A Convenient Synthesis and Biological Activity of Novel Pyrido[4,3-d] pyrimidin-4(3H)-ones

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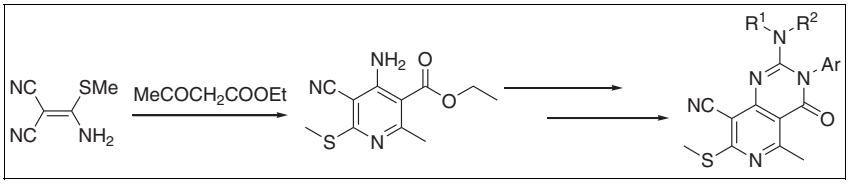

Fifteen novel 2-alkylamino-3-aryl-8-cyano-5-methyl-7-(methylthio)-pyrido[4,3-d]pyrimidin-4(3H)-ones **6a-6o** were designed and have been successfully synthesized via tandem aza-Wittig and annulation reactions with the corresponding iminophosphorances **4**, aryl isocyanate, and amines in good yields. Their structures were clearly verified by IR, ^{1}H NMR, EI-MS spectroscopy and elemental analysis, and in the case of compound **6i**, analyzed by single-crystal X-ray diffraction further. The preliminary results of an in *vivo* bioassay showed that some compounds display moderate antifungal activity.

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INTRODUCTION

Derivatives of pyridopyrimidines have been the focus of great interest over many years. This is because of the wide range of biological activities associated with heterocyclic scaffold. Some of these compounds have shown remarkable biological properties such as antitumor, antiviral, antibacterial, antihypertensive, antibronchitis, antiallergic, antiarthritic, and anti-HIV activities [1–7], whereas others exhibited good insecticidal, growth regulatory, herbicidal, and fungicidal activities [8–10]. For example, some related 4-(phenylamino)pyrido[d]pyrimidines have been reported as selective inhibitors of tyrosine phosphorylation by the epidermal growth-factor receptor and have become an important class of potential anticancer drugs [1,2].

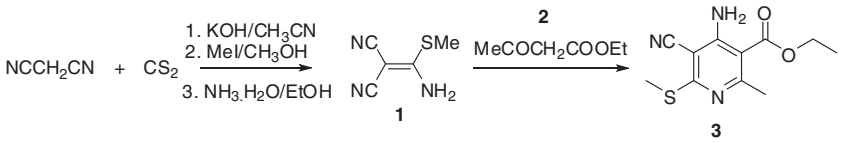
The pyrido[2,3-d]pyrimidine system has been studied in more detail, because examples have medicinal applications as inhibitors of tyrosine kinase [11] (AK) or dihydrofolate reductase [11,12]. However, few reports [1,13] are available so far to the preparation of pyrido[4,3-d]pyrimidine derivatives, which are of considerable interest as potential biologically active compounds or pharmaceuticals. The methods described so far for the preparation of some representative derivatives of this ring system involve two general routes [1–16]: (a) formation of the pyridine ring by cyclization of suitable substituents of a pyrimidine and (b) formation of the pyrimidine ring by cyclization of a suitable pyridine derivative. However, these methods often require forcing conditions, long reaction times, and complex synthetic pathways.

For the reason given earlier, the synthesis of pyrido[4,3-d] pyrimidine derivatives is challenging. The aza-Wittig reactions of iminophosphoranes have recently received increasing attention in view of their utility in the synthesis of *N*-heterocyclic compounds [17]. Previously, we had designed and synthesized fused pyrimidine derivatives such as thiazolopyrimidinones [18], thienopyrimidinones [19], and triazolopyrimidinones [20] prepared from various iminophosphoranes, and it was found that these novel fused pyrimidnes compounds displayed good antifungal activity against *Rhizoctonia solani*. As a continuation of our ongoing project related to the discovery and optimization of pesticide leads, we would like to describe a facile synthesis of novel pyrido[4,3-d]pyrimidine derivatives *via* the tandem aza-Wittig and cyclization reaction. The preliminary results of an *in vivo* bioassay indicated that some of these compounds display moderate antifungal activity.

RESULTS AND DISCUSSION

The intermediate **1** was prepared by the reaction of malononitrile and carbon bisulfide in the presence of KOH as base in acetonitrile and then treated with ammonium hydroxide in ethanol [21]. It was reported that 4-aminonicotinonitrile **3** was prepared from reaction of ketene *N,S*-acetal **1** with ethyl acetoacetate **2** in moderate yield (48%) by using anhydrous stannic chloride as catalyst [22]. Herein, we improved the conditions by using zinc nitrate as catalyst [21] instead of anhydrous stannic chloride to achieve much higher yield (95%) than the reported method(Scheme 1).

Scheme 1. Synthesis of 4-aminonicotinonitrile **3**.



Compound **3** was further converted to iminophosphorane **4** *via* reaction with triphenylphosphine, hexachloroethane, and triethylamine in good yield. Iminophosphorane **4** reacted with aromatic isocyanates and gave carbodiimides **5**. In refluxing toluene, the direct reaction of carbodiimides **5** with alkylamines did not react to give the target compounds **6**. However, when the solvent was changed to CH_2Cl_2 and in the presence of a catalytic amount of EtONa/EtOH, compound **5** was converted smoothly to the 2-alkylamino-pyrido[4,3-d]pyrimidin-4(3H)-ones **6** in satisfactory yields at room temperature (Scheme 2, Table 1).

Worth noting the reaction between carbodiimides **5** and primary amines were obtained mainly 2-alkylamino-pyrido [4,3-d]pyrimidin-4(3H)-ones **6** and another kind of cyclization compounds 2-arylamino-pyrido[4,3-d]pyrimidin-4(3H)-ones **8** were not founded (Scheme 3); this might be because of the geometry of the guanidine intermediate and conjugative effect of compounds **6**. As the amines were reacted with **5**, intermediates **7a** were formed because the amines would attack **5** mainly from the opposite direction of CO_2Et group because of the steric hindrance of CO_2Et group. At the same time, the compounds **6** are more stable than compounds **8** because of the conjugative effect between the pyridopyrimidine ring and phenyl ring. Therefore, only compounds **6** were obtained regioselectively.

All the compounds of the **6** series were obtained as white solids after recrystallization from ethanol. Their structures were fully characterized by IR, ^{1}H-NMR, EI-MS, and

Scheme 2. Synthesis of 2-alkylamino- pyrido[4,3-d]pyrimidin-4(3H)-ones **6**.

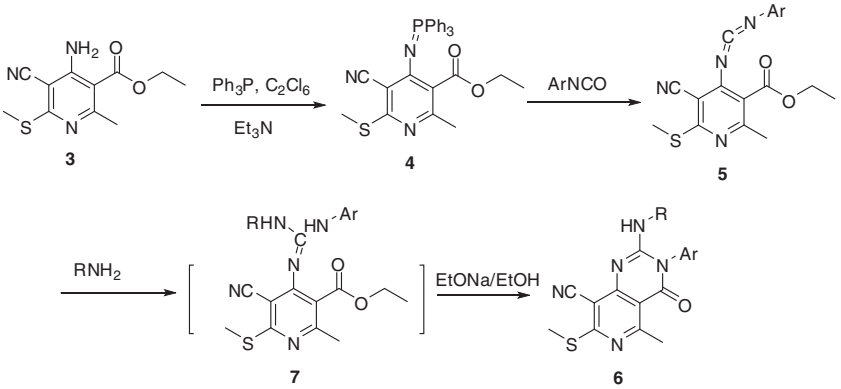


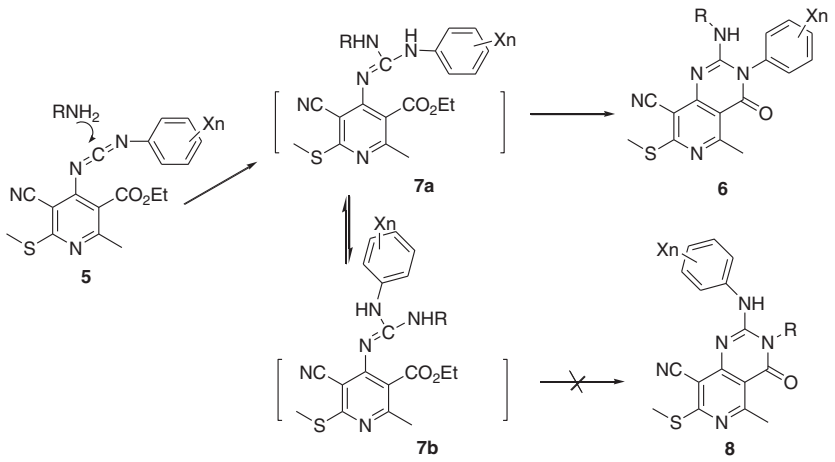
Table 1

Synthesis of compound **6**.

Compounds	NHR	Ar	Yield(%)[a]	Compounds	NHR	Ar	Yield(%)[a]
6a	PrNH	4-FPh	76	**6b**	BuNH	4-FPh	86
6c	*i*-BuNH	4-FPh	88	**6d**	*t*-BuNH	4-FPh	91
6e	$CH_3(CH_2)_4NH$	4-FPh	85	**6f**	$CH_3(CH_2)_5NH$	4-FPh	78
6g	Allylamino	4-FPh	87	**6h**	PrNH	4-ClPh	83
6i	BuNH	4-ClPh	82	**6j**	Cyclohexylamino	4-ClPh	91
6k	2-ClPhCH_2NH	Ph	85	**6l**	4-FPhCH_2NH	Ph	81
6m	3-MePhCH_2NH	Ph	87	**6n**	4-FPh CH_2CH_2NH	Ph	83
6o	4-MeOPh CH_2CH_2NH	Ph	79				

[a]Yields of isolated products based on iminophosphorane **4**.

Scheme 3. Possible mechanism of the formation of compound **6**.



elemental analysis. In the case of **6o**, the structure was additionally solved by single-crystal X-ray diffraction [23] (Fig. 1). All structures were supported spectroscopically. For example, the IR spectra of **6l** revealed CN and C=O absorption bands at 2216 and 1684 cm^{-1}, respectively; the signals attributable to the NH are found at 3390 cm^{-1}. The corresponding ^{1}H-NMR spectrum showed the 5-Me group at δ(H) 2.90 (s), and the CH_3 signals of the SMe appeared at δ(H) 2.69. The other signals resonated at δ(H) 6.97–7.65 (m, 9 arom. H), 4.63 (d, J = 5.6 Hz, CH_2), and 4.89 (s, NH). The mass spectrum of **6l** showed the molecular ion peak at *m/z* 431 as the base peak (100%). The structures of **6l** and the other analogs were further confirmed on the basis of elemental analysis.

The preliminary antifungal activity of compounds **6** series was measured at a concentration of 500 mg/L using a reported procedure [24] and the *in vivo* inhibition rates are listed in Table 2. Fungicidal activities of commercial fungicides chlorothalonil, dimethomorph, and thiophanate methyl as a control against the aforementioned five fungi were evaluated at the same condition. It was found that most of the compounds exhibited inhibitory activity against *Cladosporium cucumerinum* and *Phytophthora capsici*. They did have any obvious influence on the growth of *Corynespora cassiicola*, *Rhizoctonia solani*, and *Fusarium oxysporum*. Although the antifungal activity of these compounds are lower than that of commercial fungicides, the inhibitory activity of title compounds could be further improved by incorporating appropriate functional groups.

In summary, we have developed an efficient and convenient synthetic method for the preparation of pyrido[4,3-d] pyrimidin-4(3H)-ones in good yield *via* tandem aza-Wittig reaction and annulation reactions. The preliminary results of an *in vivo* bioassay showed that some compounds display moderate antifungal activities and further bioassay, optimization, and structure-activity relationships of the title compounds are underway.

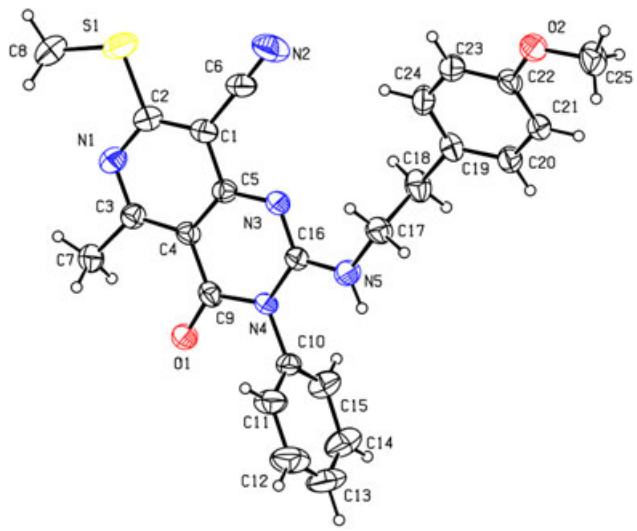


Figure 1. Perspective view of the X-ray crystal structure of **6o**. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

EXPERIMENTAL

Melting points were measured on an electrothermal melting point apparatus X-4 (Beijing Tektronix Instrument Inc., Beijing) and uncorrected. Mass spectra were measured on a Finnigan Trace MS 2000 spectrometer (Thermo Finnigan, Silicon Valley, CA). IR spectra were recorded on a NicoletAvatar 360 FT-IR spectrometer (Thermo Electron Inc., San Jose, CA) as KBr pellets with absorption in cm^{-1}. ^{1}H NMR spectra were recorded in $CDCl_3$ on a Mercury Plus 400 MHz (Varian, Palo Alto, CA), and resonances were given in ppm (δ) relative to TMS (δ 0.00 ppm). ^{13}C NMR spectra were recorded using $CDCl_3$ as the solvent on a Unity-Inova 600 MHz (Varian, Palo Alto, CA), and resonances are given in ppm (δ) relative to $CDCl_3$ (δ 77.00 ppm). The elementary analysis was performed on a Vario EL III elementary analysis instrument (Elementar Analysensysteme GmbH, Germany). The X-ray single crystal diffractometer is Bruker Smart Apex CCD X-ray crystal (Elementar Analysensysteme GmbH, Germany). All of the solvents and materials were reagent grade and purified as required.

Table 2

Antifungal activity (relative inhibition (%), *in vivo*, 500 mg/L) of compound **6**.

Compounds	*Cladosporium cucumerinum*	*Corynespora cassiicola*	*Rhizoctonia solani*	*Phytophthora capsici*	*Fusarium oxysporum*
6a	44	50	54	3	0
6b	51	47	0	63	0
6c	21	19	0	54	0
6d	72	21	0	44	0
6e	58	46	0	49	0
6f	7	39	0	23	6
6g	65	3	37	68	28
6h	0	25	77	24	11
6i	69	46	0	46	0
6j	9	58	0	36	6
6k	37	0	0	58	17
6l	18	17	50	68	0
6m	25	38	31	71	6
6n	70	47	0	55	11
6o	40	29	0	39	6
Chlorothalonil	94	95	77		
Dimethomorph				98	
Thiophanatemethyl					90

Synthesis of 6-methylsulfanyl-4-amino-5-cyano-2-methyl-nicotinic acid ethyl esters 3. A mixture of 2-(methylsulfanyl-amino-methylene)-malononitrile **1** (10 mmol) and $Zn(NO_2)_2.6H_2O$ (20 mmol) were added to a stirred solution of ethyl acetoacetate **2** (20 mmol) in ethanol (30 ml). The solution was refluxed for 6 ~ 8 h and then cooled at room temperature. The crude precipitated product was collected by using filtration. Further purification was accomplished by recrystallization from ethanol to give pure products of **3** in 95% yield. White solid. Mp 136.0–138.0°C; ^{1}H NMR ($CDCl_3$) δ 1.41 (t, J = 7.2 Hz, 3H, $\underline{OCH_2}CH_3$), 2.62 (s, 3H, SCH_3), 2.70 (s, 3H, py-$\underline{CH_3}$), 4.39 (q, J = 7.2 Hz, 2H, OCH_2), 6.68 ppm (s, 2H, NH_2); ms: *m/z* 252 (M^++1, 23), 251 (M^+, 100), 250 (M^+-1, 41), 223 (47), 205 (31), 177(30); *Anal*. Calcd for $C_{11}H_{13}N_3O_2S$: C, 52.57; H, 5.21; N, 16.72; S, 12.76. Found: C, 52.75; H, 4.80; N, 16.89; S, 12.72.

Synthesis of 6-methylsulfanyl-5-cyano-2-methyl-4-[(triphenyl-phosphanylidene)amino]-nicotinic acid ethyl esters 4. To a solution of **3** (2.51 g, 10 mmol) in MeCN (40 mL), Ph_3P (7.86 g, 30 mmol), C_2Cl_6 (7.12 g, 30 mmol), and in this order, Et_3N (8.0 ml) was added. The mixture was stirred for 6–7 h at room temperature. Then, the solution was concentrated, and the residue was recrystallized from EtOH to give **4** in 90% yield. Mp 153–155°C. ^{1}H NMR ($CDCl_3$) δ 1.09(t, J = 7.2 Hz, 3H, CH_3), 2.37(s, 3H, SCH_3), 2.51(s, 3H, py-CH_3), 3.95(q, J = 7.2 Hz, 2H, OCH_2), 7.44 ~ 7.71 ppm (m, 15H, Ph-H).

General procedure for the preparation of 2-alkylamino-3-aryl-8-cyano-5-methyl- 7-(methylthio)-pyrido[4,3-d]pyrimidin-4(3H)-ones (6a–o). To a solution of iminophosphorane **4** (0.51 g, 1 mmol) in dry methylene chloride (10 mL), aryl isocyanate (1.1 mmol) was added under nitrogen at room temperature. After the reaction mixture was left unstirred for 6–12 h, the solvent was removed under vacuum and Et_2O/petroleum ether (1:2 20 mL) was added to precipitate triphenylphosphine oxide. Removal of the solvent gave carbodiimides **5**, which were used directly without further purification.

Alkylamine (1.1 mmol) was added into the solution of **5** prepared earlier in CH_2Cl_2 (10 mL). After the reaction mixture was stirred continuously for an additional 6 h, the solvent was removed and 10 mL of anhydrous ethanol with several drops of sodium ethoxide in ethanol (3 M) were added. After stirring for another 0.5–1 h, the solution was condensed and the residue was recrystallized from dichloromethane/petroleum ether to give pure 2-alkylamino-3-aryl-8-cyano-5-methyl-7-(methylthio)-pyrido [4,3-d]pyrimidin-4(3H)-ones (**6a–o**).

8-Cyano-3-(4-fluorophenyl)-5-methyl-7-(methylthio)-2-propylamino-pyrido[4,3-d]pyrimidin-4(3H)-one (6a). This compound was obtained as white solid, mp 270.5–272.0°C, yield 0.29 g, 76%; IR (KBr): 3357, 2963, 2221(CN), 1682(C=O), 1584, 1559, 1508 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 0.88(t, J = 7.2 Hz, 3H, CH_3), 1.55 ~ 1.60(m, 2H, CH_2), 2.67(s, 3H, SCH_3), 2.87(s, 3H, py-CH_3), 3.50 ~ 3.52(m, 2H, NCH_2), 4.46(s, 1H, NH), 7.26 ~ 7.33 ppm (m, 4H, Ar-H). ms: *m/z* 385 (M^++2, 11), 384 (M^++1, 38), 383 (M^+, 100), 382 (M^+−1, 24), 355 (13), 340 (75), 325 (8), 95 (33), 75 (15), 41 (76); *Anal*. Calcd for $C_{19}H_{18}FN_5OS$: C, 59.51; H, 4.73; N, 18.26. Found: C, 59.85; H, 4.54; N, 18.60.

2-Butylamino-8-cyano-3-(4-fluorophenyl)-5-methyl-7-(methylthio)-pyrido[4,3-d]pyrimidin-4(3H)-one (6b). This compound was obtained as white solid, mp >280°C, yield 0.34 g, 86%; IR (KBr): 3344, 2951, 2222(CN), 1682(C=O), 1584, 1559, 1510 cm^{-1}; ^{1}H NMR (DMSO) δ 0.88(t, J = 7.2 Hz, 3H, CH_3), 1.23 ~ 1.26(m, 2H, CH_2), 1.50 ~ 1.53(m, 2H, CH_2), 2.63(s, 3H, SCH_3), 2.78(s, 3H, py-CH_3), 3.32 ~ 3.36(m, 2H, NCH_2), 6.84(s, 1H, NH), 7.41 ~ 7.44 ppm (m, 4H, Ar-H). ms: *m/z* 399 (M^++2, 4), 398 (M^++1, 38), 397 (M^+, 30), 396 (M^+−1, 14), 382 (31), 368 (14), 340 (50), 325 (9), 43 (100); *Anal*. Calcd for $C_{20}H_{20}FN_5OS$: C, 60.44; H, 5.07; N, 17.62. Found: C, 60.95; H, 4.65; N, 18.00.

8-Cyano-3-(4-fluorophenyl)-2-(iso-butylamino)-5-methyl-7-(methylthio)-pyrido[4,3-d]pyrimidin-4(3H)-one (6c). This compound was obtained as white solid, mp > 280°C, yield 0.35 g, 88%; IR (KBr): 3374, 2930, 2220(CN), 1682(C=O), 1559, 1531 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 0.86(d, J = 6.8 Hz, 6H, 2*CH_3), 1.85 ~ 1.89(m, 1H, CH), 2.68(s, 3H, SCH_3), 2.89(s, 3H, py-CH_3), 3.34 ~ 3.38(m, 2H, NCH_2), 4.48(s, 1H, NH), 7.26 ~ 7.34 ppm (m, 4H, Ar-H). *Anal*. Calcd for $C_{20}H_{20}FN_5OS$: C, 60.44; H, 5.07; N, 17.62. Found: C, 60.58; H, 4.63; N, 17.31.

8-Cyano-3-(4-fluorophenyl)-5-methyl-7-(methylthio)-2-(tert-butylamino)-pyrido[4,3-d]pyrimidin-4(3H)-one (6d). This compound was obtained as white solid, mp 278.0–278.9°C, yield 0.36 g, 91%; IR (KBr): 3416, 2928, 2213(CN), 1685 (C=O), 1564 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 1.45(s, 9H, 3*CH_3), 2.68(s, 3H, SCH_3), 2.87(s, 3H, py-CH_3), 4.30(s, 1H, NH), 7.29 ~ 7.32 ppm (m, 4H, Ar-H). ^{13}C NMR($CDCl_3$) δ 13.1, 26.4, 28.8, 53.9, 98.5, 108.0, 114.9, 117.9, 118.2, 129.6, 130.7, 151.8, 157.7, 161.6, 164.6, 167.2 ppm; *Anal*. Calcd for $C_{20}H_{20}FN_5OS$: C, 60.44; H, 5.07; N, 17.62. Found: C, 60.72; H, 4.61; N, 17.30.

8-Cyano-3-(4-fluorophenyl)-5-methyl-7-(methylthio)-2-(pentylamino)-pyrido[4,3-d]pyrimidin-4(3H)-one (6e). This compound was obtained as white solid, mp 263.0–264.0°C, yield 0.35 g, 85%; IR (KBr): 3356, 2857, 2220(CN), 1682(C=O), 1558, 1508 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 0.87(t, J = 7.2 Hz, 3H, CH_3), 1.23 ~ 1.32(m, 4H, 2*CH_2CH_2), 1.52 ~ 1.55(m, 2H, CH_2), 2.67(s, 3H, SCH_3), 2.88(s, 3H, py-CH_3), 3.50 ~ 3.53(m, 2H, NCH_2), 4.48(s, 1H, NH), 7.30 ~ 7.33 ppm (m, 4H, Ar-H). *Anal.* Calcd for $C_{21}H_{22}FN_5OS$: C, 61.29; H, 5.39; N, 17.02. Found: C, 61.20; H, 5.11; N, 16.76.

8-Cyano-3-(4-fluorophenyl)-2-(hexylamino)-5-methyl-7-(methylthio)-pyrido[4,3-d]pyrimidin-4(3H)-one (6f). This compound was obtained as white solid, mp 162.0–164.0°C; yield 0.33 g, 78%; IR (KBr): 3366, 2929, 2220(CN), 1683(C=O), 1558, 1532 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 0.86(t, J = 6.4 Hz, 3H, CH_3), 1.26 ~ 1.53(m, 8H, 4*CH_2), 2.68(s, 3H, SCH_3), 2.89(s, 3H, py-CH_3), 3.50 ~ 3.53(m, 2H, NCH_2), 4.42(s, 1H, NH), 7.29 ~ 7.35 ppm (m, 4H, Ar-H). *Anal*. Calcd for $C_{22}H_{24}FN_5OS$: C, 62.10; H, 5.68; N, 16.46. Found: C, 62.38; H, 5.35; N, 16.19.

2-Allylamino-8-cyano-3-(4-fluorophenyl)-5-methyl-7-(methylthio)-pyrido[4,3-d]pyrimidin-4(3H)-one (6g). This compound was obtained as white solid, mp 285.0–286.0°C; yield 0.33 g, 87%; ^{1}H NMR ($CDCl_3$) δ 2.63(s, 3H, SCH_3), 2.78(s, 3H, CH_3), 3.93 ~ 3.96(m, 2H, NCH_2), 5.07(d, J = 10 Hz, 1H, CH), 5.17(d, J = 16.8 Hz, 1H, CH), 5.81 ~ 5.85 (m, 1H, CH), 7.07(s, 1H, NH), 7.41 ~ 7.49 ppm (m, 4H, Ar-H). ms: *m/z* 382 (M^++1, 6), 381 (M^+, 19), 380 (M^+-1, 19), 366 (100), 348 (19), 340 (10), 204 (4), 190 (6), 135 (14); *Anal*. Calcd for $C_{19}H_{16}FN_5OS$: C, 59.83; H, 4.23; N, 18.36. Found: C, 60.10; H, 4.22; N, 17.91.

3-(4-Chlorophenyl)-8-cyano-5-methyl-7-(methylthio)-2-propylamino-pyrido[4,3-d]pyrimidin-4(3H)-one (6h). This compound was obtained as white solid, mp 235.0–238.0°C, yield 0.33 g, 83%; ^{1}H NMR ($CDCl_3$) δ 0.88(t, J = 7.2 Hz, 3H, CH_3), 1.55 ~ 1.61(m, 2H, CH_2), 2.68(s, 3H, SCH_3), 2.89(s, 3H, py-CH_3), 3.51 ~ 3.53(m, 2H, NCH_2), 4.46(s, 1H, NH), 7.25 ~ 7.63 ppm (m, 4H, Ar-H). ms: *m/z* 401 (M^++2, 37), 400 (M^++1, 26), 399 (M^+, 96), 356 (100), 341 (9), 246 (7), 231 (8), 204 (10), 152 (21); *Anal*. Calcd for $C_{19}H_{18}ClN_5OS$: C, 57.07; H, 4.54; N, 17.51. Found: C, 57.25; H, 4.56; N, 16.94.

3-(4-Chlorophenyl)-8-cyano-2-butylamino-5-methyl-7-(methylthio)-pyrido[4,3-d]pyrimidin-4(3H)-one (6i). This compound was obtained as white solid, mp 233.0–234.0°C, yield 0.34 g, 82%; ^{1}H NMR ($CDCl_3$) δ 0.91(t, J = 7.2 Hz, 3H, CH_3), 1.24 ~ 1.32(m, 2H, CH_2), 1.49 ~ 1.56(m, 2H, CH_2), 2.68 (s, 3H, SCH_3), 2.88(s, 3H, py-CH_3), 3.53 ~ 3.55(m, 2H, NCH_2), 4.44(s, 1H, NH), 7.25 ~ 7.63 ppm (m, 4H, Ar-H). ms: *m/z* 415 (M^++2, 8), 414 (M^++1, 26), 413 (M^+, 100), 322 (34), 204 (52), 182 (55), 167 (67), 106 (31), 91 (65), 77(20); *Anal*. Calcd for $C_{20}H_{20}ClN_5OS$: C, 58.03; H, 4.87; N, 16.92. Found: C, 58.12; H, 4.41; N, 16.61.

3-(4-Chlorophenyl)-2-cyclohexylamino-8-cyano-5-methyl-7-(methylthio)-pyrido[4,3-d]pyrimidin-4(3H)-one (6j). This compound was obtained as white solid, mp >280°C, yield 0.40 g, 91%; ^{1}H NMR ($CDCl_3$) δ 1.06 ~ 1.36(m, 4H, 2*CH_2), 1.38 ~ 1.47(m, 2H, CH_2), 1.59 ~ 1.62(m, 2H, CH_2), 1.97 ~ 1.99(m, 2H, CH_2), 2.68(s, 3H, SCH_3), 2.87(s, 3H, CH_3), 4.10 ~ 4.14(m, 1H, CH), 4.26(d, J = 7.2 Hz, 1H, NH), 7.24 ~ 7.63 ppm (m, 4H, Ar-H). ms: *m/z* 441 (M^++2, 32), 440 (M^++1, 18), 439 (M^+, 83), 358 (93), 356 (100), 341 (15), 247 (8), 192 (19), 177(6), 152(7); *Anal*. Calcd for $C_{22}H_{22}ClN_5OS$: C, 60.06; H, 5.04; N, 15.92. Found: C, 59.84; H, 4.44; N, 15.47.

2-(2-Chlorobenzylamino)-8-cyano-5-methyl-7-(methylthio)-3-phenyl-pyrido[4,3-d]pyrimidin-4(3H)-one (6k). This compound was obtained as white solid, mp 202.9–204.9°C, yield 0.38 g, 85%; IR (KBr): 3352, 2919, 2224(CN), 1688(C=O), 1557, 1532 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 2.68(s, 3H, SCH_3), 2.87(s, 3H, py-CH_3), 4.68(d, J = 6.4 Hz, 2H, NCH_2), 5.31(s, 1H, NH), 7.17 ~ 7.80 ppm (m, 9H, Ar-H). ms: *m/z* 450 (M^++3, 11), 449 (M^++2, 41), 448 (M^++1, 34), 447 (M^+, 100), 446 (M^+-1, 17), 412 (78), 322 (33), 140 (14); *Anal*. Calcd for $C_{23}H_{18}ClN_5OS$: C, 61.67; H, 4.05; N, 15.63. Found: C, 61.84; H, 3.88; N, 15.24.

8-Cyano-2-(4-fluorobenzylamino)-5-methyl-7-(methylthio)-3-phenyl-pyrido[4,3-d]pyrimidin-4(3H)-one (6l). This compound was obtained as white solid, mp 230.0–232.0°C, yield 0.35 g, 81%; IR (KBr): 3390, 2925, 2216(CN), 1684(C=O), 1556, 1529 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 2.69(s, 3H, SCH_3), 2.90(s, 3H, py-CH_3), 4.63(d, J = 5.6 Hz, 2H, NCH_2), 4.89(s, 1H, NH), 6.97 ~ 7.65 ppm (m, 9H, Ar-H). ms: *m/z* 433 (M^++2, 10), 432 (M^++1, 28), 431 (M^+, 100), 430 (M^+-1, 17), 322 (12), 185 (13); *Anal*. Calcd for $C_{23}H_{18}FN_5OS$: C, 64.02; H, 4.20; N, 16.23. Found: C, 64.79; H, 3.98; N, 15.92.

8-Cyano-5-methyl-2-(3-methylbenzylamino)-7-(methylthio)-3-phenyl-pyrido[4,3-d]pyrimidin-4(3H)-one (6m). This compound was obtained as white solid, mp 190.3–192.9°C, yield 0.37 g, 87%; IR (KBr): 3429, 2922, 2215(CN), 1698(C=O), 1581, 1540 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 2.33(s, 3H, CH_3), 2.68(s, 3H, SCH_3), 2.89(s, 3H, py-CH_3), 4.65(d, J = 5.6 Hz, 2H, CH_2), 4.86(s, 1H, NH), 7.07 ~ 7.64 ppm (m, 9H, Ar-H). ms: *m/z* 429 (M^++2, 9), 428 (M^++1, 29), 427 (M^+, 100), 426 (M^+-1, 16), 322 (14), 196 (33), 120 (32); *Anal*. Calcd for $C_{24}H_{21}N_5OS$: C, 67.43; H, 4.95; N, 16.38. Found: C, 67.89; H, 4.77; N, 16.18.

8-Cyano-2-(4-fluorophenethylamino)-5-methyl-7-(methylthio)-3-phenyl-pyrido[4,3-d]pyrimidin-4(3H)-one (6n). This compound was obtained as white solid, mp 207.0–208.7°C, yield 0.37 g, 83%; IR (KBr): 3429, 2928, 2222(CN), 1693(C=O), 1557, 1535 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 2.69(s, 3H, SCH_3), 2.86(t, J = 7.2 Hz, 2H, CH_2), 2.89(s, 3H, py-CH_3), 3.70–3.72(m, 2H, NCH_2), 4.46(s, 1H, NH), 6.91 ~ 7.58 ppm (m, 9H, Ar-H). ms: *m/z* 447 (M^++2, 5), 446 (M^++1, 14), 445 (M^+, 63), 425 (20), 322 (100), 307 (7), 122 (50); *Anal*. Calcd for $C_{24}H_{20}FN_5OS$: C, 64.70; H, 4.52; N, 15.72. Found: C, 64.67; H, 4.17; N, 15.30.

8-Cyano-2-(3-methoxyphenethylamino)-5-methyl-7-(methylthio)-3-phenyl-pyrido[4,3-d]pyrimidin-4(3H)-one (6o). This compound was obtained as white solid, mp 185.5–188.5°C, yield 0.36 g, 79%; IR (KBr): 3412, 2931, 2219(CN), 1689(C=O), 1556, 1535 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 2.69(s, 3H, SCH_3), 2.85(t, J = 6.4 Hz, 2H, CH_2), 2.90(s, 3H, py-CH_3), 3.72 ~ 3.75(m, 2H, NCH_2), 3.79(s, 3H, OCH_3), 4.47(s, 1H, NH), 6.61 ~ 7.54 ppm (m, 9H, Ar-H). ms: *m/z* 459 (M^++2, 4), 458 (M^++1, 13), 457 (M^+, 41), 322 (35), 307 (10), 134 (100), 91 (23), 77(23); *Anal*. Calcd for $C_{25}H_{23}N_5O_2S$: C, 65.63; H, 5.07; N, 15.31. Found: C, 65.48; H, 4.97; N, 15.07.

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