Synthesis, characterization and biological activity of novel (5-*RS*,6-*S*)-5-*sec*-butyl-3-(1-substituted-amino)ethylidene--1*H*-pyrrolidine-2,4-diones

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Abstract

A series of novel tetramic acid derivatives, 5-*sec*-butyl-3-(1-substitutedamino)ethylidene-1*H*-pyrrolidine-2,4-diones **5a-y** were synthesized by reaction with aryl amines or alkyl amines under reflux. Each title compound was formed as (5*S*,6*S*) and (5*R*,6*S*) C-5 epimers, and the structure of **5l** was proved by X-ray diffraction analysis. Our preliminary bioassay results show the title compounds to exhibit some herbicidal activities and better antifungal activities than the leading compound tenuazonic acid at 100 mg L^{-1} *in vitro*, and the compound **5u** displayed excellent herbicidal activity and antifungal activity.

Keywords: Pyrrolidine-2,4-dione, tenuazonic acid, synthesis, crystal structure, biological activity

Introduction

Tenuazonic acid, a metabolic toxin from widely differing phytopathogenic fungi,[1] was first isolated from the culture filtrates of *Alternaria tenuis*.[2] Its structure was established as 3-acetyl-5-*sec*-butyltetramic acid Figure 1, **A** and it is believed to be the first substituted tetramic acid isolated from natural sources.[3] It is a potent inhibitor of protein biosynthesis[4] and possesses a wide range of biological activities, including anti-tumor, antibacterial, antiviral, and insecticidal properties.[5–8] Tenuazonic acid can inhibit seed germination and cause a brown leaf spot disease in many plants.[9–12] The previous studies seemed to support the view that tenuazonic acid inhibited protein synthesis in eukaryotic cells,[4] and had weak inhibition of HPPD (*p*-hydroxyphenylpyruvate dioxygenase).[13] Moreover, recent work revealed that tenuazonic acid

inhibited photosynthesis by blocking the photosystem II electron flow Q_A to Q_B,[14] and its half-life was only about 3.22 days in soil.[15]

Structural modification of tenuazonic acid to produce better bioactive compounds has attracted broad interest. Folkes reported a series of tetramic acid derivatives containing an imide function at the 3-position and found that some of them show excellent potency against plasminogen activator inhibitor-1 Figure 1, **B**.[16] Zhu synthesized 3-[(*α*-hydroxy-substituted)benzylidene]pyrrolidine-2,4-diones and found them to exhibit good herbicidal activities Figure 1, **C**.[17] Recently, Raghunandan reported *N*-Substituted-3- acetyltetramic acid derivatives displaying antibacterial activities (Figure 1, **D**).[18] Additionally, some tenuazonic acid analogs such as, reutericyclin, cryptocin, *β*-cyclopiazonic acid, and melophlin A and B, derived from natural products, have been found and displayed a wide range of biological activities.[19–21]

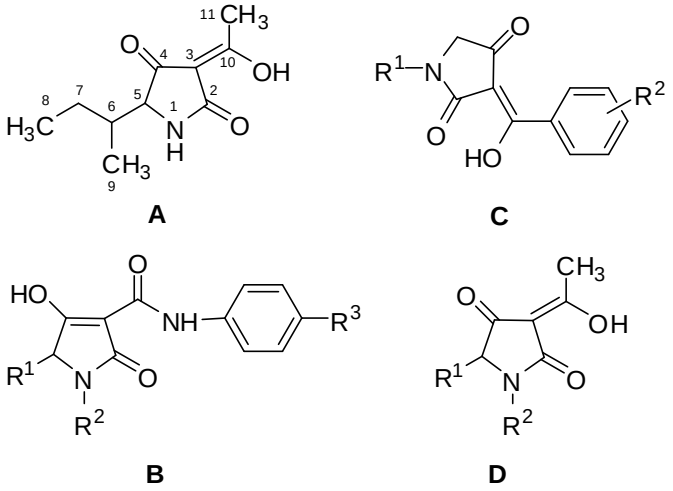


Figure 1. Structures of the compounds.

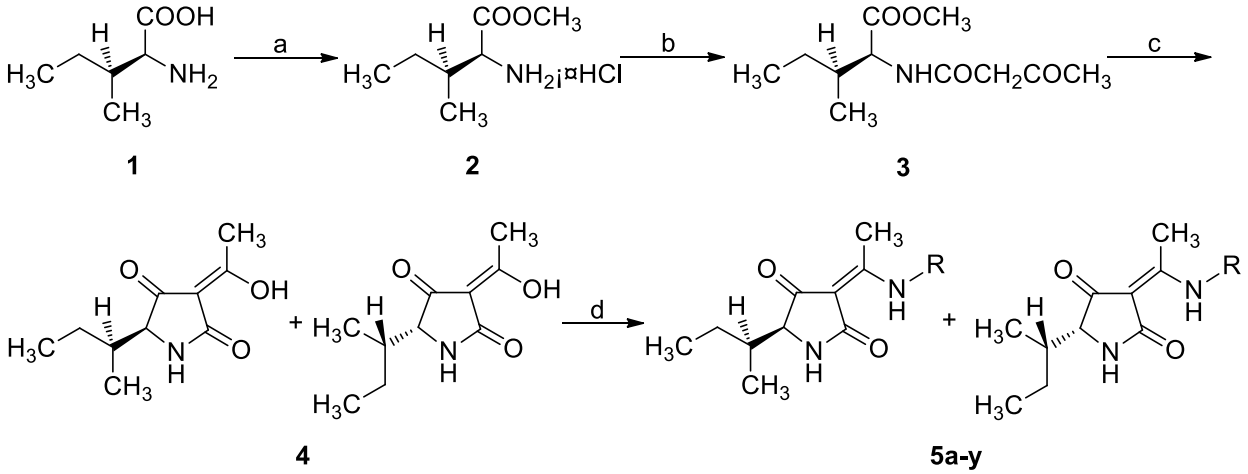
We synthesized twenty-five novel tetramic acid derivatives, 5-*sec*-butyl-3-(1-substituted-amino)ethylidene-1*H*-pyrrolidine-2,4-diones, **5a-y**, containing (5*S*,6*S*) and (5*R*,6*S*) C-5 epimers. Only one compound, **5i**, had been studied for antitumor activity.[22] Their structures were confirmed by IR, ^{1}H- NMR, MS, and elemental analysis. Fortunately, a single crystal of **5l** was grown, and the structure was characterized by X-ray diffraction analysis to characterize the two isomers of the title compounds. The preliminary bioassay tests against rape (*Brassica campestris*) and barnyard grass (*Echinochloa crusgalli*) showed that some of the compounds possessed a certain degree of inhibition activity against the stem or root growth at 100mg L^{-1} *in vitro*. Some of the compounds were found to exhibit better antifungal activities than tenuazonic acid against five fungi (*Fusarium graminearum*, *Rhizoctonia cerealis*, *Colletotrichum capsici*, *Botrytis cinerea* and *Fusarium moniliforme*) at 100 mg L^{-1} *in vitro*. We shall also discuss their structure-activity relationships.

Results and Discussion

Chemistry

The intermediate **4** was prepared by the reported method, starting from *L*-isoleucine, through esterification, *N*-aceto-acetylation and a cyclization reaction as shown in Scheme 1.[23] It was noted that during the cyclization, epimerization occurred at the position C-5 to give two epimers (5*S*, 6*S*) and (5*R*, 6*S*), and the diastereoisomer ratio was related to the sodium methoxide concentration and reaction time.[24] Therefore **4,** a buff powder, is a mixture of two epimers.[3] Owing to the existence of different keto-enol tautomeric forms, 3-acetyl-5-*sec*-butyltetramic acid are represented herein in the 3-*exo*-enol form **A** found in the solid state, and as the major tautomer in solution for many cases.[25]

Subsequently, a series of the compounds 5-*sec*-butyl-3-(1-substitutedamino)ethylidene-1*H*-pyrrolidine-2,4-diones **5a-y** were synthesized by reacting the compounds **4** with aryl amine or alkyl amine in ethanol under refluxing condition (Scheme 1). The reaction progress was monitored by TLC (light petroleum/ethyl acetate, v/v 1:1). The reaction time was significantly reduced by adding two drops of concentrated hydrochloric acid or glacial acetic acid as catalysts, but the catalyst was not needed for the reaction with alkyl amine. Each target compound was found being a diastereoisomer with two C-5 epimers of (5*S*, 6*S*) and (5*R*, 6*S*) isomers. These two C-5 epimers showed almost same properties, such as, same state, same melting point with short melting range, almost same solubility. They usually appeared in dimer forms by strong hydrogen bonds interaction (Figure 2). It is difficult to isolate the single isomer from two epimers.



Scheme 1. Synthetic route to the title compounds **5a-y**. Reagents and conditions: (a) $SOCl_2$, CH_3OH, reflux; (b) CH_3ONa then $(CH_2CO)_2$, RT; (c) CH_3ONa, benzene, reflux, then 20%HCl, H_2O; (d) $R–NH_2$, C_2H_5OH, reflux.

The structures of all synthesized compounds were determined by spectroscopic techniques

and elemental analysis. The IR spectra of the target compounds had low values for NH (3200–3350 cm^{-1}) because of formation of hydrogen bonds. In the MS, each target compound showed a weak molecular ion peak, but had an intense characteristic ion peak (M^+–C_4H_9). In the ^{1}H- NMR spectra, the chemical shifts and integration of the CH_3CNH protons showed the existence of two isomers in each target compound, with a ratio of approximately 1:1. Thus, for (5*RS*, 6*S*)-5-*sec*-butyl-3-(1-4-methylphenylamino)ethylidene-1*H*-pyrrolidine-2,4-dione **5j**, the signals of three methyl protons in C*H*$_3$CNH appeared at 2.52 (52%) and 2.54 (48%), and the NH proton signals in CH_3CN*H* appeared at 12.03 (52%) and 12.48 (48%), respectively.

Crystal structures of two isomers of the compound 5l

The 5*S*,6*S*- isomer and 5*R*,6*S*- isomer of the compound **5l** were characterized by single crystal X-ray determination. The molecular structures of the two isomers of the compound **5l** shown in Figure 2 reveal the 5*S*,6*S*-isomer's (molecule *A*) and the 5*R*,6*S*-isomer's (molecule *B*) absolute configuration in the asymmetric unit. They are connected by intermolecular hydrogen bonds N2–H2A···O4 and N4–H4A···O2 (Table 1) and are a diastereomeric mixture.

The bond lengths C7–C8 [1.379(8) Å] and C25–C26 [1.376(10) Å] are close to the C═C bond distance (1.38 Å, conjugated system). In addition, C25, C7 are coplanar with respective connected pyrrolidine-2,4-dione ring, and the deviation from the least-squares plane α (defined by C25, C26, C27, C28, C29, O3, O4, N4) through the ring atoms is less than 0.0603 Å in molecule *A*, and less than 0.0334 Å from the least-squares plane β (defined by C7, C8, C9, C10, C11, O1, O2, N2) in the molecule *B*. It is confirmed that C25–C26 and C7–C8 are double bonds. The bond lengths of C26–C27 [1.444(7) Å], C26–C29 [1.493(11) Å], C8–C9 [1.420(7) Å] and C8–C11 [1.484(8) Å] are shorter than the normal C–C bond (1.54 Å). N3–C25 [1.321(9) Å], N4–C29 [1.345(7) Å], N1–C7 [1.337(8) Å] and N2–C11 [1.351(6) Å] bonds are shorter than the normal C–N bond (1.49 Å), suggesting the existence of an electron-density delocalization among N3–C25–C26–C29–N4, C27 and among N1–C7–C8–C11–N2, C9, and form a large conjugate system with O3═C27 [1.208(9) Å], O4═C29 [1.283(7) Å] in molecule *A* and O1═C9 [1.222(8) Å] and O2═C11 [1.283(6) Å] in the molecule *B*, respectively.

In the packing diagram of **5l** (Figure 3), a one-dimensional chain structure was formed by two intermolecular hydrogen bonds N1–H1A···O3a and N3a–H3Aa···O1 (Table 1), between two adjacent dimers.

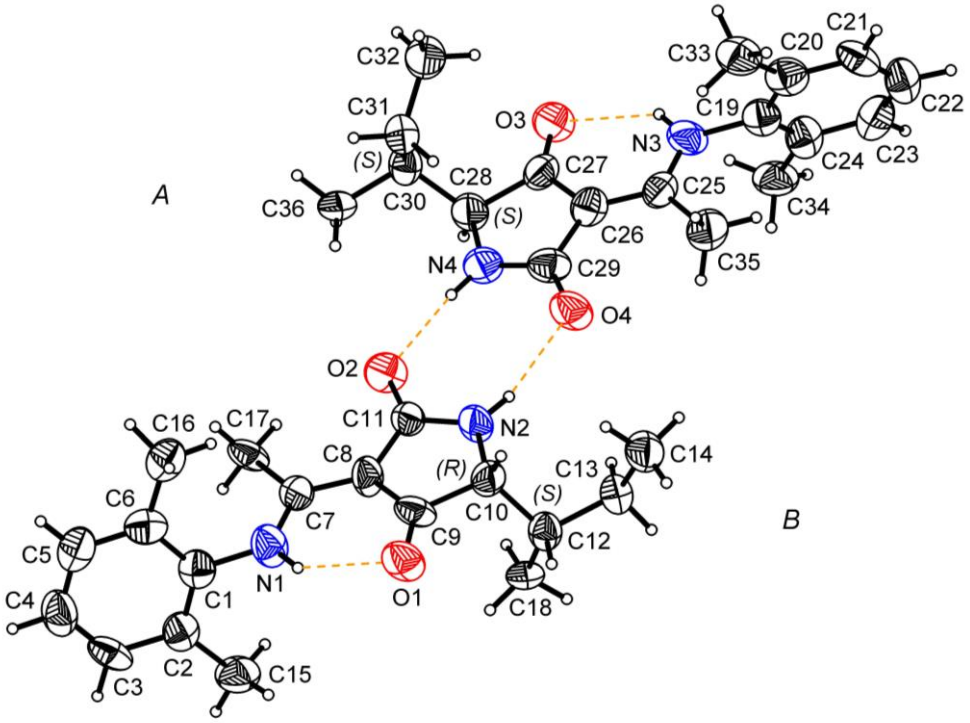


Figure 2. The molecular structure of the compound **5l**, with thermal ellipsoids drawn at the 50% probability level. Dashed lines show hydrogen bonds.

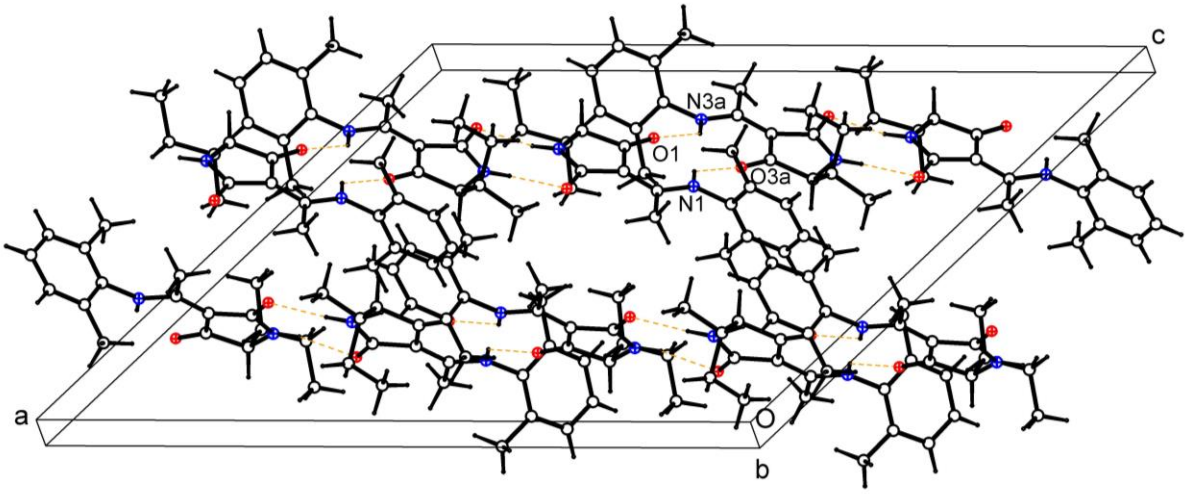


Figure 3. Packing diagram of the compound **5l** showing intermolecular interactions (dashed lines).

Table 1. Hydrogen bonding geometry of the compound **5l** (Å,°)

D–H···A	D–H	H···A	D···A	D–H···A
N1–H1A···O1	0.86	2.05	2.745(6)	137
N1–H1A···O3[a]	0.86	2.43	3.083(6)	134
N2–H2A···O4	0.86	2.03	2.868(8)	163
N3–H3A···O3	0.86	2.06	2.741(6)	135
N3–H3A···O1[a]	0.86	2.41	3.082(6)	136
N4–H4A···O2	0.86	1.99	2.843(8)	172

Symmetry code: [a] -1/2+x, -1/2+y, z.

Biological evaluation

The inhibition activities of the title compounds against the stem and root of rape and barnyard grass were determined using the reported method.[26] The results as shown in Table 2 indicated that some compounds showed certain inhibition activities at the concentration of 100 mg L^{-1}, almost all the compounds displayed better inhibition activities against the root than the corresponding stem. The data in Table 2 also indicated that most compounds had higher inhibition rates when R were aryl groups than alkyl groups. Further, when the substituents on the phenyl ring were changed, the inhibition activity showed significant differences. With an unsubstituted phenyl ring, the compound **5i** displayed inhibition rates of 89.5% and 52.5% against the root and stem growth of rape, respectively. When the phenyl ring had an electron-donating group, the corresponding target compounds gave weaker inhibition effects, but when modified by the electron- attracting group, of which the 3-position at the phenyl ring was brominated, the inhibition rates of compound **5u** reached 94.4% and 67.4% against the root and stem growth of rape, respectively.

In addition, all the target compounds were screened for antifungal activities *in vitro* against five selected phytopathogenic fungi, *F. graminearum*, *R. cerealis*, *C. capsici*, *B. cinerea*, and *F. moniliforme*. As shown in Table 3, some of the compounds displayed moderate antifungal activities at a concentration of 100 mg L^{-1}. It was similar to the herbicidal activity in that when R were substituted-phenyl groups, the corresponding target compounds always had higher antifungal activities. Introducing different substituted groups at the phenyl ring could enhance the fungicidal activities **5j**–**v**, especially, against the mycelia growth of *B. cinerea*. For example, the compounds **5m** and **5u** with, respectively, 2-CH_3-6-C_2H_5- and 3-Br- at the phenyl ring, inhibited mycelia growth of *B. cinerea* at rates of 63.4% and 70.9%, respectively. The compounds **5r** and **5t,** with respectively 4-Cl and 3,4-Cl_2 on the phenyl ring inhibited three fungi, *R. cerealis*, *C. capsici*, and *B. cinerea*, at rates of all above 50%. However, when R was replaced with thiazole, or benzidine respectively, the target compounds did not present satisfactory antifungal activities against the tested fungi. It is worth noting that most of the target compounds showed better antifungal activities than does tenuazonic acid.

Table 2. Herbicidal activities of the title compounds (100 mg L^{-1}, inhibition %)[a]

Compd.	R	*B. campestris*		*E. crusgalli*	
		Stem	Root	Stem	Root
5a	CH_3–	16.4 ± 2.7	11.5 ± 1.5	22.3 ± 7.4	28.0 ± 9.6
5b	CH_3CH_2–	N.A.[b]	3.5 ± 1.2	23.1 ± 5.9	24.8 ± 6.3
5c	$CH_3CH_2CH_2$–	20.5 ± 9.0	49.5 ± 2.5	18.8 ± 5.2	26.6 ± 7.8
5d	$(CH_3)_2CH$–	11.1 ± 2.7	10.4 ± 1.2	5.8 ± 1.3	10.9 ± 6.0
5e	$CH_3CH_2CH_2CH_2$–	N.A.	30.0 ± 1.8	4.1 ± 1.9	24.5 ± 3.5
5f	$CH_3CH_2(CH_3)CH$–	N.A.	24.7 ± 3.1	10.4 ± 2.2	19.5 ± 4.7
5g	$(CH_2)_5CH$–	29.2 ± 1.0	78.9 ± 2.2	9.7 ± 2.6	42.7 ± 5.0
5h	$C_6H_5CH_2$–	16.3 ± 6.3	21.5 ± 5.0	2.7 ± 1.3	34.3 ± 3.8
5i	C_6H_5–	52.5 ± 6.4	89.5 ± 4.5	3.6 ± 1.9	43.2 ± 4.4
5j	4–$CH_3C_6H_4$–	21.6 ± 2.3	30.1 ± 4.0	N.A.	48.1 ± 8.6
5k	3,4–$(CH_3)_2C_6H_3$–	19.5 ± 2.3	29.0 ± 1.7	N.A.	31.9 ± 4.3
5l	2,6–$(CH_3)_2C_6H_3$–	N.A.	6.4 ± 1.6	3.6 ± 1.3	40.4 ± 4.1
5m	2–CH_3–6–$C_2H_5C_6H_3$–	8.3 ± 2.7	N.A.	10.1 ± 1.6	13.1 ± 5.4
5n	4–CH_3O–C_6H_4–	23.7 ± 4.8	67.8 ± 1.9	N.A.	26.8 ± 8.8
5o	2–C_2H_5O–C_6H_4–	16.8 ± 4.3	56.0 ± 10.3	4.8 ± 3.7	33.5 ± 3.6
5p	4–HO–C_6H_4–	N.A.	10.7 ± 4.9	N.A.	N.A.
5q	4–F–C_6H_4–	11.7 ± 2.3	44.6 ± 3.0	N.A.	63.0 ± 3.6
5r	4–Cl–C_6H_4–	17.8 ± 2.4	27.1 ± 3.4	3.9 ± 1.9	34.7 ± 3.1
5s	2,4–$(Cl)_2C_6H_3$–	20.0 ± 5.8	29.7 ± 4.1	4.9 ± 2.1	31.4 ± 4.7
5t	3,4–$(Cl)_2C_6H_3$–	N.A.	N.A.	N.A.	33.8 ± 8.3
5u	3–Br–C_6H_4–	67.4 ± 6.8	94.4 ± 1.1	3.9 ± 2.4	44.8 ± 4.0
5v	4–NO_2–C_6H_4–	26.9 ± 0.7	52.2 ± 3.7	N.A.	67.8 ± 10.0
5w	$C_{10}H_7$–1–	9.5 ± 3.7	13.5 ± 4.3	N.A.	7.5 ± 3.1
5x	4–(4´–NH_2–C_6H_4)–C_6H_4–	20.0 ± 7.6	39.6 ± 1.5	N.A.	N.A.
5y	C_3H_2NS–2–	8.1 ± 1.5	46.0 ± 4.9	N.A.	46.3 ± 1.5
4	—	77.5 ± 4.3	97.7 ± 1.1	50.2 ± 3.7	98.7 ± 0.8

[a] The values are expressed as means ± SD of the replicates; n = 3 for all groups. [b] N.A.= Not active.

Table 3. Results of antifungal activities of the title compounds (100 mg L^{-1}, inhibition %)[a]

Compd.	*F. graminearum*	*R. cerealis*	*C. capsici*	*B. cinerea*	*F. moniliforme*
5a	13.0 ± 1.3	18.8 ± 2.5	13.1 ± 1.8	25.8 ± 2.1	2.6 ± 0.8
5b	N.A.[b]	N.A.	1.4 ± 0.8	44.8 ± 1.6	N.A.
5c	3.6 ± 2.7	N.A.	N.A.	1.5 ± 1.0	N.A.
5d	14.0 ± 0.7	N.A.	N.A.	2.4 ± 1.1	N.A.
5e	10.3 ± 1.3	N.A.	2.2 ± 2 0	5.9 ± 1.3	2.2 ± 0.9
5f	9.0 ± 0	N.A.	3.5 ± 0	3.2 ± 1.6	N.A.
5g	16.1 ± 0.8	N.A.	7.4 ± 0	N.A.	N.A.
5h	26.6 ± 1.6	27.7 ± 1.5	15.6 ± 0.8	29.6 ± 1.4	17.7 ± 1.4
5i	23.5 ± 1.5	23.2 ± 4.2	29.0 ± 2.3	33.8 ± 3.7	9.8 ± 1.1
5j	35.3 ± 3.9	42.4 ± 1.5	30.5 ± 2.3	53.5 ± 2.4	36.3 ± 1.8
5k	52.6 ± 2.2	40.4 ± 2.0	51.0 ± 2.5	55.4 ± 3.1	46.8 ± 2.7
5l	13.5 ± 1.4	44.1 ± 2.4	36.9 ± 0.9	51.2 ± 0.8	17.3 ± 1.6
5m	14.7 ± 3.8	39.9 ± 1.7	46.7 ± 1.5	63.4 ± 8.7	27.1 ± 0.8
5n	29.4 ± 2.9	40.5 ± 3.1	20.4 ± 0.9	40.4 ± 3.5	11.2 ± 1.3
5o	27.1 ± 1.6	34.4 ± 2.4	34.1 ± 1.3	50.7 ± 1.4	13.5 ± 1.4
5p	9.8 ± 0.8	5.1 ± 2.4	11.8 ± 2.6	11.7 ± 3.3	15.9 ± 2.9
5q	22.8 ± 1.6	42.6 ± 2.3	10.4 ± 2.2	27.2 ± 0.8	11.7 ± 2.1
5r	35.8 ± 1.7	58.2 ± 4.2	52.3 ± 1.8	55.9 ± 3.3	41.9 ± 1.6
5s	26.6 ± 2.1	34.4 ± 3.2	22.2 ± 1.6	53.5 ± 3.8	15.4 ± 2.1
5t	48.8 ± 2.1	54.9 ± 2.4	50.7 ± 0.8	56.8 ± 3.4	38.2 ± 0.8
5u	35.4 ± 4.3	46.7 ± 0.9	51.2 ± 2.1	70.9 ± 4.1	36.8 ± 2.2
5v	30.3 ± 5.0	50.8 ± 1.5	16.1 ± 1.4	36.2 ± 2.9	30.3 ± 4.6
5w	27.1 ± 0.8	33.9 ± 3.2	44.1 ± 0.8	58.7 ± 2.9	25.7 ± 2.1
5x	30.3 ± 3.5	N.A.	11.7 ± 2.2	N.A.	N.A.
5y	10.3 ± 1.2	21.1 ± 2.4	5.1 ± 2.2	20.2 ± 4.5	12.1 ± 1.4
4	10.9 ± 1.7	13.7 ± 3.1	14.6 ± 3.0	18.3 ± 2.8	10.3 ± 2.9

[a] The values are expressed as means ± SD of the replicates; n = 3 for all groups. [b] N.A.= Not active.

Conclusions

In summary, a series of novel tetramic acid derivatives containing two C-5 epimers were synthesized by the treatment of intermediate **4** with aryl- amines or alkyl- amines under refluxing condition. Their structures were verified by spectroscopic data and single- crystal structure

characterization. Bioassays showed that the title compounds exhibit lower herbicidal activities, but better antifungal activities than tenuazonic acid. The compound (5*RS*,6*S*)-5-*sec*-butyl-3-[1-(3-bromophenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione **5u** displayed both excellent herbicidal activity and antifungal activity. It was found that the 3-acetyl function is very important for maintaining the herbicidal activity of tenuazonic acid, and introducing a substituted phenylamino group at the 3-position group can obviously enhance its antifungal activity. Further investigations are in progress.

Experimental Section

General. Melting points were determined on a WRS-1B digital melting-point apparatus and were uncorrected. IR spectra (4000–400 cm^{-1}) were recorded on a Bruker Tensor27 FT-IR spectrometer, using KBr disks. ^{1}H- NMR spectra were recorded on a Varian Mercury-Plus at 300MHz at room temperature, in $CDCl_3$ or DMSO solutions. Chemical shifts are given in δ units (ppm) relative to TMS as internal standard. Optical rotations were measured on an ATAGO Automatic Polarimeter (AP-100) in a 1dm cell at 25 °C. Mass spectra were recorded on a GC/MS-QP2010 spectrometer using the direct injection technique. Elemental analyses were performed on a Vario EL III elemental analysis instrument. The progress of the reactions was routinely monitored by thin layer chromatography (TLC) on silica gel GF254 and the products were visualized with an ultraviolet lamp (254 and 365 nm). All reagents and starting materials were obtained from commercial suppliers and were used without further purification.

Synthesis of 3-acetyl-5-*sec*-butyltetramic acid 4. The intermediates **4** were prepared with modified procedures according to the reported method.[23] Methyl *N*-acetoacetyl- L-iso-leucinate **3** (0.2 mol, 42.7 g) and sodium methoxide solution (Na metal: 0.25 mol, 5.75 g; methanol: 60 mL) were mixed and the mixture refluxed in benzene (60 mL) with stirring for 3 h. Then the solvent was removed under reduced pressure, and water (100 mL) was added. The impurities were extracted with ethyl acetate (3x30 mL). The aqueous layer was separated and acidified to pH 2–3 with 20% HCl. The acidic solution was extracted with ethyl acetate (3x30 mL), and the extracts washed with saturated brine. After drying (Na_2SO_4), the solvent was evaporated off under reduced pressure to give crude 3-acetyl-5-*sec*-butyltetramic acid **4** as an orange-red oil, and direct crystallization from ethanol below 0 °C gave **4** as a buff powder, yield 71%, mp 61–63 °C (lit.,[2] 61–62.5 °C); $[\alpha]_D^{23}$ +14.53 (*c* 1.0, methanol) {lit.,[2] $[\alpha]_{5461}^{22}$ +23 (*c* 0.5, chloroform)}; ^{1}H-NMR (300 MHz, $CDCl_3$): δ 0.80 (d, 3H, *J*=6.7, C*H*$_3$CH), 1.00 (t, 3H, *J*=7.1, C*H*$_3$CH$_2$), 1.20–1.46 (m, 2H, CH$_3$C*H*$_2$), 1.92–2.07 (m, 1H, CH$_3$C*H*), 2.46 (s, 3H, CH$_3$CO), 3.79–4.01 (m, 1H, C*H*NH), 6.83 (br, 1H, NH), 11.55 (br, 1H, OH); IR (KBr) 3340, 3201, 3075, 2968, 2877, 1717, 1676, 1646, 1582, 1450, 1233 cm^{-1}; EI-MS, *m/z* (%): 197 (M^+, 2), 141 (100), 123 (18), 98 (10), 57 (12); Anal. Calcd for $C_{10}H_{15}NO_3$ (197.23): C 60.90; H 7.67; N 7.10. Found C 60.74; H 7.71; N 7.23%.

General procedure for preparation of (5*RS*,6*S*)-5-*sec*-butyl-3-(1-substituted-amino)-ethylidene -1*H*-pyrrolidine-2,4-diones 5a–y

The solid compound **4** (5.08 mmol, 1.00 g) was dissolved in absolute ethanol (20 mL), and an equimolar quantity of substituted amine was added. The mixture was refluxed under stirring. When TLC analysis showed that most of the starting material had been converted, the reaction was stopped. The reaction mixture was concentrated under reduced pressure, and the oily residue was crystallized from petroleum/ethyl acetate (2 mL, 1:1 v/v). After standing overnight at 0 °C, the precipitate was filtered off, washed with water, dried, and recrystallized from light petroleum/ethyl acetate (1:1, v/v) or methanol **5h, 5v, 5y** to provide the desired products **5a–y**.

(5*RS*,6*S*)-5-*sec*-Butyl-3-(1-methylamino)ethylidene-1*H*-pyrrolidine-2,4-dione 5a. White powder, yield 57%; mp 113–114 °C; $[\alpha]_D^{25}$ +20.52 (*c* 0.50, methanol); IR (KBr) 3320, 3207, 2961, 2875, 1677, 1626, 1597, 1057, 1356, 1172 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.75 (d, 3H, *J*=6.3, C*H*$_3$CH), 0.97 (t, 3H, *J*=6.6, C*H*$_3$CH$_2$), 1.23–1.43 (m, 2H, CH$_3$C*H*$_2$), 1.88–2.02 (m, 1H, CH$_3$C*H*), 2.55, 2.57 (s, s, 40/60, 3H, C*H*$_3$CNH), 3.06 (s, 3H, NHC*H*$_3$), 3.66–3.81 (m, 1H, C*H*NH), 5.68 (br, 1H, NH), 10.45, 10.96 (s, s, 40/60, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 210 (M^+, 8), 154 (100), 126 (21), 98 (15), 56 (25); Anal. Calcd for $C_{11}H_{18}N_2O_2$ (210.27): C, 62.83; H, 8.63; N, 13.32. Found: C, 62.71; H, 8.49; N, 13.11%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-(1-ethylamino)ethylidene-1*H*-pyrrolidine-2,4-dione 5b. White powder, yield 53%; mp 94–96 °C; $[\alpha]_D^{25}$ +15.29 (*c* 0.50, methanol); IR (KBr) 3295, 3226, 2963, 2875, 1679, 1635, 1599, 1504, 1339, 1162 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.76 (d, 3H, *J*=7.2, C*H*$_3$CH), 0.98 (t, 3H, *J*=6.6, C*H*$_3$CH$_2$CH), 1.18–1.45 (m, 5H, CH$_3$C*H*$_2$CH+NHCH$_2$C*H*$_3$), 1.89–2.02 (m, 1H, CH$_3$C*H*), 2.56, 2.57 (s, s, 45/55, 3H, C*H*$_3$CNH), 3.43 (br, 2H, NHC*H*$_2$CH$_3$), 3.65–3.80 (m, 1H, C*H*NH), 5.53 (br, 1H, NH), 10.45, 10.96 (s, s, 45/55, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 224 (M^+, 9), 168 (100), 140 (17), 112 (14), 70 (15); Anal. Calcd for $C_{12}H_{20}N_2O_2$ (224.30): C, 64.26; H, 8.99; N, 12.49. Found: C, 64.02; H, 9.19; N, 12.34%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-(1-*n*-propylamino)ethylidene-1*H*-pyrrolidine-2,4-dione 5c. Colorless crystals, yield 39%; mp 117–119 °C; $[\alpha]_D^{25}$ +37.93 (*c* 0.50, methanol); IR (KBr) 3321, 3227, 2966, 1706, 1623, 1581, 1512, 1309, 1182 cm^{-1} ; ^1H NMR (300 MHz, $CDCl_3$): δ 0.75–1.17 (m, 9H, C*H*$_3$CH+C*H*$_3$CH$_2$CH+CH$_2$CH$_2$C*H*$_3$), 1.28–1.75 (m, 4H, CH$_3$C*H*$_2$CH+CH$_2$C*H*$_2$CH$_3$), 1.95–2.05 (m, 1H, CH$_3$C*H*), 2.55, 2.56 (s, s, 49/51, 3H, C*H*$_3$CNH), 3.34 (t, 2H, *J*=8.4, C*H*$_2$CH$_2$CH$_3$), 3.65–3.80 (m, 1H, C*H*NH), 5.87 (br, 1H, NH), 10.56, 11.05 (s, s, 49/51, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 238 (M^+, 13), 182 (10), 126 (100), 97 (13), 57 (15); Anal. Calcd for $C_{13}H_{22}N_2O_2$ (238.33): C, 65.51; H, 9.30; N, 11.75. Found: C, 65.68; H, 9.21; N, 11.49%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-(1-*iso*-propylamino)ethylidene-1*H*-pyrrolidine-2,4-dione 5d. Colorless crystals, yield 54%; mp 159–160 °C; $[\alpha]_D^{25}$ +172.21 (*c* 0.50, methanol); IR (KBr) 3270, 3195, 2960, 2876, 1685, 1640, 1595, 1505, 1396, 1220 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.76 (d, 3H, *J*=6.6, C*H*$_3$CHCH$_2$), 0.96 (t, 3H, *J*=7.4, C*H*$_3$CH$_2$), 1.25–1.47 (m, 8H, CH$_3$C*H*$_2$+NCH(C*H*$_3$)$_2$), 1.92–2.03 (m, 1H, CH$_3$C*H*CH$_2$), 2.57, 2.59 (s, s, 47/53, 3H, C*H*$_3$CNH), 3.71–3.80 (m, 1H, CHC*H*NH), 3.87–3.99 (m, 1H, NC*H*(CH$_3$)$_2$), 5.41 (br, 1H, NH), 10.48, 10.99 (s, s, 47/53, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 238 (M^+, 7), 182 (100), 139 (11), 96 (14), 58 (16);

Anal. Calcd for $C_{13}H_{22}N_2O_2$ (238.33): C, 65.51; H, 9.30; N, 11.75. Found: C, 65.57; H, 9.27; N, 11.93%.

(5*RS*, 6*S*)-5-*sec*-Butyl-3-(1-*n*-butylamino)ethylidene-1*H*-pyrrolidine-2,4-dione 5e. Colorless crystals, yield 55%; mp 64–66 °C; $[\alpha]_D^{25}$ +13.69 (*c* 0.50, methanol); IR (KBr) 3275, 3211, 2960, 2874, 1685, 1635, 1598, 1504, 1405, 1217 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.76–1.00 (m, 9H, C*H*$_3$CH+C*H*$_3$CH$_2$CH+ CH$_2$CH$_2$C*H*$_3$), 1.19–1.71 (m, 6H, CH$_3$C*H*$_2$CH+C*H*$_2$CH$_2$CH$_3$+CH$_2$C*H*$_2$CH$_3$), 1.84–2.05 (m, 1H, CH$_3$C*H*), 2.55, 2.56 (s, s, 59/41, 3H, C*H*$_3$CNH), 3.27–3.43 (m, 2H, C*H*$_2$CH$_2$CH$_2$CH$_3$), 3.65–3.81 (m, 1H, C*H*NH), 5.57 (br, 1H, NH), 10.55, 11.06 (s, s, 59/41, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 252 (M^+, 6), 196 (100), 140 (14), 97 (10), 57 (9); Anal. Calcd for $C_{14}H_{24}N_2O_2$ (252.35): C, 66.63; H, 9.59; N, 11.10. Found: C, 66.87; H, 9.48; N, 11.29%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-(1-*sec*-butylamino)ethylidene-1*H*-pyrrolidine-2,4-dione 5f. Colorless crystals, yield 42%; mp 102–104 °C; $[\alpha]_D^{25}$ +16.35 (*c* 0.50, methanol); IR (KBr) 3291, 3224, 2966, 2876, 1705, 1638, 1596, 1503, 1403, 1151 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.74–0.97 (m, 9H, C*H*$_3$CHCH$_2$+C*H*$_3$CH$_2$CH+ NHCHCH$_2$C*H*$_3$), 1.13–1.42 (m, 5H, CH$_3$C*H*$_2$CH+NHCHC*H*$_3$), 1.56–1.65 (m, 2H, NHCHC*H*$_2$CH$_3$), 1.83–2.04 (m, 1H, CH$_3$C*H*CH), 2.54, 2.57 (s, s, 57/43, 3H, C*H*$_3$CNH), 3.62–3.79 (m, 2H, CHC*H*NH+NHC*H*CH$_3$), 5.76 (br, 1H, NH), 10.50, 10.98 (s, s, 57/43, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 252 (M^+, 5), 196 (100), 140 (20), 112 (8), 57 (9); Anal. Calcd for $C_{14}H_{24}N_2O_2$ (252.35): C, 66.63; H, 9.59; N, 11.10. Found: C, 66.91; H, 9.45; N, 11.33%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-(1-cyclohexylamino)ethylidene-1*H*-pyrrolidine-2,4-dione 5g. White powder, yield 71%; mp 137–138 °C; $[\alpha]_D^{25}$ +9.40 (*c* 0.50, methanol); IR (KBr) 3280, 3195, 2960, 2875, 1690, 1647, 1593, 1506, 1401, 1143 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.75–2.25 (m, 19H, C*H*$_3$C*H*$_2$(C*H*$_3$)C*H*+ 5C*H*$_2$(cyclohexyl)), 2.56, 2.58 (s, s, 47/53, 3H, C*H*$_3$CNH), 3.51–3.79 (m, 2H, C*H*NH+C*H*(cyclohexyl)), 5.63 (br, 1H, NH), 10.60, 11.11 (s, s, 47/53, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 278 (M^+, 10), 221 (100), 140 (26), 84 (13), 55 (17); Anal. Calcd for $C_{16}H_{26}N_2O_2$ (278.39): C, 69.03; H, 9.41; N, 10.06. Found: C, 69.15; H, 9.33; N, 10.12%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-(1-benzylamino)ethylidene-1*H*-pyrrolidine-2,4-dione 5h. Colorless crystals, yield 90%; mp 157–158 °C; $[\alpha]_D^{25}$ +16.44 (*c* 0.50, methanol); IR (KBr) 3209, 3198, 3059, 2967, 2877, 1672, 1624, 1592, 1581, 1500, 1451, 1394, 1134 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.78 (d, 3H, *J*=6.7, C*H*$_3$CH), 0.98 (t, 3H, *J*=6.9, C*H*$_3$CH$_2$), 1.23–1.45 (m, 2H, CH$_3$C*H*$_2$), 1.86–2.07 (m, 1H, CH$_3$C*H*), 2.59, 2.61 (s, s, 47/53, 3H, C*H*$_3$CNH), 3.67–3.83 (m, 1H, C*H*NH), 4.59 (s, 2H, ArC*H*$_2$), 5.78 (br, 1H, NH), 7.27–7.40 (m, 5H, ArH), 10.88, 11.34 (s, s, 47/53, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 286 (M^+, 6), 230 (100), 144 (19), 139 (159), 91 (100); Anal. Calcd for $C_{17}H_{22}N_2O_2$ (286.37): C, 71.30; H, 7.74; N, 9.78. Found: C, 71.14; H, 7.81; N, 9.65%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-(1-phenylamino)ethylidene-1*H*-pyrrolidine-2,4-dione 5i. White powder, yield 44%; mp 71–72 °C; $[\alpha]_D^{25}$ +24.74 (*c* 0.50, methanol); IR (KBr) 3339, 3201, 3075, 2968, 2899, 1717, 1676, 1646, 1582, 1450, 1194 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.83 (d, 3H, *J*=6.7, C*H*$_3$CH), 1.03 (t, 3H, *J*=7.4, C*H*$_3$CH$_2$), 1.27–1.50 (m, 2H, CH$_3$C*H*$_2$), 1.94–2.08 (m,

1H, CH_3CH), 2.55, 2.57 (s, s, 57/43, 3H, CH_3CNH), 3.74–3.92 (m, 1H, $CHNH$), 5.74 (br, 1H, NH), 7.21–7.46 (m, 5H, ArH), 12.11, 12.54 (s, s, 57/43, 1H, CH_3CNH); EI-MS, *m/z* (%): 272 (M^+, 7), 216 (100), 158 (17), 130 (20), 118 (23), 77 (15); Anal. Calcd for $C_{16}H_{20}N_2O_2$ (272.34): C, 70.56; H, 7.40; N, 10.29. Found: C, 70.42; H, 7.34; N, 10.35%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(4-methylphenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione 5j. White powder, yield 59%; mp 105–106 °C; $[\alpha]_D^{25}$ +16.81 (*c* 0.50, methanol); IR (KBr) 3290, 3177, 3046, 2960, 2873, 1683, 1633, 1600, 1574, 1520, 1494, 1390, 1213 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.83 (d, 3H, *J*=6.6, CH_3CH), 1.02 (t, 3H, *J*=7.2, CH_3CH_2), 1.25–1.50 (m, 2H, CH_3CH_2), 1.92–2.04 (m, 1H, CH_3CH), 2.40 (s, 3H, $ArCH_3$), 2.52, 2.54 (s, s, 52/48, 3H, CH_3CNH), 3.72–3.91 (m, 1H, $CHNH$), 5.49 (br, 1H, NH), 7.08–7.26 (m, 4H, ArH), 12.03, 12.48 (s, s, 52/48, 1H, CH_3CNH); EI-MS, *m/z* (%): 286 (M^+, 10), 230 (100), 172 (13), 132 (24), 91 (15), Anal. Calcd for $C_{17}H_{22}N_2O_2$ (286.37): C, 71.30; H, 7.74; N, 9.78. Found: C, 71.19; H, 7.82; N, 9.67%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(3,4-dimethylphenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione 5k. Colorless crystals, yield 79%; mp 124–125 °C; $[\alpha]_D^{25}$ +17.47 (*c* 0.50, methanol); IR (KBr) 3308, 3175, 3068, 2960, 2876, 1655, 1625, 1573, 1513, 1496, 1457,1392, 1166 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.83 (d, 3H, *J*=6.7, CH_3CH), 1.02 (t, 3H, *J*=7.2, CH_3CH_2), 1.29–1.51 (m, 2H, CH_3CH_2), 1.96–2.09 (m, 1H, CH_3CH), 2.30 (s, 6H, 2×$ArCH_3$), 2.52, 2.55 (s, s, 53/47, 3H, CH_3CNH), 3.73–3.90 (m, 1H, $CHNH$), 5.67 (br, 1H, NH), 6.91–7.20 (m, 3H, ArH), 12.00, 12.44 (s, s, 53/47, 1H, CH_3CNH); EI-MS, *m/z* (%): 300 (M^+, 13), 244 (100), 172 (13), 146 (14), 57 (7); Anal. Calcd for $C_{18}H_{24}N_2O_2$ (300.40): C, 71.97; H, 8.05; N, 9.33. Found: C, 71.78; H, 8.16; N, 9.43%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(2,6-dimethylphenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione 5l. Colorless crystals, yield 50%; mp 182–184 °C; $[\alpha]_D^{25}$ +24.51 (*c* 0.50, methanol); IR (KBr) 3297, 3182, 3054, 2957, 2871, 1682, 1635, 1599, 1578, 1500, 1469, 1390, 1199 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.85 (d, 3H, *J*=6.8, CH_3CH), 0.98 (t, 3H, *J*=6.99, CH_3CH_2), 1.27–1.51 (m, 2H, CH_3CH_2), 1.96–2.10 (m, 1H, CH_3CH), 2.22 (s, 6H, 2×$ArCH_3$), 2.24, 2.26 (s, s, 51/49, 3H, CH_3CNH), 3.74–3.95 (m, 1H, $CHNH$), 5.44 (br, 1H, NH), 7.16–7.24 (m, 3H, ArH), 11.59, 12.06 (s, s, 51/49, 1H, CH_3CNH); EI-MS, *m/z* (%): 300 (M^+, 7), 244 (100), 172 (17), 146 (22), 105 (10); Anal. Calcd for $C_{18}H_{24}N_2O_2$ (300.40): C, 71.97; H, 8.05; N, 9.33. Found: C, 72.09; H, 8.01; N, 9.45%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(2-methyl-6-ethylphenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-di one 5m. White powder, yield 51%; mp 161–162 °C; $[\alpha]_D^{25}$ +17.91 (*c* 0.50, methanol); IR (KBr) 3295, 3186, 3054, 2969, 2873, 1683, 1634, 1598, 1577, 1497, 1642, 1389, 1194 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.84 (d, 3H, *J*=6.6, CH_3CH), 1.03 (t, 3H, *J*=6.9, CH_3CH_2), 1.18 (t, 3H, *J*=7.5, CH_3CH_2Ar), 1.27–1.52 (m, 2H, CH_3CH_2CH), 1.96–2.08 (m, 1H, CH_3CH), 2.21 (s, 3H, $ArCH_3$), 2.25, 2.27 (s, s, 52/48, 3H, CH_3CNH), 2.50 (q, 2H, *J*=7.5, $ArCH_2$), 3.74–3.93 (m, 1H, $CHNH$), 6.11 (br, 1H, NH), 7.14–7.25 (m, 3H, ArH), 11.65, 12.09 (s, s, 52/48, 1H, CH_3CNH); EI-MS, *m/z* (%): 314 (M^+, 4), 258 (100), 230 (6), 160 (19), 91 (8); Anal. Calcd for $C_{19}H_{26}N_2O_2$ (314.42): C, 72.58; H, 8.33; N, 8.91; Found: C, 72.36; H, 8.15; N, 8.78%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(4-methoxylphenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione 5n. White powder, yield 34%; mp 99–101 °C; $[\alpha]_D^{25}$ +14.80 (*c* 0.50, methanol); IR (KBr) 3277, 3200, 3055, 2961, 2074, 1683, 1640, 1597, 1577, 1520, 1493, 1388, 1248, 1030 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.83 (d, 3H, *J*=6.6, C*H*$_3$CH), 1.02 (t, 3H, *J*=7.2, C*H*$_3$CH$_2$), 1.24–1.52 (m, 2H, CH$_3$C*H*$_2$), 1.93–2.10 (m, 1H, CH$_3$C*H*), 2.49, 2.52 (s, s, 53/47, 3H, C*H*$_3$CNH), 3.72–3.90 (m, 4H, C*H*NH + OCH$_3$), 5.61 (br, 1H, NH), 6.94–7.13 (m, 4H, ArH), 11.95, 12.40 (s, s, 53/47, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 302 (M^+, 21), 246 (100), 218 (8), 188 (11), 161 (13), 148 (18); Anal. Calcd for $C_{17}H_{22}N_2O_3$ (302.37): C, 67.53; H, 7.33; N, 9.26. Found: C, 67.68; H, 7.27; N, 9.39%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(2-ethoxylphenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione 5o. White powder, yield 57%; mp 85–87 °C; $[\alpha]_D^{25}$ –7.50 (*c* 0.50, methanol); IR (KBr) 3295, 3208, 3067, 2962, 2875, 1682, 1643, 1608, 1577, 1513, 1474, 1391, 1122 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.83 (d, 3H, *J*=6.6, C*H*$_3$CH), 1.02 (t, 3H, *J*=6.9, C*H*$_3$CH$_2$), 1.20–1.44 (m, 5H, CH$_3$C*H*$_2$ + OCH$_2$C*H*$_3$), 1.95–2.08 (m, 1H, CH$_3$C*H*), 2.53, 2.54 (s, s, 54/46, 3H, C*H*$_3$CNH), 3.73–3.91 (m, 1H, C*H*NH), 4.10 (q, 2H, *J*=6.7, OC*H*$_2$CH$_3$), 5.72 (br, 1H, NH), 6.96–7.31 (m, 4H, ArH), 11.95, 12.34 (s, s, 54/46, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 316 (M^+, 15), 260 (100), 175 (9), 134 (10), 57 (7); Anal. Calcd for $C_{18}H_{24}N_2O_3$ (316.39): C, 68.33; H, 7.65; N, 8.85. Found: C, 68.48; H, 7.57; N, 8.99%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(4-hydroxyphenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione 5p. Grey powder, yield 72%; mp 154–156 °C; $[\alpha]_D^{25}$ +22.45 (*c* 0.50, methanol); IR (KBr) 3281, 3203, 3063, 2964, 2876, 1674, 1634, 1594, 1575, 1519, 1493, 1385, 1221 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.82 (d, 3H, *J*=6.5, C*H*$_3$CH), 1.02 (t, 3H, *J*=6.9, C*H*$_3$CH$_2$), 1.25–1.50 (m, 2H, CH$_3$C*H*$_2$), 1.95–2.09 (m, 1H, CH$_3$C*H*), 2.50, 2.51 (s, s, 50/50, 3H, C*H*$_3$CNH), 3.76–3.92 (m, 1H, C*H*NH), 5.72 (br, 1H, NH), 6.93–7.03 (m, 4H, ArH), 11.89, 12.37 (s, s, 50/50, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 288 (M^+, 15), 232 (100), 147 (12), 134 (25), 58 (12); Anal. Calcd for $C_{16}H_{20}N_2O_3$ (288.34): C, 66.65; H, 6.99; N, 9.72. Found: C, 66.83; H, 6.87; N, 9.68%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(4-fluorophenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione 5q. Colorless crystals, yield 69%; mp 91–93 °C; $[\alpha]_D^{25}$ +18.42 (*c* 0.50, methanol); IR (KBr) 3295, 3207, 3063, 2963, 2875, 1737, 1685, 1640, 1610, 1585, 1492, 1385, 1156 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.82 (d, 3H, *J*=6.6, C*H*$_3$CH), 1.01 (t, 3H, *J*=6.8, C*H*$_3$CH$_2$), 1.32–1.50 (m, 2H, CH$_3$C*H*$_2$), 1.94–2.05 (m, 1H, CH$_3$C*H*), 2.49, 2.52 (s, s, 56/44, 3H, C*H*$_3$CNH), 3.74–3.90 (m, 1H, C*H*NH), 5.97 (br, 1H, NH), 7.13–7.16 (m, 4H, ArH), 12.02, 12.42 (s, s, 56/44, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 290 (M^+, 10), 234 (100), 176 (13), 136 (27), 95 (13), 57 (7); Anal. Calcd for $C_{16}H_{19}FN_2O_2$ (290.33): C, 66.19; H, 6.60; N, 9.65. Found: C, 66.27; H, 6.51; N, 9.49%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(4-chlorophenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione 5r. Colorless crystals, yield 80%; mp 124–126 °C; $[\alpha]_D^{25}$ +16.28 (*c* 0.50, methanol); IR (KBr) 3295, 3182, 3058, 2964, 2874, 1738, 1692, 1630, 1601, 1570, 1480, 1385, 1090 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.82 (d, 3H, *J*=6.6, C*H*$_3$CH), 1.02 (t, 3H, *J*=7.2, C*H*$_3$CH$_2$), 1.25–1.52 (m, 2H, CH$_3$C*H*$_2$), 1.96–2.06 (m, 1H, CH$_3$C*H*), 2.53, 2.56 (s, s, 54/46, 3H, C*H*$_3$CNH), 3.73–3.92 (m, 1H, C*H*NH), 5.79 (br, 1H, NH), 7.13–7.44 (m, 4H, ArH), 12.09, 12.49 (s, s, 54/46, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 306 (M^+, 9), 250 (100), 192 (14), 152 (25), 123 (14), 67 (16): Anal. Calcd for

$C_{16}H_{19}ClN_2O_2$ (306.79): C, 62.64; H, 6.24; N, 9.13. Found: C, 62.78; H, 6.20; N, 9.13%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(2,4-dichlorophenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione 5s. Colorless crystals, yield 79%; mp 149–150 °C; $[\alpha]_D^{25}$ +21.37 (*c* 0.50, methanol); IR (KBr) 3287, 3181, 3066, 2965, 2877, 1685, 1600, 1596, 1559, 1508, 1463, 1383, 1104 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.84 (d, 3H, *J*=6.7, C*H*$_3$CH), 1.03 (t, 3H, *J*=6.7, C*H*$_3$CH$_2$), 1.24–1.52 (m, 2H, CH$_3$C*H*$_2$), 1.95–2.09 (m, 1H, CH$_3$C*H*), 2.47, 2.50 (s, s, 57/43, 3H, C*H*$_3$CNH), 3.75–3.94 (m, 1H, C*H*NH), 5.70 (br, 1H, NH), 7.19–7.55 (m, 3H, ArH), 12.03, 12.38 (s, s, 57/43, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 340 ([M-H]$^-$, 5), 284 (100), 186 (15), 123 (21), 57 (8); Anal. Calcd for $C_{16}H_{18}Cl_2N_2O_2$ (341.23): C, 56.32; H, 5.32; N, 8.21. Found: C, 56.47; H, 5.23; N, 8.11%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(3,4-dichlorophenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione 5t. Colorless crystals, yield 82%; mp 139–141 °C; $[\alpha]_D^{25}$ +18.89 (*c* 0.50, methanol); IR (KBr) 3290, 3182, 3070, 2961, 2874, 1747, 1654, 1616, 1589, 1560, 1506, 1469, 1387, 1133 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.82 (d, 3H, *J*=6.4, C*H*$_3$CH), 1.02 (t, 3H, *J*=6.9, C*H*$_3$CH$_2$), 1.22–1.46 (m, 2H, CH$_3$C*H*$_2$), 1.92–2.11 (m, 1H, CH$_3$C*H*), 2.56, 2.58 (s, s, 58/42, 3H, C*H*$_3$CNH), 3.76–3.96 (m, 1H, C*H*NH), 5.55 (br, 1H, NH), 7.05–7.56 (m, 3H, ArH), 12.12, 12.51 (s, s, 58/42, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 340 ([M-H]$^-$, 4), 284 (100), 226 (14), 186 (22), 123 (17), 57 (9); Anal. Calcd for $C_{16}H_{18}Cl_2N_2O_2$ (341.23): C, 56.32; H, 5.32; N, 8.21. Found: C, 56.19; H, 5.26; N, 8.38%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(3-bromophenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione 5u. Colorless crystals, yield 61%; mp 94–95 °C; $[\alpha]_D^{25}$ +16.27 (*c* 0.50, methanol); IR (KBr) 3380, 3217, 3061, 2961, 2874, 1685, 1643, 1611, 1583, 1507, 1469, 1383, 1224 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.83 (d, 3H, *J*=6.6, C*H*$_3$CH), 1.02 (t, 3H, *J*=7.2, C*H*$_3$CH$_2$), 1.20–1.49 (m, 2H, CH$_3$C*H*$_2$), 1.94–2.08 (m, 1H, CH$_3$C*H*), 2.56, 2.58 (s, s, 59/41, 3H, C*H*$_3$CNH), 3.74–3.94 (m, 1H, C*H*NH), 5.61 (br, 1H, NH), 7.14–7.53 (m, 4H, ArH), 12.13, 12.54 (s, s, 59/41, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 350 ([M-H]$^-$, 11), 294 (100), 234 (14), 130 (14), 67 (16); Anal. Calcd for $C_{16}H_{19}BrN_2O_2$ (351.24): C, 54.71; H, 5.45; N, 7.98;. Found: C, 54.98; H, 5.32; N, 7.88%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(4-nitrophenyl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione 5v. Yellow crystals, yield 67%; mp 155–156 °C; $[\alpha]_D^{25}$ +39.42 (*c* 0.50, methanol); IR (KBr) 3295, 3188, 3066, 2960, 2875. 1689, 1651, 1615, 1578, 1519, 1484, 1394, 1384, 1304 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.83 (d, 3H, *J*=6.6, C*H*$_3$CH), 1.03 (t, 3H, *J*=6.9, C*H*$_3$CH$_2$), 1.24–1.48 (m, 2H, CH$_3$C*H*$_2$), 1.89–2.06 (m, 1H, CH$_3$C*H*), 2.69, 2.71 (s, s, 53/47, 3H, C*H*$_3$CNH), 3.77–3.97 (m, 1H, C*H*NH), 5.68 (br, 1H, NH), 7.35–8.36 (m, 4H, ArH), 12.49, 12.78 (s, s, 53/47, 1H, CH$_3$CN*H*); EI-MS, *m/z* (%): 317 (M^+, 5), 261 (100), 163 (13), 123 (11), 86 (11); Anal. Calcd for $C_{16}H_{19}N_3O_4$ (317.35): C, 60.56; H, 6.03; N, 13.24. Found: C, 60.70; H, 6.12; N, 13.11%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-(1-naphthyl-α-ylamino)ethylidene-1*H*-pyrrolidine-2,4-dione 5w. Colorless crystals, yield 65%; mp 141–143 °C; $[\alpha]_D^{25}$ +17.81 (*c* 0.50, methanol); IR (KBr) 3277, 3178, 3057, 2959, 2875, 1658, 1631, 1607, 1576, 1498, 1641, 1387, 1271 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.90 (t, 3H, *J*=6.5, C*H*$_3$CH$_2$), 1.08 (d, 3H, *J*=6.9, C*H*$_3$CH), 1.24–1.55 (m, 2H, CH$_3$C*H*$_2$), 1.99–2.13 (m, 1H, CH$_3$C*H*), 2.46, 2.47 (s, s, 56/44, 3H, C*H*$_3$CNH), 3.79–4.00 (m, 1H,

C*H*NH), 5.88 (br, 1H, NH, exchangeable with D_2O), 7.35–7.95 (m, 7H, naphthyl-H), 12.36, 12.77 (s, s, 56/44, 1H, CH_3CN*H*, exchangeable with D_2O); EI-MS, *m/z* (%): 322 (M^+, 21), 266 (100), 180 (18), 168 (20), 127 (15), 67 (8), Anal. Calcd for $C_{20}H_{22}N_2O_2$ (322.40): C, 74.51; H, 6.88; N, 8.69. Found: C, 74.59; H, 6.76; N, 8.78%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(4-aminobiphenyl-4′-yl)amino]ethylidene-1*H*-pyrrolidine--2,4-dione 5x. Khaki powder, yield 40%; mp 200 °C–decomp.; $[\alpha]_D^{25}$ +62.24 (*c* 0.50, methanol); IR (KBr) 3433, 3340, 3214, 3032, 2960, 2873, 1683, 1638, 1594, 1575, 1486, 1421, 1385, 1215 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.84 (d, 3H, *J*=6.8, C*H*$_3$CH), 1.03 (t, 3H, *J*=7.2, C*H*$_3$CH$_2$), 1.24–1.47 (m, 2H, CH$_3$C*H*$_2$), 1.94–2.11 (m, 1H, CH$_3$C*H*), 2.59, 2.65 (s, s, 40/60, 3H, C*H*$_3$CNH), 3.73–3.92 (m, 1H, C*H*NH), 5.41 (br, 1H, NH), 6.79 (br, 2H, NH_2), 7.21–7.69 (m, 8H, ArH), 12.21, 12.61 (s, s, 40/60, 1H, CH_3CN*H*); EI-MS, *m/z* (%): 363 (M^+, 83), 307 (100), 222 (36), 153 (278), 104 (47); Anal. Calcd for $C_{22}H_{25}N_3O_2$ (363.45): C, 72.70; H, 6.93; N, 11.56. Found: C, 72.87; H, 6.85; N, 11.68%.

(5*RS*,6*S*)-5-*sec*-Butyl-3-[1-(thiazolyl-2-yl)amino]ethylidene-1*H*-pyrrolidine-2,4-dione 5y. Yellow crystals, yield 54%; mp 133–134 °C; $[\alpha]_D^{25}$ +167.89 (*c* 0.50, methanol); IR (KBr) 3295, 3188, 3070, 2960, 2873, 1702, 1656, 1599, 1524, 1502, 1452, 1342, 1141 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ 0.82 (t, 3H, *J*=6.6, C*H*$_3$CH$_2$), 1.00 (t, 3H, *J*=7.5, C*H*$_3$CH), 1.31–1.53 (m, 2H, CH$_3$C*H*$_2$), 2.00–2.25 (m, 1H, CH$_3$C*H*), 2.97, 2.99 (s, s, 57/43, 3H, C*H*$_3$CNH), 3.78–3.96 (m, 1H, C*H*NH), 6.07 (br, 1H, NH), 7.11–7.60 (m, 2H, thiazolyl-H), 13.12, 13.32 (s, s, 57/43, 1H, CH_3CN*H*); EI-MS, *m/z* (%): 279 (M^+, 16), 223 (100), 193 (18), 138 (37), 86 (21), 58 (15); Anal. Calcd for $C_{13}H_{17}N_3O_2S$ (279.36): C, 55.89; H, 6.13; N, 15.04. Found: C, 55.95; H, 6.19; N, 15.18%.

X-Ray structure determination of compound 5l

A suitable single crystal of the compound **5l**, grown from ethanol and ethyl acetate mixture (1:1, v/v) at room temperature, was selected for X-ray diffraction analysis. The intensity data were recorded on a Bruker Smart APEX II CCD diffractometer equipped with graphite monochromatized Mo *Kα* radiation (λ = 0.71073 Å) at 291(2) K. The intensities were corrected for Lorentz and polarization effects, and all data were corrected using the SADABS program.[27] The crystal structure was solved by direct methods using the SHELXS-97 program.[28] All the non-hydrogen atoms were refined by full-matrix least-squares technique on F^2 with anisotropic thermal parameters. The hydrogen atoms were positioned geometrically and refined using a riding model. Crystal data and structure refinement parameters are summarized in Table 4.

The crystallographic data reported in this paper have been deposited with the Cambridge Crystallographic Data Centre as Supplementary Publication No. CCDC 731832. A copy of this information may be obtained free of charge from the Director, CCDC, 12 Union Road, Cambridge, CB2 1EZ, UK (fax: +44 1223 336033; e-mail: deposit@ccdc.cam.ac.uk or http://www.ccdc.cam.ac.uk/deposit).

Biological assays

Herbicidal tests. The compounds to be tested were dissolved in DMF at a concentration of 1.0 g L^{-1}, then diluted to the required test concentration with distilled water containing 0.1% TW-80. The biological tests were carried out in 9 cm Petri plates, each plate contained a piece of filter paper on the bottom, 10 pre-germinated seeds (1.0–0.5 mm) of rape or barnyard grass, and 5 mL of the solution (100 mg L^{-1}). Then the plates were kept at 25 ± 1 °C, with a photo-period of 12:12 (L:D) h for 2 days (rape) and 3 days (barnyard grass), respectively. In a control experiment, 5 mL of the same solution without the tested compounds was applied on each plate. Three replicates were conducted for each treatment and respective control. The lengths of root and stem were measured to calculate the means and the percentage of growth inhibition.

Antifungal tests. The antifungal activities were evaluated against five kinds of pathogenic fungi namely *F. graminearum, R. cerealis, C. capsici, B. cinerea,* and *F. moniliforme,* using a mycelia growth inhibition technique according to the literature.[29] The compounds to be tested were dissolved in 1 mL methanol, respectively, then added to 100 mL potato sucrose agar (PSA) medium and mixed uniformly to give the final concentration of 100 mg L^{-1}. The medium was poured into three 9 cm Petri plates under aseptic conditions in a laminar flow hood. Methanol (1 mL) in 100 mL PSA medium was used in the control experiment. After solidification, the mycelial disks (0.5 cm diameter) cut from the culture medium were transferred aseptically to the centers of treatment plates with a sterilized inoculation needle. Then the treated plates were incubated at 25 ± 1 °C in the dark. The diameters of the fungal colonies were measured till the fungal growth had covered two-thirds of the plates in the control treatments to calculate the percentage of mycelial growth inhibition.

Table 4. Crystal data and structure refinement for compound **5l**

Empirical formula	$C_{18}H_{24}N_2O_2$
Formula weight	300.39
Crystal size (mm^3)	0.26×0.22×0.20
Temperature (K)	291(2)
Color, Shape	Colorless, Block
Crystal system, space group	Monoclinic, *C*2
a (Å)	23.332(3)
b (Å)	12.3665(14)
c (Å)	17.806(2)
β (°)	136.2650(10)
Volume ($Å^3$)	3551.8(7)
Z	8
D_{calc} (g cm^{-3})	1.124
Absorption coefficient (mm^{-1})	0.073

Table 4. (continued)

F (000)	1296
Theta range for data collection (°)	1.65–27.51
Index ranges	–28≤h≤30, –16≤k≤15, –23≤l≤23
Reflections collected/unique	15546/4254 [$R_{(int)}$ = 0.0390]
Completeness to theta = 27.51°	99.7%
Data/restraints/parameters	4254/1/407
Goodness-of-fit on F^2	1.070
Final R indices [I > 2σ(I)]	R_1 = 0.0626, wR_2 = 0.1465
R indices (all data)	R_1 = 0.0795, wR_2 = 0.1513
Largest diff. peak and hole (e $Å^{-3}$)	0.292 and –0.529

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