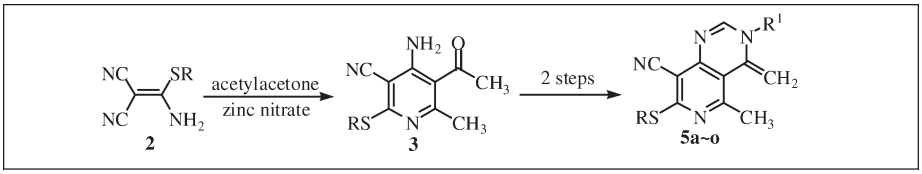
Synthesis and Biological Activity of 4-Methylene-pyrido[4,3-d]pyrimidines

Wenyan Mo,[a] Yongyan Yao,[a] Yunli Shen,[a] Hongwu He,[a]* and Yucheng Gu[a,b]

[a]Key Laboratory of Pesticide & Chemical Biology, Ministry of Education, Institute of Pesticide Chemistry, Central China Normal University, Wuhan, People's Republic of China
[b]Syngenta, Jealott's Hill International Research Centre, Bracknell, Berkshire, United Kingdom
*E-mail: he1208@mail.ccnu.edu.cn

Received January 8, 2009
DOI 10.1002/jhet.124
Published online 25 June 2009 in Wiley InterScience (www.interscience.wiley.com).



Fifteen novel 3-substituted-5-methyl-4-methylene-7-alkylsulfanyl-3,4-dihydro-pyrido[4,3-d]pyrimidine-8-carbonitriles **5a–o**, were synthesized *via* a facile annulation process in which formation of the pyrimidine ring proceeded smoothly by the regioselective attack of a formamidate group on a neighboring carbonyl group instead of a cyano group. Bioassay results indicated that these compounds showed significant herbicidal activity at a dose of 100 μg/mL on the roots of oil rape and barnyard grass. In addition, some of these compounds displayed fungicidal activity.

J. Heterocyclic Chem., **46**, 579 (2009).

INTRODUCTION

Reports in the literature on the biological activity of pyrido[4,3-d]pyrimidines relate mainly to their pharmaceutical activity as EGFR-TK and DHFR inhibitors [1]. Very little has been reported about their biological activities relating to agriculture [2].

Generally, pyrido[d]pyrimidines have been prepared by routes in which the pyrimidine ring is formed by cyclization of suitable substituents on a pyridine [3]. For example, it has been reported that formamidate reacts with a cyano group in the neighboring position to afford a pyrimidine ring [4]. However, there are no reports about formamidate regioselectively attacking a neighboring carbonyl group instead of a cyano group.

In our previous work, we have synthesized various pyrido[4,3–d]pyrimidine derivatives possessing fungicidal and herbicidal activity [5]. As a progression to this research, we herein describe a highly efficient regioselective cyclization reaction leading to the synthesis of pyrido[4,3–d]pyrimidines under mild conditions and in short reaction times. A preliminary *in vitro* bioassay indicated that these 4-methylene-pyrido[4,3–d]pyrimidines have strong herbicidal activity and that some have fungicidal activity as well.

RESULTS AND DISCUSSION

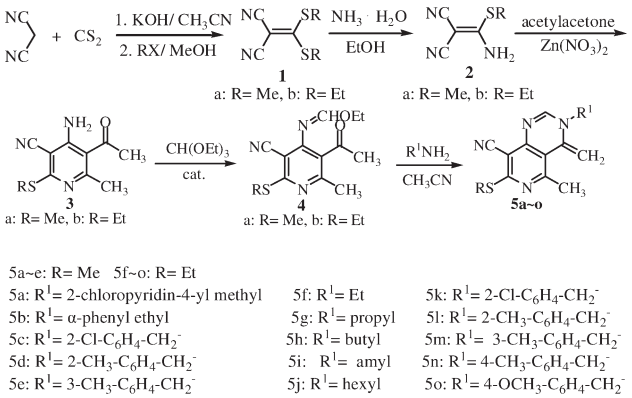
The conventional synthetic route to pyrido[4,3–d]pyrimidines involves formation of the pyrimidine ring by closure of suitable substituents on a pyridine. It was noted that the synthesis of the substituted pyridines, or the pyrimidine ring closure, always suffered from certain drawbacks, such as low yield, long reaction time, or difficulty in isolating or purifying the products [3,4]. In the protocol described here, formation of the pyrimidine ring proceeded regioselectively and with high efficiency (see Scheme 1).

The intermediates **1** and **2** can be prepared according to a published method [6]. It was reported that 4-amino nicotinonitrile 3 was prepared in moderate yield (48%) by using anhydrous stannic chloride as catalyst [7]. We improved the conditions by using zinc nitrate as catalyst to achieve much higher yield than the reported method (over 83%) [8]. By optimizing the reaction conditions, formamidate **4a** was obtained with the yield of 87% yield in the presence of *p*-toluene sulfonic acid. By comparison, only 32% yield was obtained when no catalyst was used, and formamidate **4b** was obtained with 82% yield in the presence of acetic anhydride as the catalyst.

The formamidate **4** reacted with primary amines smoothly and regioselectively in acetonitrile at 40∼50°C to give the title compounds **5a–o** in moderate to excellent yields. The products were easily isolated by filtration. The structures of compounds **5a–o** were fully elucidated by a comprehensive analysis of their IR, MS, and ^{1}H NMR spectra and by elemental analysis.

The compounds **5a–o** were screened for activity against six fungi, namely *Fusarium oxysporium, Rhizoctonia solani, Botrytis cinerea, Gibberella zeae, Dothiorella gregaria*, and *Colletotrichum gossypii*, at a

Scheme 1



concentration of 50 μg/mL according to a reported method [5]. As the results in Table 1 show, most of the compounds have weak fungicidal activity. Among these compounds, only compound **5a,** in which R^1 is (2-chloropyridin-4-yl)methyl group, exhibited moderate inhibitory activity against each of the six fungi. This might imply that the introduction of pyridine ring to the position 3 of pyrimidines was important for its fungicidal activity. All the remaining compounds, in which the substituent R^1 is alkyl or substituted benzyl, showed only weak antifungal activity.

The herbicidal activity of compounds **5a**–**o** was also evaluated, against two representative targets, oil rape and barnyard grass, at concentrations of 100 and 10 μg/mL, according to a literature method [9]. The results are listed in Table 2 and show that these compounds have moderate to good herbicidal activity against the roots of these two species at the rate of 100 μg/mL, especially against the root of barnyard grass. Compound **5a** with (2-chloropyridin-4-yl)methyl moiety in position 3 of the pyrimidine ring showed much better activity than the rest of the series with over 85% inhibition even at the lower concentration of 10 μg/mL. Switching the substituent R from methyl to ethyl has no obvious effect on the inhibition rates. In terms of R^1, the substituents with electron-donating groups on the phenyl rings seem to have somewhat higher herbicidal activity. For example, compounds **5d** and **5e** showed better activity than compound **5c**, and compounds **5l–o** were better than compound **5k.**

EXPERIMENTAL

All chemicals were of reagent grade and were commercially available. Solvents were either used directly or were purified as required. *Fusarium oxysporium, Rhizoctonia solani, Botrytis cinerea, Gibberella zeae, Dothiorella gregaria, Colletotrichum gossypii*, and seeds of oil rape and barnyard grass were provided through the courtesy of the Bioassay Center, Central China Normal University.

Melting points were measured on a WRS-1B Digital melting point apparatus. ^{1}H NMR spectra were recorded at 400 MHz in $CDCl_3$ solution on a Varian VNMR-400 Spectrometer, whereas IR spectra were recorded on a Nicolet AVATAR-360 Infrared Spectrometer. The MS spectra were determined using a Finnigan Trace MS Spectrometer, and the signals were given in *m*/*z*. Elementary analysis was carried out on a Vario EL III CHNSO elemental analyzer.

Intermediates **1** and **2** were prepared according to the literature [6].

5-Acetyl-4-amino-6-methyl-2-alkylsulfanyl-nicotinonitrile (3). Acetylacetone (1.0 g, 10 mmol) and **2** (10 mmol) were added to a stirred solution of zinc nitrate in 30 mL alcohol. The mixture was stirred and refluxed for 12 h. The precipitate

Table 1

Antifungal activity of compounds **5a–o** (% inhiblition).

			50 μg/mL					
Compounds	R	R^1	*F. oxysporium*	*R. solani*	*B. cinerea*	*G. zeae*	*D. gregaria*	*C. gossypii*
5a	Me	2-Chloropyridin-4-yl-methyl	36	52	81	43	60	54
5b	Me	α-Phenyl ethyl	42	33	48	56	14	26
5c	Me	2—Cl—C_6H_4—CH_2—	12	3	12	9	20	14
5d	Me	2—CH_3—C_6H_4—CH_2—	8	0	19	3	26	14
5e	Me	3—CH_3—C_6H_4—CH_2—	0	0	19	11	26	11
5f	Et	Ethyl	50	32	17	6	21	31
5g	Et	Propyl	35	9	14	19	21	27
5h	Et	Butyl	15	7	9	19	10	23
5i	Et	Amyl	19	0	23	16	7	27
5j	Et	Hexyl	15	4	20	9	10	15
5k	Et	2—Cl—C_6H_4—CH_2—	4	7	11	6	0	19
5l	Et	2—CH_3—C_6H_4—CH_2—	4	2	23	9	0	15
5m	Et	3—CH_3—C_6H_4—CH_2—	4	0	17	3	3	4
5n	Et	4—CH_3—C_6H_4—CH_2—	15	0	0	9	0	19
5o	Et	4—OCH_3—C_6H_4—CH_2—	19	4	0	3	0	19

Table 2

Herbicidal activity of compounds **5a-o** (% inhibition).

Compds	R	R^1	Oil rape (root/stalk)		Barnyard grass (root/stalk)	
			100 μg/mL	10 μg/mL	100 μg/mL	10 μg/mL
5a	Me	2-Chloropyridin-4-yl-methyl	96/63	91/37	97/81	87/69
5b	Me	α-Phenylethyl	68/26	37/21	72/56	67/50
5c	Me	2—Cl—C_6H_4—CH_2—	62/5	13/0	77/67	62/56
5d	Me	2—CH_3—C_6H_4—CH_2	68/16	18/11	89/77	64/56
5e	Me	3—CH_3—C_6H_4—CH_2	82/21	26/16	87/50	64/60
5f	Et	Ethyl	45/35	54/18	69/59	8/0
5g	Et	Propyl	46/41	17/6	42/41	28/41
5h	Et	Butyl	63/29	25/18	53/48	0/0
5i	Et	Amyl	43/35	32/24	92/76	19/17
5j	Et	Hexyl	71/29	45/24	89/72	39/35
5k	Et	2—Cl—C_6H_4—CH_2—	51/35	17/24	77/59	58/52
5l	Et	2—CH_3—C_6H_4—CH_2—	64/35	32/29	78/62	67/59
5m	Et	3—CH_3—C_6H_4—CH_2—	81/47	26/29	94/72	64/55
5n	Et	4—CH_3—C_6H_4—CH_2—	61/35	39/18	92/76	68/57
5o	Et	4—OCH_3—C_6H_4—CH_2—	86/41	36/35	88/66	64/48

was filtered off, washed with water (10 mL × 3), and recrystalized from alcohol to give **3**.

5-acetyl-4-amino-6-methyl-2-methylsulfanyl-nicotinonitrile (3a). White solid, m.p. 163.5–164.5°C, yield 83%. ^{1}H NMR ($CDCl_3$): δ 2.58 (s, 3H, CH_3), 2.61 (s, 3H, SCH_3), 2.68 (s, 3H, $COCH_3$), 6.60 ppm (br, 2H, NH_2).

5-acetyl-4-amino-6-methyl-2-ethylsulfanyl-nicotinonitrile (3b). Yellowish solid, m.p. 139.0–140.0°C, yield 89%. ^{1}H NMR ($CDCl_3$): δ 1.38 (t, 3H, SCH_2CH_3, J = 7.4 Hz), 2.59 (s, 3H, CH_3), 2.70 (s, 3H, $COCH_3$), 3.15 (q, 2H, SCH_2CH_3, J = 7.4 Hz), 6.68 ppm (br, 2H, NH_2).

***N*-(3-Acetyl-5-cyano-2-methyl-6-alkylsulfanyl-pyridin-4-yl)-formimidic acid ethyl ester (4).** To a solution of **4**-aminopyridine **3** (10 mmol) in triethyl orthoformate (5.92 g, 40 mmol) was added acetic anhydride or *p*-toluene sulfonic acid as catalyst. The mixture was heated and monitored by TLC. The triethyl orthoformate was removed at reduced pressure, and the residue was purified by silica gel chromatography (ether:petroleum ether=1:10) to afford formamidate **4**.

***N*-(3-acetyl-5-cyano-2-methyl-6-methylsulfanyl-pyridin-4-yl)-formimidic acid ethyl ester (4a).** White solid, m.p. 78.5–79.9°C, yield 95%. ^{1}H NMR: δ 1.40 (t, 3H, CH_2CH_3, J = 8.0 Hz), 2.46 (s, 3 H, CH_3), 2.56 (s, 3H, SCH_3), 2.62 (s, 3H, $COCH_3$), 4.39 (q, 2H, CH_2CH_3, J = 8.0 Hz), 8.06 ppm (s, 1H, =CH). ms (70 Ev): *m*/*z* 277 (M^+, 63).

***N*-(3-acetyl-5-cyano-2-methyl-6-ethylsulfanyl-pyridin-4-yl)-formimidic acid ethyl ester (4b).** Yellowish liquid, yield 82%. ^{1}H NMR: δ 1.38 (t, 3H, SCH_2CH_3, J = 3.8 Hz), 1.40 (t, 3H, CH_2CH_3, J = 3.8 Hz), 2.45 (s, 3H, CH_3), 2.46 (s, 3H, $COCH_3$), 3.27 (q, 2H, SCH_2CH_3, J = 3.6 Hz), 4.27 (q, 2H, CH_2CH_3, J = 3.6 Hz), 7.65 ppm (s, 1H, N=CH).

General procedure for the preparation of compounds (5a–m). To a solution of formamidate **4** (10 mmol) in anhydrous acetonitrile (10 mL) was added the appropriate amine (15 mmol), and the mixture was stirred and heated at 40–50°C for 30 min. The precipitate was isolated by filtration, then recrystalized from acetone/petroleum ether to give pure products **5**.

3-[(2-Chloropyridin-4-yl)methyl]-5-methyl-4-methylene-7-methylsulfanyl-3,4-dihydropyrido[4,3–d]pyrimidine-8-carbonitrile (5a). This compound was obtained as a pale yellow solid, m.p. 167.9–168.2°C, yield 59%. IR (KBr): ν 3036, 2931, 2218, 1610, 1550, 1523, 1390, 1278 cm^{-1}; ^{1}H NMR ($CDCl_3$): δ 2.61 (s, 3H, CH_3), 2.64 (s, 3H, SCH_3), 4.67 (d, 2H, $=CH_2$), 4.83 (s, 2H, CH_2), 7.20-7.64 (m, 3H, pyridine-H), 7.86 ppm (s, 1H, pyrimidine-H); ms: *m*/*z* 357 (M^++1 20). *Anal*. Calcd. for $C_{17}H_{14}ClN_5S$ (356): C, 57.38; H, 3.97; N, 19.68. Found C, 57.82; H, 3.44; N, 19.71.

3-(α-Phenylethyl)-5-methyl-4-methylene-7-methylsulfanyl-3,4-dihydropyrido[4,3–d]pyrimidine-8-carbonitrile (5b). This compound was obtained as a white solid, m.p. 147.4–148.5°C, yield 62%. IR (KBr): ν 3170, 2978, 2925, 2211, 1621, 1563, 1514, 1399, 1294 cm^{-1}; ^{1}H NMR ($CDCl_3$): δ 1.63 (d, 3H, Ph—CH_3, J = 4.0 Hz), 2.45 (s, 3H, CH_3), 2.60 (s, 3H, SCH_3), 4.65 (d, 2H, $=CH_2$), 6.41 (d, 1H, Ph—CH, J = 4.0 Hz), 6.27-7.43 (m, 5H, ArH), 7.75 ppm (s, 1H, pyrimidine-H); ms: *m*/*z* 334 (M^+ 8). *Anal*. Calcd. for $C_{19}H_{18}N_4S$ (334): C, 68.23; H, 5.42; N, 16.75. Found C, 68.29; H, 5.44;, N, 16.51.

3-(2-Chlorophenyl)-5-methyl-4-methylene-7-methyl sulfanyl-3,4-dihydropyrido[4,3–d]pyrimidine-8-carbonitrile (5c). This compound was obtained as a white solid, m.p. 143.2–144.5°C, yield 63%. IR (KBr): ν 3148, 2927, 2215, 1601, 1548, 1524, 1396, 1280 cm^{-1}; ^{1}H NMR ($CDCl_3$): δ 2.61 (s, 3H, CH_3), 2.66 (s, 3H, SCH_3), 4.46 (d, 1H, $=CH^a$, J = 2.0 Hz), 4.64 (d, 1H, $=CH^b$, J = 2.0 Hz), 4.86 (s, 2H, CH_2), 7.21-7.47 (m, 4H, ArH), 7.55 ppm (s, 1H, pyrimidine-H); ms: *m*/*z* 357 (M^++2 7), 355(M^+ 24). *Anal*. Calcd. for $C_{18}H_{15}ClN_4S$ (355): C, 60.92; H, 4.26; N, 15.79. Found C, 60.72; H, 4.44; N, 15.71.

3-(2-Methylphenyl)-5-methyl-4-methylene-7-methyl sulfanyl-3,4-dihydro-pyrido[4,3–d]pyrimidine-8-carbonitrile (5d). This compound was obtained as a white solid, m.p. 184.2–184.8°C, yield 75%. IR (KBr): ν 3001, 2926, 2218, 1604, 1553, 1521, 1399, 1278 cm^{-1}; ^{1}H NMR ($CDCl_3$): δ 2.32 (s, 3H, Ph—CH_3), 2.62 (s, 3H, CH_3), 2.69 (s, 3H, SCH_3), 4.50 (d, 1H, $=CH^a$, J = 2.0 Hz), 4.65 (d, 1H, $=CH^b$, J = 2.0 Hz), 4.69 (s, 2H, —CH_2—), 7.16-7.28 (m, 4H, ArH), 7.44 ppm (s, 1H,

pyrimidine-H); ms: *m/z* 335(M^++1 9), 334(M^+ 20). *Anal.* Calcd. for $C_{19}H_{18}N_4S$ (334): C, 68.23; H, 5.42; N, 16.75. Found C, 68.72; H, 5.44; N, 16.71.

3-(3-Methylphenyl)-5-methyl-4-methylene-7-methyl sulfanyl-3,4-dihydro-pyrido[4,3–d]pyrimidine-8-carbonitrile (5e). This compound was obtained as a white solid, m.p. 162.1–163.6°C, yield 90%. IR (KBr): ν 3074, 2926, 2219, 1607, 1555, 1522, 1401, 1262 cm^{-1}; ^1H NMR ($CDCl_3$): δ 2.34 (s, 3H, Ph—CH_3), 2.62 (s, 3H, CH_3), 2.64 (s, H, SCH_3), 4.54 (d, 1H, =CH^a, *J* = 2.0 Hz), 4.62 (d, 1H, =CH^b, *J* = 2.0 Hz), 4.74(s,2H, —CH_2—), 7.06–7.28 (m, 4H, ArH), 7.58 ppm (s, 1H, pyrimidine-H); ms: *m/z* 334(M^+ 43). *Anal.* Calcd. for $C_{19}H_{18}N_4S$ (334): C, 68.23; H, 5.42; N, 16.75. Found C, 68.39; H, 5.44; N, 16.73.

3-Ethyl-5-methyl-4-methylene-7-ethylsulfanyl-3,4-dihydro-pyrido[4,3–d]pyrimidine-8-carbonitrile (5f). This compound was obtained as a white solid, m.p. 144.1–145.0°C, yield 63%. IR (KBr): ν 3170, 2975, 2931, 2221, 1615, 1547, 1523, 1399 cm^{-1}; ^1H NMR ($CDCl_3$): δ 1.36–1.41 (m, 6H, CH_3 in —CH_2CH_3 and —SCH_2CH_3), 2.69 (s, 3H, —CH_3), 3.24 (q, 2H, —SCH_2CH_3, *J* = 7.3 Hz), 3.69 (q, 2H, —CH_2CH_3, *J* = 7.2 Hz), 4.64 (d, 1H, =CH^a, *J* = 3.2 Hz), 4.65 (d, 1H, =CH^b, *J* = 3.2 Hz), 7.52 ppm (s, 1H, pyrimidine-H); ms: *m/z* 273(M^++1 3), 272(M^+ 3). *Anal.* Calcd. for $C_{14}H_{16}N_4S$ (272) : C, 61.74; H, 5.92; N, 20.57. Found C, 61.72; H, 5.72; N, 20.30.

3-Propyl-5-methyl-4-methylene-7-ethylsulfanyl-3,4-dihydro-pyrido[4,3–d]pyrimidine-8-carbonitrile (5g). This compound was obtained as a white solid, m.p. 148.8–150.4°C, yield 61%. IR (KBr): ν 3180, 2966, 2875, 2221, 1613, 1548, 1521, 1388 cm^{-1}; ^1H NMR ($CDCl_3$): δ 0.99 (t, 3H, —$CH_2CH_2CH_3$, *J* = 7.4 Hz), 1.38 (t, 3H, —SCH_2CH_3, *J* = 8.0 Hz), 1.75^{-1}.81 (m, 2H, —$CH_2CH_2CH_3$), 2.69 (s, 3H, —CH_3), 3.25 (q, 2H, —SCH_2CH_3, *J* = 7.3 Hz), 3.58 (t, 2H, —$CH_2CH_2CH_3$, *J* = 7.2 Hz), 4.63 (d, 1H, =CH^a, *J* = 3.2 Hz), 4.65 (d, 1H, =CH^b, *J* = 3.2 Hz), 7.47 ppm (s, 1H, pyrimidine-H); ms: *m/z* 286(M^+ 64). *Anal.* Calcd. for $C_{15}H_{18}N_4S$ (286) : C, 62.91; H, 6.33; N, 19.56. Found C, 62.88; H, 6.40; N, 19.61.

3-Butyl-5-methyl-4-methylene-7-ethylsulfanyl-3,4-dihydro pyrido[4,3–d]pyrimidine-8-carbonitrile (5h). This compound was obtained as a white solid, m.p. 144.6–145.1°C, yield 59%. IR (KBr): ν 3047, 2959, 2871, 2218, 1597, 1550, 1517, 1388 cm^{-1}; ^1H NMR ($CDCl_3$): δ 0.96^{-1}.00 (t, 3H, —$CH_2CH_2CH_2CH_3$, *J* = 7.2 Hz), 1.36–1.44 (m, 5H, —$CH_2CH_2CH_2CH_3$ and —SCH_2CH_3), 1.69–1.76 (m, 2H, —$CH_2CH_2CH_2CH_3$), 2.69 (s, 3H, —CH_3), 3.25 (q, 2H, —SCH_2CH_3, *J* = 4.9 Hz), 3.62 (t, 2H, —$CH_2CH_2CH_2CH_3$, *J* = 7.0 Hz), 4.62 (d, 1H, =CH^a, *J* = 3.2 Hz), 4.64 (d, 1H, =CH^b, *J* = 3.2 Hz), 7.47 ppm (s, 1H, pyrimidine-H); ms: *m/z* 301(M^++H 31), 300(M^+ 67). *Anal.* Calcd. for $C_{16}H_{20}N_4S$ (300): C, 63.97; H, 6.71; N, 18.65. Found C, 64.02; H, 6.84; N, 18.43.

3-Amyl-5-methyl-4-methylene-7-ethylsulfanyl-3,4-dihydro pyrido[4,3–d]pyrimidine-8-carbonitrile (5i). This compound was obtained as a white solid, m.p. 134.8–135.4°C, yield 56%. IR (KBr): ν 3163, 2957, 2868, 2211, 1613, 1548, 1522, 1395cm^{-1}; ^1H NMR ($CDCl_3$): δ 0.93 (t, 3H, —$CH_2CH_2CH_2CH_2CH_3$, *J* = 6.8 Hz), 1.33–1.40 (m, 7H, —$CH_2CH_2CH_2CH_2CH_3$ and —SCH_2CH_3), 1.73–1.78 (m, 2H, —$CH_2CH_2CH_2CH_2CH_3$), 2.694 (s, 3H, —CH_3), 3.25 (q, 2H, —SCH_2CH_3, *J* = 7.4 Hz), 3.61 (t, 2H, —$CH_2CH_2CH_2CH_2CH_3$, *J* = 7.0 Hz), 4.62 (d, 1H, =CH^a, *J* = 3.2 Hz), 4.65 (d, 1H, =CH^b, *J* = 3.2 Hz), 7.47 ppm (s, 1H, pyrimidine-H); ms: *m/z* 315(M^++1 13), 314(M^+ 47), 313(M^+−H 14). *Anal.* Calcd. for $C_{17}H_{22}N_4S$ (314): C, 64.93; H, 7.05; N, 17.82. Found C, 64.59; H, 7.28; N, 17.66.

3-Hexyl-5-methyl-4-methylene-7-ethylsulfanyl-3,4-dihydro pyrido[4,3–d]pyrimidine-8-carbonitrile (5j). This compound was obtained as a white solid, m.p. 131.5–132.4°C, yield 79%. IR (KBr): ν 3174, 2950, 2865, 2222, 1615, 1547, 1523, 1397cm^{-1}; ^1H NMR ($CDCl_3$): δ 0.88–0.91 (t, 3H, —$CH_2CH_2CH_2CH_2CH_2CH_3$, *J* = 6.8 Hz), 1.33–1.40 (m, 9H, —$CH_2CH_2CH_2CH_2CH_2CH_3$ and —SCH_2CH_3), 1.72−1.77 (m, 2H, —$CH_2CH_2CH_2CH_2CH_2CH_3$), 2.69 (s, 3H, —CH_3), 3.25 (q, 2H, —SCH_2CH_3, *J* = 7.3 Hz), 3.61 (t, 2H, —$CH_2CH_2CH_2CH_2CH_3$, *J* = 7.2 Hz), 4.63 (d, 1H, =CH^a, *J* = 3.2 Hz), 4.65 (d, 1H, =CH^b, *J* = 3.2 Hz), 7.47 ppm (s, 1H, pyrimidine-H); ms: *m/z* 328(M^+ 41). *Anal.* Calcd. for $C_{18}H_{24}N_4S$ (328): C, 65.82; H, 7.36; N, 17.06. Found C, 65.53; H, 7.41; N, 16.99.

3-(2-Chlorobenzyl)-5-methyl-4-methylene-7-ethylsulfanyl-3,4-dihydropyrido[4,3–d]pyrimidine-8-carbonitrile (5k). This compound was obtained as a white solid, m.p. 148.5–149.5°C, yield 61%. IR (KBr): ν 3066, 2928, 2216, 1599, 1548, 1525, 1393 cm^{-1}; ^1H NMR ($CDCl_3$): δ 1.37–1.41 (t, 3H, —SCH_2CH_3, *J* = 7.4 Hz), 2.66 (s, 3H, —CH_3), 3.25 (q, 2H, —SCH_2CH_3, *J* = 7.3 Hz), 4.45 (d, 1H, =CH^a, *J* = 3.6 Hz), 4.63 (d, 1H, =CH^b, *J* = 3.2 Hz), 4.86 (s, 2H, —CH_2—), 7.21–7.47 (m, 4H, ArH), 7.55 ppm (s, 1H, pyrimidine-H); ms: *m/z* 370(M^++2 7), 368(M^+ 23). *Anal.* Calcd. for $C_{19}H_{17}ClN_4S$ (368): C, 61.86; H, 4.65; N, 15.19. Found C, 61.62; H, 4.74; N, 14.93.

3-(2-Methylbenzyl)-5-methyl-4-methylene-7-ethylsulfanyl-3,4-dihydro-pyrido[4,3–d]pyrimidine-8-carbonitrile (5l). This compound was obtained as a white solid, m.p. 153.9–154.9°C, yield 68%. IR (KBr): ν 3066, 2966, 2218, 1607, 1554, 1523, 1400 cm^{-1}; ^1H NMR ($CDCl_3$): δ 1.39 (t, 3H, —SCH_2CH_3, *J* = 7.6 Hz), 2.33 (s, 3H, Ph—CH_3), 2.68 (s, 3H, —CH_3), 3.24 (q, 2H, —SCH_2CH_3, *J* = 7.3 Hz), 4.49 (d, 1H, =CH^a, *J* = 3.6 Hz), 4.65 (d, 1H, =CH^b, *J* = 3.6 Hz), 4.69 (s, 2H, —CH_2—), 7.16–7.27 (m, 4H, ArH), 7.43 ppm (s, 1H, pyrimidine-H); ms: *m/z* 348(M^+ 32). *Anal.* Calcd. for $C_{20}H_{20}N_4S$ (348): C, 68.93; H, 5.79; N, 16.08. Found C, 69.06; H, 5.88; N, 15.94.

3-(3-Methylbenzyl)-5-methyl-4-methylene-7-ethylsulfanyl-3,4-dihydro-pyrido[4,3–d]pyrimidine-8-carbonitrile (5m). This compound was obtained as a white solid, m.p. 145.2–146.0°C, yield 80%. IR (KBr): ν 3001, 2967, 2926, 2217, 1606, 1550, 1524, 1401 cm^{-1}; ^1H NMR ($CDCl_3$): δ 1.38 (t, 3H, —SCH_2CH_3, *J* = 7.4 Hz), 2.35 (s, 3H, Ph—CH_3), 2.64 (s, 3H, —CH_3), 3.25 (q, 2H, —SCH_2CH_3, *J* = 7.3 Hz), 4.52 (d, 1H, =CH^a, *J* = 3.6 Hz), 4.62 (d, 1H, =CH^b, *J* = 3.6 Hz), 4.74 (s, 2H, —CH_2Ph), 7.06–7.27 (m, 4H, ArH), 7.57 ppm (s, 1H, pyrimidine-H); ms: *m/z* 348(M^+ 16). *Anal.* Calcd. for $C_{20}H_{20}N_4S$ (348): C, 68.93; H, 5.79; N, 16.08. Found C, 68.81; H, 5.86; N, 15.91.

3-(4-Methylbenzyl)-5-methyl-4-methylene-7-ethylsulfanyl-3,4-dihydro-pyrido[4,3–d]pyrimidine-8-carbonitrile (5n). This compound was obtained as a white solid, m.p. 163.5–164.5°C, yield 63%. IR (KBr): ν 3030, 2974, 2927, 2218, 1606, 1554, 1521, 1399 cm^{-1}; ^1H NMR ($CDCl_3$): δ 1.36–1.40 (t, 3H, —SCH_2CH_3, *J* = 7.2 Hz), 2.36 (s, 3H, Ph—CH_3), 2.63 (s, 3H, —CH_3), 3.25 (q, 2H, —SCH_2CH_3, *J* = 7.3 Hz), 4.53 (d, 1H, =CH^a, *J* = 3.6 Hz), 4.60 (d, 1H, =CH^b, *J* = 3.2 Hz), 4.74 (s, 2H, —CH_2Ph), 7.14-7.20 (m, 4H, ArH), 7.57 ppm (s, 1H, pyrimidine-H); ms: *m/z* 348(M^+ 5). *Anal.* Calcd. for $C_{20}H_{20}N_4S$

(348): C, 68.93; H, 5.79; N, 16.08. Found C, 68.84; H, 5.63; N, 15.90.

3-(4-Methoxybenzyl)-5-methyl-4-methylene-7-ethyl sulfanyl-3,4-dihydro-pyrido[4,3–d]pyrimidine-8-carbonitrile (5o). This compound was obtained as a white solid, m.p. 156.1–156.9°C, yield 82%. IR (KBr): ν 2934, 2220, 1612, 1547, 1515, 1404 cm^{-1}; ^1H NMR ($CDCl_3$): δ 1.38 (t, 3H, —SCH_2CH_3, J = 7.2 Hz), 2.63 (s, 3H, —CH_3), 3.24 (q, 2H, —SCH_2CH_3, J = 7.5 Hz), 3.81 (s, 3H, —OCH_3), 4.57 (d, 1H, =CH^a, J = 3.2 Hz), 4.61 (d, 1H, =CH^b, J = 3.2 Hz), 4.71 (s, 2H, —CH_2Ph), 6.90–7.20 (m, 4H, ArH) 7.54 ppm (s, 1H, pyrimidine-H); ms: *m/z* 364(M^+ 12). *Anal*. Calcd. for $C_{20}H_{20}N_4OS$ (364): C, 65.91; H, 5.53; N, 15.37. Found C, 65.84; H, 5.63; N, 15.21.

Acknowledgments. This work was supported by the National Basic Research Program of China (No. 2003CB114400), National Natural Science Foundation of China (No.20372023, 20772042) and Syngenta Ltd. We thank Dr. John Clough of Syngfenta Jealott's Hill International Research Centre for proof reading.

REFERENCES AND NOTES

[1] (a) Thompson, A. M.; Murray, D. K.; Elliott, W. L.; Fry, D. W.; Nelson, J. A.; Showalter, H. D. H.; Roberts, B. J.; Vincent, P. W.; Denny, W. A. J Med Chem 1997, 40, 3915; (b) Fry, D. W.; Nelson, J. A.; Slintak, V.; Keller, P. R.; Rewcastle, G. W.; Denny, W. A.; Zhou, H. R.; Bridges, A. J. Biochem Pharmacol 1997, 54, 877; (c) Smaill, J. B.; Palmer, D. B.; Rewcastle, G. W.; Denny, W. A.; McNamara, D. J.; Dobrusin, E. M.; Brodges, A. J.; Zhou, H. R.; Showalter, H. D. H.; Winters, R. T.; Fry, D. W.; Nelson, J. M.; Slintak, V.; Elliot, W. L.; Roberts, B. J.; Vincent, P. W.; Patmore, S. J. J Med Chem 1999, 42, 1803; (d) Elselarge, E. F.; Clarke, J.; Jacob, P. J. Heterocyclic Chem 1972, 9, 1113; (e) Rosowsky, A.; Bader, H.; Moran, R. G. J Heterocycl Chem 1989, 26, 509; (f) Rosowsky, A.; Mota, C. E.; Queener, S. F. J Heterocycl Chem 1995, 32, 335; (g) Hussein, E. S.; Suhair, M. A. Z.; Mona, A. M. J Med Chem 2000, 43, 2915.

[2] (a) Brighty, K. E.; Lowe, J. A. US Pat. 50378341, 1991; (b) Hacker, R. E.; Jourdan, G. P. Eur. Pat. 0414386, 1991; (c) Yamada, H. Eur. Pat. 0665224, 1994; (d) Mo, W.-Y.; He, H.-W. Chin J Struct Chem 2007, 26, 172.

[3] (a) Rewcastle, W. G.; Palmer, D. B.; Thompson, A. M.; Bridges, A. J.; Cody, D. R.; Zhou, H. R.; Fry, D. W.; McMichael, A.; Denny, W. A. J Med Chem 1996, 39, 1823; (b) T. B. Brown and M. F. Stevens. J Chem Soc Perkin Trans I 1975, 11, 1023. (c) Thompson, A. M.; Bridges, A. J.; Fry, D. W.; Kraker, A. J.; Denny, W. A. J Heterocycl Chem 1995, 38, 3780; (d) Liu, J.-C.; He, H.-W.; Ding, M.-W. Helv Chim Acta 2007, 90, 1337.

[4] Hosmane, R. S.; Lim, B. B.; Summers, M. F. J Org Chem 1988, 53, 5309.

[5] Ren, Q.-Y.; Cui, Z.-P.; He, H.-W.; Gu, Y.-C. J Fluorine Chem 2007, 128, 1369.

[6] Zhao, W.-G.; Wang, S.-H.; Wang, W.-Y.; Li, Z.-M. Huaxue Shiji 2000, 22, 376.

[7] Zhao, W.-G.; Liu, Z.-X.; Li, Z.-M.; Wang, B.-L. Synth Commun 2003, 33, 4229.

[8] Mo, W.-Y.; Liao, G.-H.; Wang, T.; He, H.-W. J Fluorine Chem 2008, 129, 519.

[9] Hostettmann, K.; Harborne, J. B. In Methods in plant Biochemistry; Dey, P. M., Eds.; Academic Press: London, 1991; Vol. 6, pp 33–46.