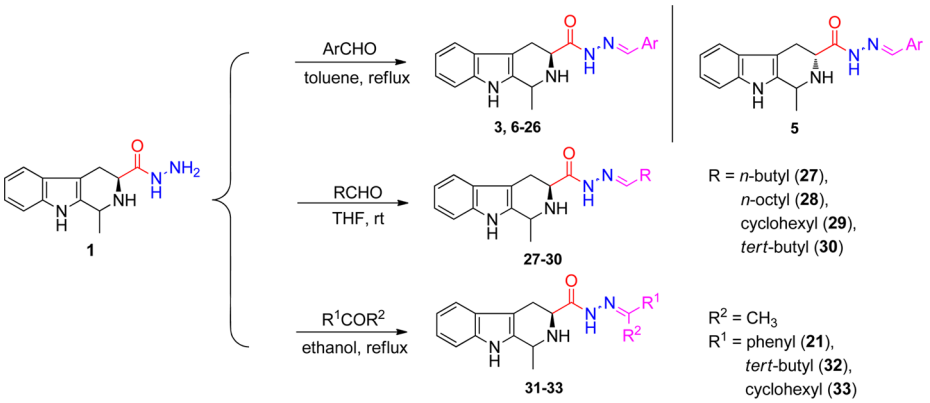
Scheme 1. Synthesis of Compound 1



Scheme 2. Synthesis of Compounds 3 and 5−33



Ar = phenyl (**3**, **5**), 4-*tert*-butylphenyl (**6**), 4-(dimethylamino)phenyl (7), 4-nitrophenzl (**8**), 4-chlorophenyl (**9**), 2,4-dichlorophenyl (**10**), 3,4-dichlorophenyl (**11**), 4-methoxyphenyl (**12**), 3-methoxyphenyl (**13**), 2-methoxyphenyl (**14**), 3,4-dimethoxyphenyl (**15**), benzo[*d*][1,3]dioxol-5-yl (**16**), 2,3-dihydrobenzo[*b*][1,4]dioxin-6-yl (**17**), 6-hydroxynaphthalen-2-yl (**18**), pyridin-4-yl (**19**), pyridin-3-yl (**20**), pyridin-2-yl (**21**), furan-2-yl (**22**), 1*H*-pyrrol-2-yl (**23**), thiophen-2-yl (**24**), 1*H*-imidazol-2-yl (**25**), (*E*)-but-2-enyl (**26**)

at room temperature, After 6 h, TLC analysis showed the complete consumption of compound **1**. Then the mixture was concentrated in vacuo and dried without further operation to give **30** as a yellow solid in 95% yield: mp = 140−141 °C; ^{1}H NMR (100 MHz, $CDCl_3$) δ 9.93 and 9.39 (s, 1H, NH), 8.13 and 8.09 (s, 1H, N=CH), 7.53−7.02 (m, 4H, Ar−H), 4.51 and 3.67 (d, $^3J_{HH}$ = 8.4 Hz, 1H, C**H**), 4.37−4.16 (m, 1H, C**H**), 3.33 and 3.13 (d, $^2J_{HH}$ = 14.4 Hz, 1H, C**H**$_2$), 2.79 (t, $^2J_{HH}$ = 13.2 Hz, 1H, C**H**$_2$), 2.09 (br, 1H, NH), 1.58−1.41 (m, 3H, C**H**$_3$), 1.15 and 1.06 (s, 9H, C**H**$_3$); ^{13}C NMR (100 MHz, $CDCl_3$) δ 176.0 and 169.3, 160.9 and 156.5, 137.3, 136.7 and 136.6, 128.1 and 127.9, 122.6 and 122.4, 120.4 and 120.2, 119.1 and 118.8, 111.7, 109.0 and 108.5, 58.1 and 55.1, 50.2 and 48.7, 35.8 and 35.6, 31.0 and 27.9, 28.1 and 28.0, 26.1 and 25.8, 20.9; HRMS (ESI) calcd for $C_{18}H_{25}N_4O$ $[M + H]^+$ 313.2023, found 313.2028.

Synthesis of *N′*-(1-Phenylethylidene)-(3*S*)-1-methyl-2,3,4,9-tetrahydro-1*H*-pyrido[3,4-*b*]indole-3-carbohydrazide (31).[20] To a solution of compound **1** (0.50 g, 2.05 mmol) in EtOH (35 mL) was added acetophenone (0.27 g, 2.25 mmol), and then the reaction mixture was heated to reflux. After 6 h, the reaction was complete detected by TLC. The mixture was concentrated in vacuo, and the crude product was purified by column chromatography on silica gel eluted with DCM/MeOH (30:1 → 10:1) to give **31** as a light yellow solid in 56% yield: mp = 221−224 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.84 and 10.81 (s, 1H, N−H), 10.69 and 10.48 (s, 1H, O=C-NH),7.89−7.77 and 7.76−7.67 (m, 2H, Ph-H), 7.48−7.27 (m, 5H, Ph-H and Ar−H), 7.08−6.88 (m, 2H, Ar−H), 4.53 and 3.78 (dd, $^3J_{HH}$ = 10.8 Hz, 3.6 Hz, 1H, C**H**), 4.23−4.11 (m, 1H, C**H**), 3.03−2.93 (m, 1H, C**H**$_2$), 2.76−2.59 (m, 1H, C**H**$_2$), 3.31 and 3.30 (s, 3H, C**H**$_3$), 1.46 (d, $^3J_{HH}$ = 6.4 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 175.3 and 169.2, 151.9 and 148.0, 138.1, 137.9 and 137.6, 135.8 and 135.8, 129.3 and 129.1, 128.4 and 128.4, 126.9, 126.4 and 125.9, 120.6, 118.4, 117.5 and 117.5, 111.0, 106.4 and 106.1, 56.5 and 53.7, 48.4 and 47.7, 25.5 and 25.3, 19.9, 13.7 and 13.4; HRMS (ESI) calcd for $C_{21}H_{23}N_4O$ $[M + H]^+$ 347.1867, found 347.1872.

Compounds **32** and **33** were synthesized using a similar procedure as compound **31**.

*Data for N′-(3,3-Dimethylbutan-2-ylidene)-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**32**).* The crude product was purified by column chromatography on silica gel eluted with DCM/MeOH (30:1 → 10:1) to give **32** as a yellow solid in 63% yield: mp = 103−107 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.80 and 10.79 (s, 1H, N−H), 10.18 and 10.02 (s, 1H, O=C-NH),7.40 and 7.35 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.29 (d, $^3J_{HH}$ = 8.0 Hz, 1H, Ar−H), 7.03 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 6.95 and 6.93 (t, $^3J_{HH}$ = 6.8 Hz, 1H, Ar−H), 4.26 and 3.65 (dd, $^3J_{HH}$ = 10.8 Hz, 4.0 Hz, 1H, C**H**), 4.16−4.07 (m, 1H, C**H**), 2.94 (dd, $^2J_{HH}$ = 14.8 Hz, $^3J_{HH}$ = 2.8 Hz, 1H, C**H**$_2$), 2.70−2.53 (m, 1H, C**H**$_2$), 1.87 and 1.85 (s, 3H, C**H**$_3$), 1.43 (d, $^3J_{HH}$ = 6.4 Hz, 3H, C**H**$_3$), 1.12 and 1.04 (s, 9H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 174.7 and 168.6, 163.2 and 158.5, 137.9 and 137.5, 135.8, 126.9, 120.6 and 120.5, 118.4, 117.5 and 117.4, 111.0, 106.5 and 106.2, 56.6 and 54.4, 48.4 and 47.8, 38.5 and 38.3, 27.6 and 27.5, 26.0, 25.3 and 24.8, 20.0 and 19.9, 12.0 and 11.8;

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HRMS (ESI) calcd for $C_{19}H_{27}N_4O$ $[M + H]^+$ 327.2180, found 327.2186.

*Data for N′-Cyclohexylidene-(3S)-1-methyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole-3-carbohydrazide (**33**).* The crude product was purified by column chromatography on silica gel eluted with DCM/MeOH (30:1 → 10:1) to give **33** as a yellow solid in 60% yield: mp = 131−135 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.81 and 10.78 (s, 1H, NH), 10.39 and 10.24 (s, 1H, O═C-NH),7.39 and 7.36 (d, $^3J_{HH}$ = 8.0 Hz, 1H, Ar−H), 7.29 (d, $^3J_{HH}$ = 7.6 Hz, 1H, Ar−H), 7.02 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 6.94 (t, $^3J_{HH}$ = 7.2 Hz, 1H, Ar−H), 4.36 and 3.62 (dd, $^3J_{HH}$ = 10.0 Hz, 3.6 Hz, 1H, C**H**), 4.18−4.04 (m, 1H, C**H**), 2.94−2.84 (m, 1H, C**H**$_2$), 2.69−2.58 (m, 1H, C**H**$_2$), 2.40−2.12 (m, 4H, C**H**$_2$), 1.71−1.51 (m, 6H, C**H**$_2$), 1.42 (d, $^3J_{HH}$ = 6.0 Hz, 3H, C**H**$_3$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 168.7, 161.5, 137.8, 135.8, 126.9 and 126.8, 120.6 and 120.5, 118.4, 117.5, 111.0, 106.3, 56.3 and 53.3, 48.3 and 47.7, 35.2 and 35.0, 27.0 and 26.8, 25.7 and 25.5, 25.1, 19.9 and 19.8; HRMS (ESI) calcd for $C_{19}H_{25}N_4O$ $[M + H]^+$ 325.2023, found 325.2023.

Biological Assay. The anti-TMV and fungicidal and insecticidal activities of the synthesized compounds were tested using our previously reported methods.[21,22]

Field Trials. The experiments were carried out between June and July in 2014. The trial against TMV was performed in the trial area of Institute of Environment and Resources, Yunnan Provincial Academy of Agricultural Sciences, Kunming, Yunnan Province, China. The experimental design was a randomized complete block with three treatments. The untreated plots served as a blank control. The formulation of compounds **9** (1% EC) and **29** (1% ME) was prepared in our lab. They were diluted to 10, 50, or 100 g(ai)/ha (weight (gram) of active ingredient/hectare) before use. The formulation of moroxydine hydrochloride−cupric acetate (20% WP) and amino-oligosaccharins (5% aqueous solution) were obtained from Wansheng Biological Pesticide Co., Ltd. (Shandong, China) and Hainan zhengye zhong nong Co., Ltd. (Hainan, China) respectively. The incidence was surveyed 10 days after the third time spraying. The severity of the virus disease was divided into 0−4 grade. The disease index and control effect can be calculated by using the following formulas:

disease index

$$I\% = [\sum (N_n n)/(N4)] \times 100$$

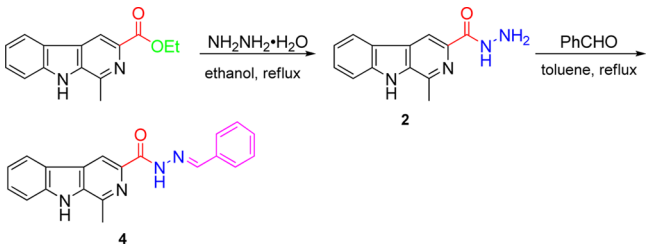
$$\text{control effect } (\%) = 1 - (I'_{bc}I)/(I_{bc}I') \times 100$$

where N_n is the number of diseased plants of each relative level values, n is the relative level values (n = 0−4), N is the total number of diseased plants, I'_{bc} is the disease index of the blank control area before spraying, I_{bc} is the disease index of the blank control area 10 days after the third time spraying, I' is the disease index of the spraying area before spraying; and I is the disease index of the spraying area 10 days after the third time spraying.

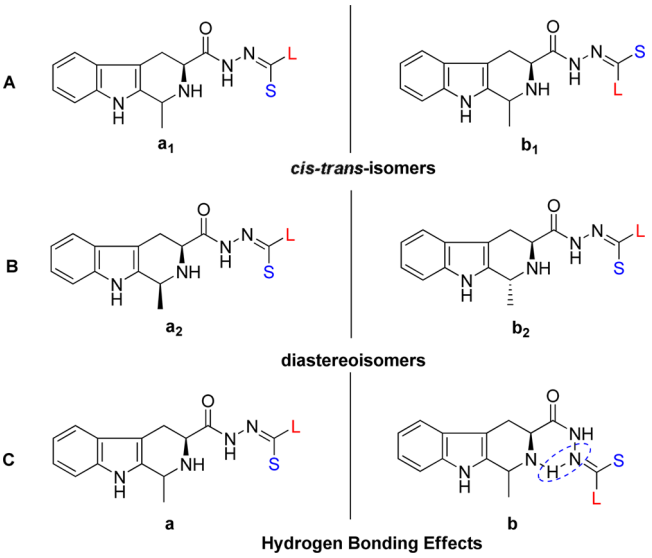
RESULTS AND DISCUSSION

Synthesis. The synthesis of the lead compound **1** is shown in Scheme 1. Using L-tryptophan as starting material, the tetrahydrocarboline skeleton of **1** was constructed through Pictet−Spengler cyclization, and then the given carboxylic acid reacted with thionyl chloride and methanol to form methyl ester, which subsequently reacted with hydrazine hydrate to give compound **1**.

Scheme 3. Synthesis of Compounds 2 and 4



Scheme 4. Prediction of the Configuration



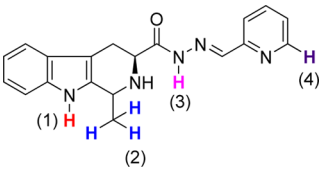


Figure 3. Compound **21**.

The compounds **3** and **6−33** containing the acylhydrazone fragment all can be synthesized from lead compound **1** by reaction with the corresponding aldehydes or ketones under different reaction conditions (Scheme 2). To be specific, when aromatic aldehydes were used as starting materials, the corresponding reactions were carried out in toluene under reflux conditions. When the reaction was complete, the reaction system was cooled to room temperature, and the solid products precipitated because of their poor solubility in toluene. By this approach, compounds **3** and **6−26** could be easily obtained in 60%−91% yields. When aliphatic aldehydes were used as starting materials, the corresponding reactions were carried out in tetrahydrofuran at room temperature. When the reaction was complete, compounds **27−30** could be obtained by column chromatography in 71%−95% yields. When ketones were used, these reactions were carried out in ethanol under reflux conditions. When the reaction was complete, compounds **31−33** could be obtained by column chromatography in 56%−63% yields.

In addition, 1-methyl-9*H*-carbazole-3-carboxylic acid hydrazide **2** could be prepared from ethyl 1-methyl-9*H*-pyrido[3,4-*b*]indole-3-carboxylate, which was synthesized from tryptophan through a literature protocol.[17] Compound **2** reacted with benzaldehyde using a similar procedure as the preparation of compound **3** to give **4** in 91% yield (Scheme 3).

Configuration. At room temperature, the ^{1}H and ^{13}C NMR spectra of most of target compounds exhibited two sets of signals, which aroused our attention. Initially, this phenomenon was thought to be due to the presence of *cis−trans* isomers (prediction A, Scheme 4); however, in the ^{1}H NMR spectrum

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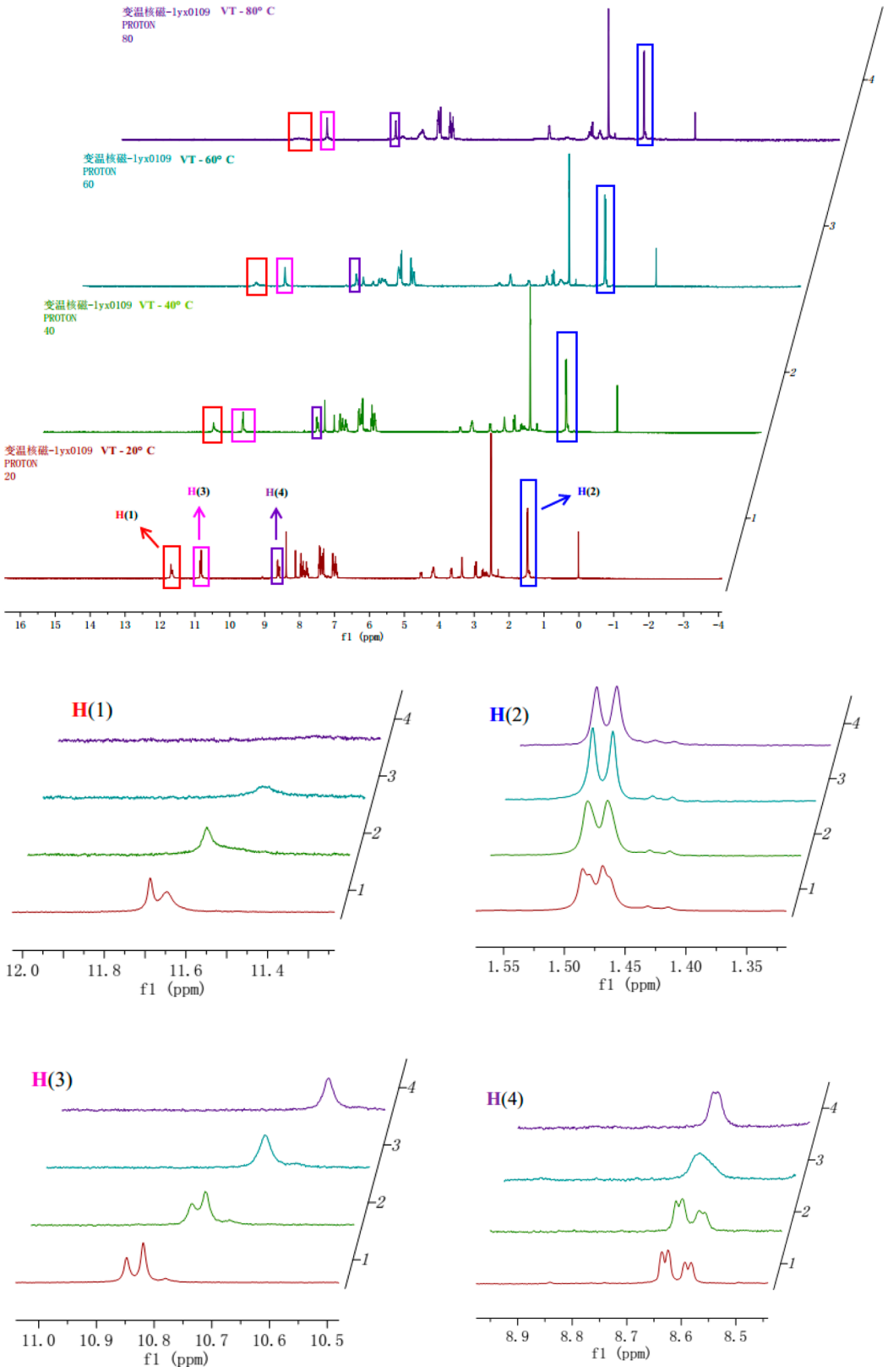

Figure 4. ^{1}H NMR of Compound **21** at Different Temperatures.

of **4** only one set of signals was observed, which suggested such phenomenon resulted from other reasons. Meanwhile, the presence of diastereoisomers (prediction B, Scheme 4) as a possible reason was excluded (the ^{1}H NMR spectra of precursor **1** as well as compound **21** (Figures 3, 4 and 5) clearly showed the presence of diastereoisomeric mixtures, and the ratio is 10:1). Eventually, hydrogen-bonding effects (prediction C, Scheme 4) was put forward, and variable-temperature (VT) ^{1}H NMR studies (Figures 4 and 5) of compound **21** were performed in order to confirm this

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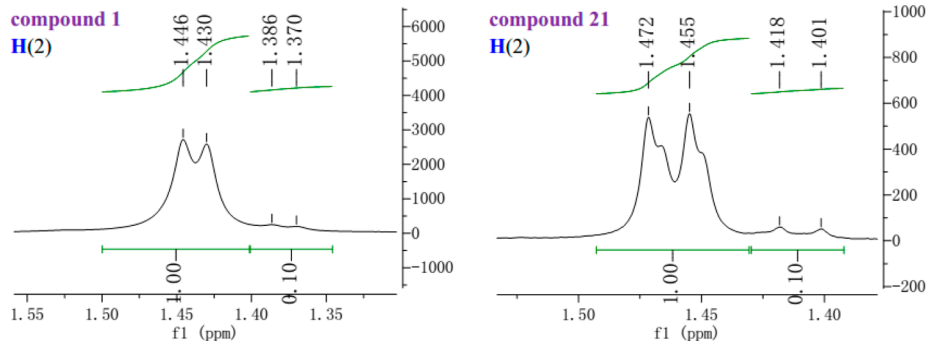

Figure 5. ^{1}H NMR (H2) of compounds **1** and **21**.

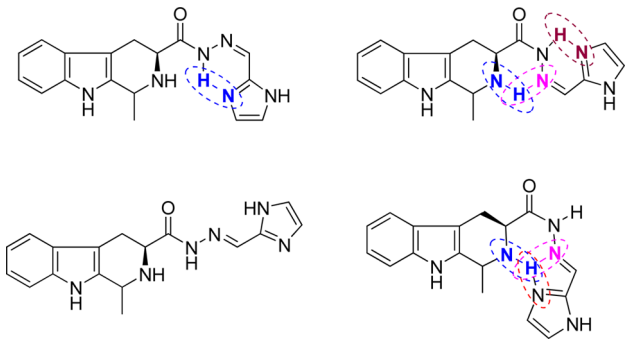

Figure 6. Hydrogen-bonding effects of compound **25**.

hypothesis. As we can see, the coalescence of signals was observed in the entire ^{1}H NMR spectrum with the rise of the temperature (20 → 40 → 60 → 80 °C). Therefore, the VT ^{1}H NMR studies indicated that the doubling of signals in the ^{1}H and ^{13}C NMR spectra was very likely due to hydrogen-bonding effects. In addition, it also could explain the appearance of four sets of signals in compound **25** (Figure 6).

Antiviral Activities. In our previous work, high antiviral lead compound **1** was found, on the basis of which, a series of derivatives containing acylhydrazone structure were synthesized to investigate the influence of the variation in the basic skeleton of **1**. The acylhydrazone compounds exhibited excellent antiviral activity. The antiviral results *in vitro* and *in vivo* (protection, inactivation, and curative) are listed in Tables 1 and 2. Overall, compared with ribavirin, most of the newly designed compounds exhibited desirable anti-TMV activity *in vitro*. Compounds **8**, **9**, **12**, **16**, **28**, **29**, and **30** showed much higher inhibition than that of ribavirin and their precursor **1** at 500 and 100 μg/mL. Especially, the activity of compound **16** was 36.9% at 100 μg/mL, which was very close to that of Ribavirin (40%) at 500 μg/mL.

When the C ring (Figure 3) was tetrahydropyridine, the in vitro activity of compound **1** (40.4%, 500 μg/mL) was a little lower than that of the β-carboline compound **2** (43.9%, 500 μg/mL); however, activity of compound **3** (55.8%, 500 μg/mL) was obviously higher than that of compound **4** (41.8%, 500 μg/mL). On the other hand, the activity of compound **1** (40.4%, 500 μg/mL) showed obviously lower inhibition than that of compound **3** (55.8%, 500 μg/mL), which proved that it is advantageous to improve the activity if the hydrazide structure at the 3-position of tetrahydrogen-β-carboline was changed to acylhydrazone. Hence, a representative set of compounds containing acylhydrazone structure were designed to search for better-activity compounds. Additionally, the

Table 1. *In Vitro* Antiviral Activities of Compounds 1−33 against TMV

compd	concn (μg/mL)	inhibition rate (%)	compd	concn (μg/mL)	inhibition rate (%)
1	500	40.4	18	500	40.9
	100	14.9		100	0
2	500	43.9	19	500	36.6
	100	23.8		100	0
3	500	55.8	20	500	33.3
	100	28.4		100	0
4	500	41.8	21	500	32.7
	100	10.5		100	0
5	500	49.6	22	500	48.8
	100	10.8		100	15.4
6	500	52.4	23	500	49.5
	100	17.6		100	8.8
7	500	56.8	24	500	58.5
	100	20.4		100	23
8	**500**	**62.8**	25	500	42.4
	100	19.6		100	0
9	**500**	**67.3**	26	500	35.2
	100	**30.8**		100	0
10	500	56.8	27	500	32.6
	100	20.1		100	7.8
11	500	50.7	**28**	**500**	**61.3**
	100	15.1		**100**	**23.2**
12	**500**	**62.5**	**29**	500	59.6
	100	**28.7**		**100**	**31.2**
13	500	53.2	**30**	**500**	**64.4**
	100	13.2		**100**	**27.5**
14	500	38.5	31	500	30.2
	100	0		100	9.2
15	500	37.2	32	500	58.1
	100	7.8		100	25.6
16	**500**	**72.6**	33	500	43.4
	100	**36.9**		100	8.6
17	500	46.5	Ribavirin	500	40
	100	19.8		100	12.9

structure−activity relationship (SAR) demonstrated that the chirality at the 3-position of tetrahydrogen-β-carboline affected slightly the antiviral activity; for instance, compound **3** (3*S*,

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Table 2. *In Vivo* Antiviral Activities of Compounds 1−33 against TMV

compd	concn (μg/mL)	inactivation effect (%)	curative effect (%)	protection effect (%)
1	500	40	42.3	39.6
	100	8.5	10.3	13.4
2	500	55.4	58.9	49.7
	100	21.4	16.9	19.2
3	500	56.8	53.1	58.4
	100	23.4	26.5	24.7
4	500	47.1	45	47.9
	100	13.8	7.9	19.4
5	500	51.7	47.6	48.2
	100	12.3	19.5	8.9
6	500	50.3	47	53.8
	100	13.3	18.2	21.7
7	**500**	**64.5**	**60.4**	**62.4**
	100	**25**	**23.1**	**18.9**
8	500	58.3	57.1	61.5
	100	27.4	20.8	21.4
9	**500**	**70.4**	**71.5**	**64.2**
	100	**35.9**	**29.2**	**34.1**
10	**500**	**66.7**	**60.4**	**57.2**
	100	**28.5**	**31.5**	**18.6**
11	500	55.2	48.9	48.1
	100	20.4	23.5	14.8
12	**500**	**69.1**	**66.4**	**70**
	100	**30.4**	**34.8**	**31.8**
13	500	59.2	54	54.7
	100	22.1	16.5	20.6
14	500	43.2	36.4	34.8
	100	0	10.9	0
15	500	47.1	40.5	42.3
	100	20.3	16.2	11
16	**500**	**74.5**	**69**	**68.1**
	100	**33.6**	**30.2**	**35.8**
17	500	43.6	48.1	46
	100	15.2	18.6	10.3
18	500	45.6	43.9	47.8
	100	16.9	8.2	0
19	500	43.4	39.6	37.5
	100	8.8	12.9	0
20	500	41	36.9	33.6
	100	11.2	5.7	0
21	500	38.9	36.8	31.4
	100	9.2	0	0
22	500	54.6	44.2	47.5
	100	18.2	9.3	11.5
23	**500**	**61.3**	**63.4**	**54.7**
	100	**20.2**	**27.8**	**30**
24	500	59.1	52.3	61.3
	100	29.6	25.1	31.8
25	500	49.6	45.3	48.1
	100	21.3	12.6	18.5
26	500	47	33.2	37.1
	100	8.5	0	6.7
27	500	47.1	41.3	40.5
	100	13.4	10.4	11.3
28	**500**	**73.4**	**70.9**	**59.6**
	100	**32.8**	**31.7**	**27.3**
29	**500**	**75.8**	**62.8**	**69.2**
	100	**36.3**	**30.4**	**25.7**
30	**500**	**63.7**	**68.2**	**66.8**
	100	**28.9**	**36.8**	**33.4**
31	500	39.5	34.8	37.2
	100	13.6	11.1	7.9
32	500	59.7	55.4	56.8
	100	21.3	16.7	18.9
33	500	33	41.9	46.2
	100	12.8	10.9	14.3
Ribavirin	500	37.6	39.4	37.9
	100	12.3	11.9	14

55.8%, 500 μg/mL) was slightly more active than compound **5** (3*R*, 49.6%, 500 μg/mL).

Furthermore, it was noteworthy that the substituents of the acylhydrazone moiety had a significant impact on anti-TMV activity.

When R^2 was hydrogen and R^1 was a benzene ring, it was universally advantageous to anti-TMV activity if substituents were introduced to the 4-position of the benzene ring such as **7** (56.8%, 500 μg/mL), **8** (62.8%, 500 μg/mL), **9** (67.3%, 500 μg/mL), and **12** (62.5%, 500 μg/mL). Monosubstitution or multisubstitution on the benzene ring affected anti-TMV activity to a certain extent, for instance, compared with compounds **10** (56.8%, 500 μg/mL) and **11** (50.7%, 500 μg/mL), the monosubstituted compound **9** exhibited significantly higher activity. In addition, the position of the substituent group also was favored to increase anti-TMV activity; the activity of compound **12** (62.5%, 500 μg/mL) was much better than that of **13** (53.2%, 500 μg/mL) and **14** (38.5%, 500 μg/mL), the order of their activities was para > meta > ortho. However, the activity was greatly reduced when the benzene ring was changed to a naphthalene ring, such as **18** (40.9%, 500 μg/mL).

Interestingly, compound **16** (72.6%, 500 μg/mL) exhibited excellent inhibitory effect; in contrast, compounds **15** (37.2%, 500 μg/mL) and **17** (46.5%, 500 μg/mL) were far less effective. We speculated it could be ascribed to the tension on the five-membered ring of **16**, which inhibited the conjugation of the lone-pair electrons of oxygen atoms with the benzene ring.

When R^2 was hydrogen and R^1 was a heterocyclic ring, the anti-TMV activities of these compounds (**19**−**25**) reduced obviously compared with those of compounds with substituted benzene (**3**, **5**−**18**).

When R^2 was hydrogen and R^1 was an aliphatic substituent, both the length of the aliphatic chain and the number of branched chains affected anti-TMV activity; the more branched and the longer was the aliphatic chain, the better was the

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Table 3. Fungicidal Activity of Compounds 1−4 and Compounds 6−33 against Fourteen Kinds of Phytopathogens

compd	fungicidal activity (%)/50 mg/kg													
	F.C[a]	*C.H*	*P.P*	*A.S*	*F.G*	*F.M*	*S.S*	*P.C*	*R.C*	*B.M*	*W.A*	*P.I*	*R.S*	*B.C*
1	13.6	18.2	39.4	9.5	38.5	29.4	30.8	51.5	54.1	24.0	24.0	26.1	4.2	27.8
2	34.6	35.3	55.6	25.0	24.1	0.0	11.8	7.9	43.1	12.0	11.1	16.0	13.2	13.6
3	36.4	**72.7**	36.4	**57.1**	**53.8**	**58.8**	26.9	**69.7**	**86.5**	**60.0**	**52.0**	30.4	48.6	**88.9**
4	15.4	17.6	55.6	20.0	44.8	7.1	35.3	18.4	43.1	12.0	14.8	16.0	13.2	36.4
6	**65.4**	**82.4**	**97.2**	**70.0**	**41.4**	**57.1**	**94.1**	**73.7**	**98.0**	**84.0**	**77.8**	**76.0**	**88.2**	**86.4**
7	38.5	76.5	**97.2**	65.0	31.0	35.7	41.2	**78.9**	**96.1**	**64.0**	48.1	72.0	63.2	**72.7**
8	38.5	58.8	**72.2**	45.0	58.6	42.9	**88.2**	**78.9**	**88.2**	**60.0**	48.1	52.0	59.2	**72.7**
9	**61.5**	**76.5**	**97.2**	**55.0**	**75.9**	**64.3**	**88.2**	**81.6**	**94.1**	**68.0**	**77.8**	**80.0**	**81.6**	**59.1**
10	27.3	45.5	**48.5**	42.9	53.8	47.1	46.2	**60.6**	**56.8**	**56.0**	60.0	39.1	45.8	**52.8**
11	**77.3**	**72.7**	**97.0**	**71.4**	**76.9**	**70.6**	**84.6**	**84.8**	**97.3**	**80.0**	**80.0**	**65.2**	**62.5**	**80.6**
12	23.1	64.7	**72.2**	30.0	65.5	64.3	**94.1**	**73.7**	**84.3**	**68.0**	55.6	52.0	59.2	**59.1**
13	34.6	58.8	**69.4**	65.0	41.4	57.1	**88.2**	**81.6**	**92.2**	**48.0**	40.7	40.0	68.4	**68.2**
14	26.9	23.5	**88.9**	45.0	72.4	21.4	**64.7**	**63.2**	**78.4**	**56.0**	40.7	44.0	32.9	**77.3**
15	22.7	45.5	**66.7**	52.4	30.8	47.1	**21.2**	**84.8**	**89.2**	**52.0**	72.0	30.4	44.4	**66.7**
16	31.8	63.6	**63.6**	52.4	53.8	58.8	**44.2**	**81.8**	**86.5**	**56.0**	48.0	39.1	50.0	**88.9**
17	22.7	45.5	**66.7**	52.4	30.8	47.1	**21.2**	**84.8**	**89.2**	**52.0**	72.0	30.4	44.4	**66.7**
18	19.2	47.1	**77.8**	30.0	10.3	28.6	**88.2**	**84.2**	**86.3**	**56.0**	48.1	44.0	28.9	**72.7**
19	23.1	35.3	**52.8**	15.0	27.6	21.4	**64.7**	28.9	**66.7**	28.0	29.6	24.0	23.7	36.4
20	38.5	41.2	**83.3**	15.0	62.1	50.0	35.3	26.3	**80.4**	32.0	40.7	52.0	39.5	54.5
21	23.1	23.5	**75.0**	20.0	20.7	21.4	5.9	18.4	**41.2**	16.0	25.9	12.0	21.1	36.4
22	11.5	52.9	**69.4**	35.0	6.9	28.6	**64.7**	39.5	**68.6**	20.0	33.3	36.0	32.9	81.8
23	23.1	23.5	**97.2**	15.0	41.4	14.3	**58.8**	42.1	**74.5**	28.0	29.6	24.0	32.9	22.7
24	26.9	47.1	**55.6**	45.0	62.1	28.6	**88.2**	68.4	**72.5**	44.0	44.4	36.0	46.1	72.7
25	34.6	47.1	**97.2**	15.0	27.6	28.6	35.3	18.4	**64.7**	12.0	25.9	24.0	15.8	36.4
26	26.9	29.4	**52.8**	30.0	20.7	14.3	41.2	31.6	**64.7**	36.0	22.2	32.0	6.6	40.9
27	19.2	17.6	**69.4**	45.0	6.9	0.0	64.7	5.3	43.1	12.0	14.8	20.0	6.6	9.1
28	9.1	36.4	24.2	19.0	61.5	35.3	46.2	18.2	29.7	36.0	32.0	17.4	22.2	44.4
29	4.5	0.0	33.3	19.0	30.8	41.2	21.2	9.1	29.7	36.0	24.0	17.4	5.6	66.7
30	4.5	18.2	36.4	19.0	46.2	29.4	15.4	12.1	48.6	20.0	24.0	17.4	8.3	36.1
31	0.0	36.4	39.4	33.3	61.5	29.4	11.5	36.4	51.4	36.0	52.0	26.1	15.3	69.4
32	4.5	36.4	36.4	14.3	53.8	23.5	3.8	18.2	48.6	28.0	32.0	30.4	8.3	41.7
33	0.0	18.2	9.1	19.0	46.2	29.4	7.7	12.1	48.6	32.0	32.0	26.1	15.3	52.8
carbendazim	<50	<50	<50	<50	100	<50	100	<50	100	100	100	100	100	<50
chlorothalonil	100	73.3	100	73.3	<50	100	<50	100	100	91.3	91.3	86.4	100	100

[a]*F.C., Fusarium oxysporum* sp. *cucumeris*; *C.H., Cercospora arachidicola* Hori; *P.P., Physalospora piricola*; *A.S., Alternaria solani*; *F.G., Fusarium graminearum*; *F.M., Fusarium moniliforme*; *S.S., Sclerotinia sclerotiorum*; *P.C., Phytophthora capsici*; *R.C., Rhizoctonia cerealis*; *B.M., Bipolaris maydis*; *W.A.*, watermelon anthracnose; *P.I., Phytophthora infestans*; *R.S., Rhizoctonia solani*; *B.C., Botrytis cinerea*.

Table 4. Insecticidal Activity of Compounds 1−4 and Compound 6−33 against Four Kinds of Insects

compd	larvicidal activity (%) at concn (mg/kg)				
	M. separata	*H. armigera*	*O. nubilalis*	*C. pipiens pallens*	
	600 mg/kg	600 mg/kg	600 mg/kg	10 mg/kg	5 mg/kg
1	20	20	30	40	—
2	45	40	40	100	40
3	**75**	**55**	**60**	**100**	**80**
4	5	30	0	15	—
6	10	30	5	20	—
7	25	40	30	55	—
8	30	35	35	65	—
9	25	25	20	50	—
10	45	35	40	100	20
11	5	30	5	10	—
12	20	35	15	35	—
13	35	30	40	75	—
14	25	30	30	50	—
15	65	40	55	100	40
16	15	30	15	15	—
17	30	15	35	60	—
18	40	35	45	75	—
19	15	15	20	30	—
20	20	10	30	40	—
21	25	20	20	50	—
22	10	20	15	25	—
23	**70**	**50**	**60**	**100**	**80**
24	35	20	30	75	—
25	40	45	35	85	—
26	40	20	50	85	—
27	15	10	10	35	—
28	45	20	50	100	10
29	45	20	55	100	10
30	25	20	30	55	—
31	20	30	30	40	—
32	45	45	40	100	20
33	20	25	15	45	—

activity. For example, compound **26** containing an unsaturated aliphatic chain (35.2%, 500 μg/mL) and **27** (32.6%, 500 μg/mL) containing a saturated short-chain displayed lower anti-TMV activities than **28** (61.3%, 500 μg/mL), **29** (59.6%, 500 μg/mL), and **30** (64.4%, 500 μg/mL). When R^2 was methyl, the compounds **31** (30.2%, 500 μg/mL), **32** (58.1%, 500 μg/mL), and **33** (43.4%, 500 μg/mL) exhibited much lower antiviral activities than **3** (55.8%, 500 μg/mL), **30** (64.4%, 500 μg/mL), and **29** (59.6%, 500 μg/mL), which meant that the hydrogen-bond in NH═CH played an important role in the anti-TMV activity.

Further bioassay was conducted to investigate their inactivation, curative, and protection effect *in vivo* (Table 2). These results also showed that most of the compounds exhibited excellent antiviral activity and similar structure−activity relationship as the activities *in vitro*. Especially the activities of compounds **9** (70.4%, 71.5%, and 64.2% at 500 μg/mL), **12** (69.1%, 66.4%, and 70% at 500 μg/mL), **16** (74.5%, 69%, and 68.1% at 500 μg/mL), **28** (73.4%, 70.9%, and 59.6% at 500 μg/mL), **29** (75.8%, 62.8%, and 69.2% at 500 μg/mL), and **30** (63.7%, 68.2%, and 66.8% at 500 μg/mL) were much superior to ribavirin (37.6%, 39.4%, and 37.9% at 500 μg/mL). What is more, their anti-TMV activities (≥30%) at 100 μg/mL were close to ribavirin at 500 μg/mL.

Fungicidal Activity. The fungicidal results are listed in Table 3. Most of β-carboline, tetrahydrogen-β-carboline alkaloids, and their derivatives showed good fungicidal activities against 14 kinds of phytopathogens; especially compounds **6**, **9**, and **11** exhibited desirable fungicidal activities against each of the phytopathogens. Additionally, tetrahydrogen-β-carboline derivatives exhibited higher activities than β-carboline derivatives. When the skeleton was tetrahydrogen-β-carboline (C ring was tetrahydropyridine), the fungicidal activities were greatly affected by the substituents (R^1 and R^2) on the acylhydrozone. The compounds with one hydrogen and one aryl group (**3**, **5**−**25**) exhibited high activities against fungi; whereas the compounds with no hydrogen or with at least one alkyl group (**26**−**33**) exhibited relatively poor activities. Noteworthily, the compounds containing aryl groups exhibited fungicidal activities to certain fungi selectively; they showed apparently higher activities against *Physalospora piricola, Sclerotinia sclerotiorum, Phytophthora capsici, Rhizoctonia cerealis, Bipolaris maydi*, and *Botrytis cinerea* than the others.

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Table 5. Results of Anti-TMV Activities of Field Trial in 2014 after 10 Days of Application of 9, 29, Amino-oligosaccharins, and Moroxydine Hydrochloride−Cupric Acetate

compd[a]	concn (gai/ha)	I'_{eq} [b]	I_{eq} [c]	E [d] (%)
9 (1% EC)	10	1.14	1.52	65.28
	50	1.86	4.92	80.82
	100	0.00	0.00	100.00
29 (1% ME)	10	1.10	1.52	70.91
	50	0.79	0.00	100.00
	100	0.00	0.00	100.00
blank control		2.56	5.23	
amino-oligosaccharins (5% aqueous solution)	100	3.33	3.33	66.67
moroxydine hydrochloride−cupric acetate (20% WP)	600	1.23	2.67	66.67

[a]EC, emulsifiable concentrate; ME, microemulsion; WP, wettable powder. [b]I'_{eq}, the average disease index of the spraying area before spraying. [c]I_{eq}, the average disease index of the spraying area 10 days after the third time spraying. [d]E, the average control effect 10 days after the third time spraying.

Figure 7. Effect of anti-TMV activities in field trials of compounds **9** and **29**.

Insecticidal Activity. These derivatives also showed broad-spectrum insecticidal activities (Table 4). What is more, compounds **3** and **23**, which contain a phenyl group or a pyrryl group, respectively, exhibited moderate insecticidal activity against the four kinds of insects (*Mythimna separata*, *Helicoverpa armigera*, *Ostrinia nubilalis*, and *Culex pipiens pallens*).

Field Trials. Because compounds **9** and **29** exhibited excellent anti-TMV activity in laboratory, 1% EC of compound **9** and 1% ME of compound **29** were evaluated for their anti-TMV activities in Tengchong County, Yunnan Province, China. Their efficacy was compared with the amino-oligosaccharins, moroxydine hydrochloride−cupric acetate, and an untreated control. Table 5 presents part of the results of field trials. Compounds **9** and **29** (Figure 7) exhibited better activities than control plant virus inhibitors. The results showed that the series of compounds, especially compound **29**, are promising for plant virus control.

In summary, we designed and synthesized a series of novel tetrahydrogen-β-carboline and β-carboline derivatives containing an acylhydrazide or acylhydrazone moiety at the 3-position. The hydrogen bonding effect was verified via variable-temperature (VT) ^{1}H NMR studies. What is more, the target compounds exhibited good anti-TMV activity in the laboratory; especially, *in vivo*, inactivation, curative, and protection activities of compounds **8**, **9**, **12**, **16**, **28**, **29**, and **30** were much higher (57.1−75.8% at 500 μg/mL) than that of ribavirin (37.6%, 37.6%, 39.4%, and 37.9% at 500 μg/mL) and the lead compound **1** (40.4%, 40.0%, 42.3%, and 39.6% at 500 μg/mL), and the relevant SAR was summarized. At the same time, field trials of compounds **9** and **29** further demonstrated the antiviral efficacy against TMV. In addition, we were pleased to find that these compounds also showed broad-spectrum of fungicidal activity and insecticidal activity. For instance, compounds **3**, **6**, **9**, and **11** exhibited good inhibition of the 14 kinds of phytopathogens; compounds **3** and **23** exhibited moderate insecticidal activity against four kinds of investigated insects. Further studies on structural optimization are in progress in our laboratory.

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Notes

The authors declare no competing financial interest.

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