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RE 444 RESPUESTA AL OFICIO

Señores

SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO
DIVISIÓN DE NUEVAS CREACIONES

E. _____ S. _____ D. _____

Ref: Expediente No. 03-112.613

Solicitud de patente de invención titulada: "PIRIMIDINAS INHIBIDORAS DE
CDK, SU PREPARACIÓN Y USO COMO MEDICAMENTO."

Solicitante: SCHERING AKTIENGESELLSCHAFT.

N. REF: P2003/0000193

RESPUESTA AL OFICIO No. 11329 NOTIFICADO POR FIJACIÓN EN LISTA No. 289,
DE FECHA 5 DE SEPTIEMBRE DEL 2008
(REQUERIMIENTO ARTÍCULO 46)

J. IAN RAISBECK, mayor de edad y vecino de Bogotá, identificado tal como aparece al pie de mi firma, en mi carácter de apoderado de la sociedad solicitante, me permito manifestar lo siguiente ante el auto emanado por ese Despacho, dentro del plazo legal previsto en el artículo 46 de la Decisión 486:

1. Presento copia completa de los siguientes documentos exigidos por el Examinador en el examen de búsqueda de anterioridades, según lo previsto en el Artículo 46 de la Decisión 486 de la Comisión de la Comunidad Andina:

- Boschelli et al, Journal of medicinal chemistry (1998), 41(22), 4365-4377;
- Sprague et al, Journal of the Chemical Society (1936), 58:423-6;
- Curd et al, Journal of the Chemical Society (1946), 378-84;

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Synthesis and Tyrosine Kinase Inhibitory Activity of a Series of 2-Amino-8*H*-pyrido[2,3-*d*]pyrimidines: Identification of Potent, Selective Platelet-Derived Growth Factor Receptor Tyrosine Kinase Inhibitors

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Screening of a compound library led to the identification of 2-amino-6-(2,6-dichlorophenyl)-8-methylpyrido[2,3-*d*]pyrimidine (**1**) as a inhibitor of the platelet-derived growth factor receptor (PDGFr), fibroblast growth factor receptor (FGFr), and c-src tyrosine kinases (TKs). Replacement of the primary amino group at C-2 of **1** with a 4-(*N,N*-diethylaminoethoxy)phenylamino group yielded **2a**, which had greatly increased activity against all three TKs. In the present work, variation of the aromatic group at C-6 and of the alkyl group at N-8 of the pyrido[2,3-*d*]pyrimidine core provided several analogues that retained potency, including derivatives that were biased toward inhibition of the TK activity of PDGFr. Analogues of **2a** with a 3-thiophene or an unsubstituted phenyl group at C-6 were the most potent inhibitors. Compound **54**, which had IC₅₀ values of 31, 88, and 31 nM against PDGFr, FGFr, and c-src TK activity, respectively, was active in a variety of PDGF-dependent cellular assays and blocked the in vivo growth of three PDGF-dependent tumor lines.

Introduction

The tyrosine kinases (TKs) are a key group of enzymes that catalyze the specific phosphorylation of tyrosine residues on proteins. The TKs can be divided into two classes, the cytoplasmic TKs such as c-src and the transmembrane growth factor receptor TKs (RTKs). When growth factors such as platelet-derived growth factor (PDGF), fibroblast growth factor (FGF), epidermal growth factor (EGF), or vascular endothelial growth factor (VEGF) bind their receptors, the receptors dimerize and become active TKs. The subsequent autophosphorylation of the receptors leads to a signal transduction cascade which triggers a wide variety of events including proliferation and migration.^{1,2}

Since distinct TKs are implicated in such diverse conditions as angiogenesis,^{3,4} restenosis,^{5,6} atherosclerosis,⁷ and tumor growth,⁸ selective TK inhibitors are highly desirable. The obstacles to identifying selective inhibitors are the large number of TKs that have been described and the structural homology between the TKs, especially in their ATP-binding regions.⁹ Despite these difficulties several selective TK inhibitors have been identified¹⁰⁻³⁸ including those of EGF²⁷⁻³⁵ and FGFr TK³⁶⁻³⁸ activity reported by Parke-Davis. We were also interested in identifying compounds that could selectively modulate the activity of PDGFr TK due to the

importance of PDGF and its receptors in cancer.¹⁰⁻¹⁷ Several years ago it was found that PDGF was the human homologue of the simian oncoprotein v-sis.^{39,40} Later studies showed that both PDGF and its receptor are expressed in several types of human gliomas.⁴¹⁻⁴⁴ Furthermore, a shortened form of the PDGF receptor inhibited the growth of C6 glioma cells.⁴⁵ Therefore a small molecule capable of suppressing PDGFr TK activity has potential utility as an anticancer therapeutic agent.

We previously reported that screening of a compound library led to the identification of 2-amino-6-(2,6-dichlorophenyl)-8-methyl-8*H*-pyrido[2,3-*d*]pyrimidin-7-one (**1**) as an inhibitor of PDGFr, FGFr, and c-src TK activity.⁴⁶ Optimization of the various groups at C-2 of **1** determined that aromatic amino substituents were greatly preferred over primary or alkyl amino groups. This finding was exemplified by the broad-spectrum TK inhibitor **2a** which has a 4-(*N,N*-diethylaminoethoxy)phenylamino group at C-2 and had IC₅₀ values of 79, 43, and 9 nM against PDGFr, FGFr, and c-src TK activity, respectively.⁴⁷ While the initial focus was on variation of the C-2 amino group, it was also determined that replacement of the 8-methyl group of **2a** with an ethyl group provided **2b**, which was roughly equipotent to **2a** but more PDGFr-selective. It was considered that further modification at the N-8 position of the pyrido[2,3-*d*]pyrimidin-7-one could possibly lead to more active and/or more selective congeners.

In the initial screen that identified **1** as a TK inhibitor, similar activity was found for the related pyrido[2,3-*d*]pyrimidine **3a**.⁴⁸ This nonspecific kinase inhibi-

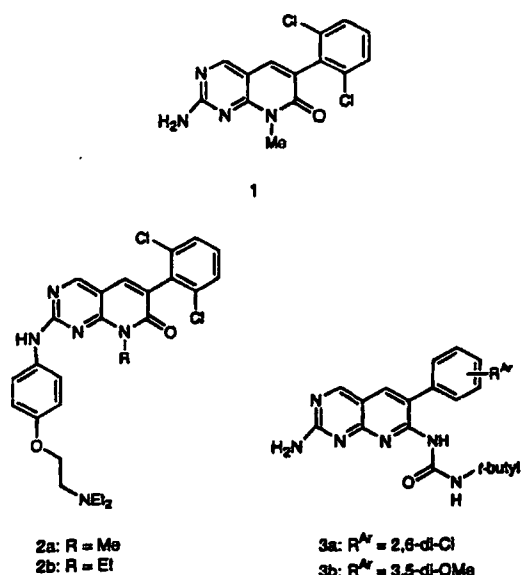
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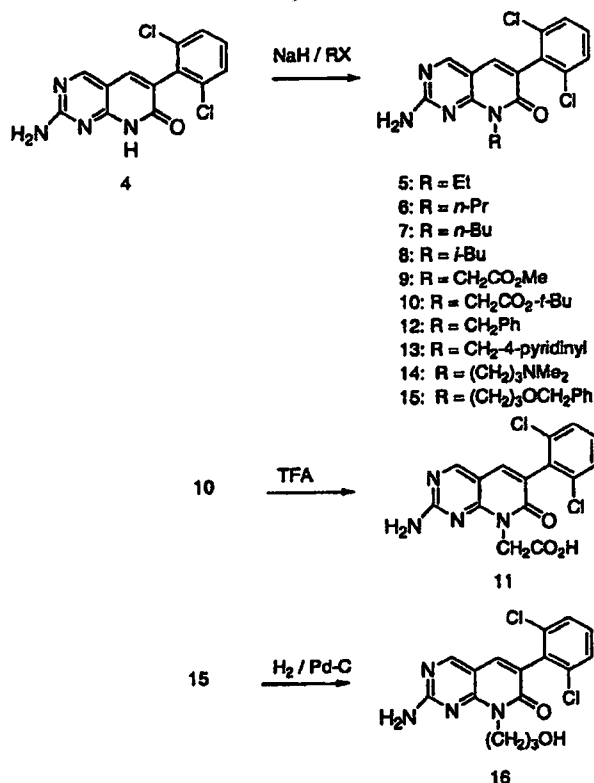
tor was converted into the selective FGFR TK inhibitor **3b** by replacement of the 2,6-dichlorophenyl group at C-6 of **3a** with a 3,5-dimethoxyphenyl group.^{36,37} In addition, a model of the binding of these two classes of pyrido[2,3-*d*]pyrimidines to the kinase domains of TKs proposed that modification of the group at C-6 could confer selectivity.⁴⁹ All of these findings encouraged us to utilize **2a** as a template for the preparation of potential selective inhibitors, through variation of the C-6 and N-8 moieties.⁵⁰

Chemistry

Additional N-8 alkyl derivatives of **1** were obtained by alkylation of **4**⁴⁶ with various halides employing sodium hydride as the base. The minor O-alkylation product was readily separated from the major product by chromatography. Compounds **5**–**10** and **12**–**15** were prepared by this route as shown in Scheme 1. Cleavage of the *tert*-butyl ester group of **10** with trifluoroacetic acid provided the acid derivative **11**. Catalytic hydrogenation of **15** removed the benzyl group to give **16**. We were not able to find conditions to convert **15** to **16** without some concomitant dechlorination.

Analogues of **2a** with various aromatic groups at C-6 of the pyrido[2,3-*d*]pyrimidin-7-one could be prepared as shown in Scheme 2. To this end, condensation of the 2-methyl sulfide derivative **17**⁴⁶ with phenylacetonitrile or 3-thiopheneacetonitrile in DMF in the presence of potassium carbonate gave the pyridopyrimidin-7-imines **18** and **19** that were then hydrolyzed to the 7-ones **20** and **21**. Oxidation of the methyl sulfide groups of **20** and **21** with *m*-chloroperbenzoic acid provided the sulfones **22** and **23**. The sulfone group was readily displaced by arylamines such as aniline, converting **22** and **23** to the 2-phenylamino derivatives **24** and **25**. In a similar fashion treatment of **22** and **23** with 4-aminopyridine provided **26** and **27**, while 4-(*N,N*-diethylaminoethoxy)aniline⁵¹ provided **28** and **29**, respectively. While for arylamines it was necessary to employ the more labile sulfone group, in the case of the more nucleophilic alkylamines the sulfide group was sufficiently reactive. Therefore, treatment of **26** with

Scheme 1

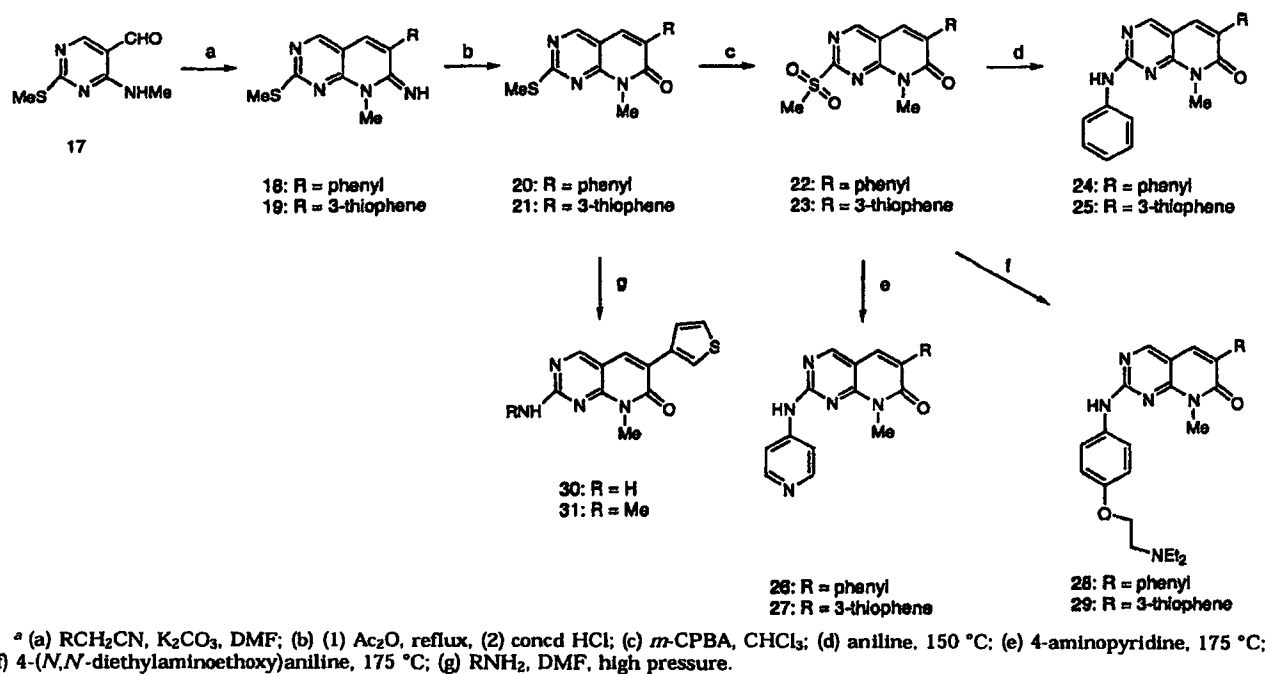


ammonia or methylamine in a sealed tube provided **30** and **31**, respectively (listed in Table 2).

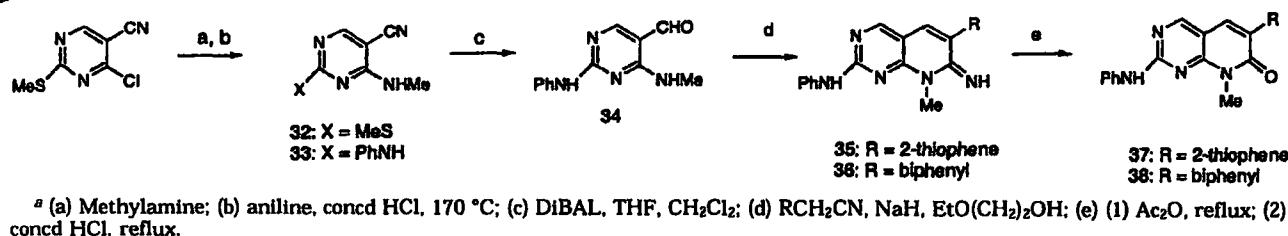
A more direct route to the preparation of analogues retaining the methyl group at N-8 and a phenylamino group at C-2 while varying the aromatic group at C-6 is shown in Scheme 3. Treatment of 4-chloro-5-cyano-2-methylsulfanyl-pyrimidine⁵² with methylamine provided **32**. Displacement of the 2-methyl sulfide group of **32** with aniline in the presence of a catalytic amount of concentrated hydrochloric acid gave **33**. Reduction of the nitrile group of **33** with diisobutylaluminum hydride followed by hydrolysis gave the corresponding aldehyde **34**. It should be noted that while the desired product was obtained by this protocol, it was necessary to stop the reaction while some **33** remained since extended reaction time led to the formation of additional products. Condensation of **34** with 2-thiopheneacetonitrile or 4-biphenylacetonitrile in 2-ethoxyethanol and sodium hydride gave good yields of the pyridopyrimidin-7-imines **35** and **36**, respectively. Acetylation of the imines followed by acid hydrolysis provided the corresponding 7-ones **37** and **38**.

2-Phenylamino analogues with an ethyl group at N-8 group were prepared from **44**, the 4-ethylamino analogue of **34**. While a similar route to that used to prepare **34** could be adapted, a more reproducible route to **44** starting with commercially available **39** was employed (Scheme 4). Thus, the chloro group of **39** was displaced with aqueous ethylamine to give **40**. Oxidation of the 2-methyl sulfide group of **40** with *m*-chloroperbenzoic acid gave mixtures of products thought to arise due to the instability of the corresponding sulfone. It was found that use of (±)-*trans*-2-phenylsulfonyl-3-phenyloxaziridine⁵³ as the oxidant provided

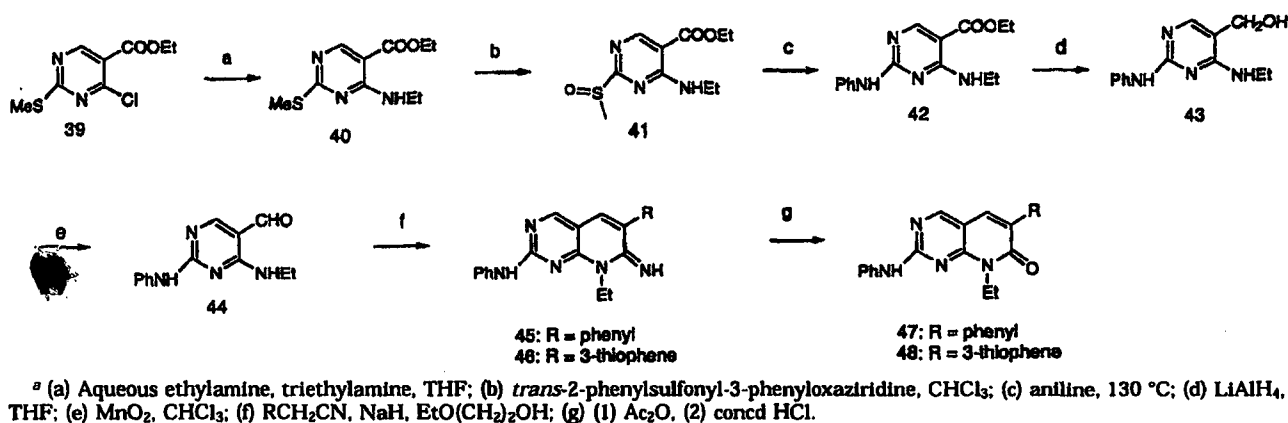
Scheme 2^a



Scheme 3^a



Scheme 4^a



the corresponding sulfoxide **41** in good yield. Displacement of the sulfoxide group of **41** with aniline at elevated temperature gave the desired 2-phenylamino derivative **42**. The ester group at C-5 was sequentially reduced to the alcohol **43** with lithium aluminum hydride and then oxidized to the aldehyde **44** with manganese oxide. Condensation of **44** with phenylacetone in the presence of sodium hydride in ethoxyethanol gave the pyrido[2,3-*d*]pyrimidin-7-imine **45** which was hydrolyzed to the 7-one derivative **47** as described previously. The 7-imine (**46**) and 7-one (**48**)

derivatives containing a 3-thiophene group at C-6 were prepared in an analogous fashion from **44**.

The more flexible route employing the 2-methyl sulfide group was then used to allow for variation of the amino group at C-2 as shown in Scheme 5. Aldehyde **49**⁴⁶ was condensed with phenylacetone, and the resultant imine **50** was hydrolyzed to the 7-one **51**. Oxidation of the 2-methyl sulfide group of **51** with *m*-chloroperoxybenzoic acid gave sulfone **52** that was treated with aniline to provide **47** by an alternate route to that used initially (Scheme 5). The additional 2-ary-

Conclusion

We have demonstrated that a screening lead, **1**, could be converted into a potent inhibitor of the activity of PDGFR TK. By replacement of the 2-amino group with 4-(*N,N*-diethylaminoethoxy)phenylamino, the C-6 2,6-dichlorophenyl group with phenyl, and the N-8 methyl group with ethyl, **1** was converted into **54**. The IC₅₀ values for the inhibition of the TK activity of PDGFR, FGFR, and c-src improved from 3.8, 1.3, and 0.26 μ M for **1** to 0.031, 0.088, and 0.031 μ M for **54**. This agent was active in several PDGF-dependent cellular assays including PDGFR autophosphorylation and inhibition of tumor cell growth. In addition, **54** exhibited meaningful *in vivo* activity against three tumor model systems. Studies to evaluate the potential use of **54** as an anticancer therapeutic agent are continuing.

Experimental Section

General Methods. Melting points were determined with a Thomas-Hoover capillary melting point apparatus or a MEL-TEMP melting point apparatus and are uncorrected. ¹H NMR spectra were recorded using a Varian Unity 400-MHz spectrometer. Chemical shifts are in parts per million (δ) referenced to Me₄Si. Chemical ionization mass spectra (CI) were recorded on a VG Trio 2 mass spectrometer instrument using a reagent gas of 1% NH₃ in CH₄. Atmospheric pressure chemical ionization (APCI) and electrospray mass (ES) spectra were recorded using a VG Trio 2000 mass spectrometer. Flash chromatography was performed with silica gel 60 (230–400 mesh; E. Merck Darmstadt). Combustion analyses (CHN) were performed by the Parke-Davis Pharmaceutical Research Analytical Department or Robertson Microlit (Madison, NJ).

General Procedure for the Preparation of 8-*N*-Substituted-2-amino-6-(2,6-dichlorophenyl)-8*H*-pyrido[2,3-*d*]pyrimidin-7-ones from 4, Used To Prepare 5–10, 12, and 15. 2-Amino-6-(2,6-dichlorophenyl)-8-ethyl-8*H*-pyrido[2,3-*d*]pyrimidin-7-one (**5**). To a suspension of NaH (60% in mineral oil, 27 mg) in 5 mL of DMF was added **4** (172 mg, 0.56 mmol). The mixture was heated at 50 °C for 1 h resulting in a clear solution. Ethyl iodide (60 μ L, 0.75 mmol) was added, and the solution was stirred at 50 °C for 3.5 h, then cooled to room temperature, and poured onto 30 mL of ice water. The resulting precipitate was collected by filtration, washed with water, and purified by flash chromatography, eluting with 1:1 hexane/EtOAc to provide 104 mg (55%) of **5**; mp 207–209 °C; ¹H NMR (DMSO-*d*₆) δ 1.20 (t, *J* = 7 Hz, 3H), 4.30 (q, *J* = 7 Hz, 2H), 7.41–7.46 (m, 3H), 7.57 (d, *J* = 8 Hz, 2H), 7.70 (s, 1H), 8.65 (s, 1H); MS (CI) *m/z* 335 (M + 1). Anal. (C₁₅H₁₂Cl₂N₄O) C, H, N.

2-Amino-6-(2,6-dichlorophenyl)-8-propyl-8*H*-pyrido[2,3-*d*]pyrimidin-7-one (**6**): yield 68%; mp 196–197 °C; ¹H NMR (DMSO-*d*₆) δ 0.89 (t, *J* = 7 Hz, 3H), 1.66 (m, 2H), 4.23 (t, *J* = 7 Hz, 2H), 7.41–7.46 (m, 3H), 7.57 (d, *J* = 8 Hz, 2H), 7.77 (s, 1H), 8.64 (s, 1H); MS (CI) *m/z* 351 (M + 1). Anal. (C₁₆H₁₄Cl₂N₄O) C, H, N.

2-Amino-8-butyl-6-(2,6-dichlorophenyl)-8*H*-pyrido[2,3-*d*]pyrimidin-7-one (**7**): yield 64%; mp 202–205 °C; ¹H NMR (DMSO-*d*₆) δ 0.91 (t, *J* = 7 Hz, 3H), 1.31 (m, 2H), 1.62 (m, 2H), 4.26 (t, *J* = 7 Hz, 2H), 7.39 (br s, 2H), 7.44 (t, *J* = 8 Hz, 1H), 7.56 (d, *J* = 8 Hz, 2H), 7.76 (s, 1H), 8.63 (s, 1H); MS (CI) *m/z* 365 (M + 1). Anal. (C₁₇H₁₆Cl₂N₄O·0.08EtOAc) C, H, N.

2-Amino-6-(2,6-dichlorophenyl)-8-isobutyl-8*H*-pyrido[2,3-*d*]pyrimidin-7-one (**8**): yield 51%; mp 193–195 °C; ¹H NMR (DMSO-*d*₆) δ 0.87 (t, *J* = 6.5 Hz, 6H), 2.24 (m, 1H), 4.13 (d, *J* = 7.5 Hz, 2H), 7.37 (br s, 2H), 7.44 (t, *J* = 7.5 Hz, 1H), 7.57 (d, *J* = 7.5 Hz, 2H), 7.77 (s, 1H), 8.64 (s, 1H); MS (CI) *m/z* 365 (M + 1). Anal. (C₁₇H₁₆Cl₂N₄O) C, H, N.

[2-Amino-6-(2,6-dichlorophenyl)-7-oxo-7*H*-pyrido[2,3-*d*]pyrimidin-8-yl]acetic acid methyl ester (**9**): yield 61%; mp 188–190 °C; ¹H NMR (DMSO-*d*₆) δ 3.68 (s, 3H), 5.03 (s, 2H), 7.50 (br s, 2H), 7.45 (t, *J* = 8 Hz, 1H), 7.57 (d, *J* = 8 Hz,

2H), 7.87 (s, 1H), 8.69 (s, 1H); MS (CI) *m/z* 381 (M + 1). Anal. (C₁₆H₁₂Cl₂N₄O₃) C, H, N.

[2-Amino-6-(2,6-dichlorophenyl)-7-oxo-7*H*-pyrido[2,3-*d*]pyrimidin-8-yl]acetic acid *tert*-butyl ester (**10**): yield 30%; mp 171–173 °C; ¹H NMR (DMSO-*d*₆) δ 1.38 (s, 9H), 4.88 (s, 2H), 7.47 (br s, 2H), 7.45 (t, *J* = 8 Hz, 1H), 7.57 (d, *J* = 8 Hz, 2H), 7.85 (s, 1H), 8.68 (s, 1H); MS (CI) *m/z* 423 (M + 1). Anal. (C₁₉H₁₈Cl₂N₄O₃) C, H, N.

[2-Amino-6-(2,6-dichlorophenyl)-7-oxo-7*H*-pyrido[2,3-*d*]pyrimidin-8-yl]acetic Acid (**11**). To a solution of **10** (157 mg, 0.37 mmol) in 4 mL of CH₂Cl₂ was added 2 mL of trifluoroacetic acid. The solution was stirred at room temperature for 5 h and then concentrated. The resultant oil was partitioned between CH₂Cl₂ and brine. The aqueous layer was washed with EtOAc, and the organic layers were combined, dried over MgSO₄, filtered, and concentrated to give a gummy solid. Diethyl ether was added, and the resultant precipitate was collected. Hexane was added to the filtrate, and the resultant precipitate was collected. The solids were combined to give 71 mg (52%) of **11** that was >97% pure by HPLC: mp 297–300 °C dec; ¹H NMR (DMSO-*d*₆) δ 4.93 (s, 2H), 7.45 (t, *J* = 8 Hz, 1H), 7.48 (br s, 2H), 7.57 (d, *J* = 8 Hz, 2H), 7.85 (s, 1H), 8.68 (s, 1H); MS (CI) *m/z* 367 (M + 1). Anal. (C₁₅H₁₀Cl₂N₄O₃) Calcd: C, 49.34; H, 2.76; N, 15.34. Found: C, 46.01; H, 2.77; N, 13.28.

2-Amino-8-benzyl-6-(2,6-dichlorophenyl)-8*H*-pyrido[2,3-*d*]pyrimidin-7-one (**12**): yield 43%; mp 220–222 °C; ¹H NMR (DMSO-*d*₆) δ 5.48 (s, 2H), 7.21–7.35 (complex m, 5H), 7.43–7.47 (complex m, 3H), 7.57 (d, *J* = 8 Hz, 2H), 7.84 (s, 1H), 8.67 (s, 1H); MS (CI) *m/z* 399 (M + 1). Anal. (C₂₀H₁₄Cl₂N₄O) C, H, N.

2-Amino-6-(2,6-dichlorophenyl)-8-(pyridin-4-ylmethyl)-8*H*-pyrido[2,3-*d*]pyrimidin-7-one (**13**). To a suspension of NaH (60% in mineral oil, 32 mg) in 6 mL of DMF was added **4** (200 mg, 0.65 mmol), and the mixture was heated to 70 °C. In a second flask containing Et₃N (220 μ L, 1.59 mmol) in 4 mL of DMF was added 4-picoyl chloride hydrochloride (137 mg, 0.84 mmol). This dark red mixture was added to the solution of the sodium salt of **4**. The mixture was heated to 70 °C, then cooled to room temperature, and poured into 20 mL of ice water. The resulting precipitate was collected by filtration and washed with water followed by 10% MeOH in EtOAc to provide 96 mg of crude product. The organic filtrate was concentrated to provide an additional 77 mg of crude **13**. An analytical sample was obtained by flash chromatography, eluting with a gradient of EtOAc to 10% MeOH in EtOAc to provide **13**; mp 268–270 °C dec; ¹H NMR (DMSO-*d*₆) δ 5.49 (s, 2H), 7.22 (dd, *J* = 4.5, 1.5 Hz, 2H), 7.44 (t, *J* = 8 Hz, 1H), 7.48 (br s, 2H), 7.57 (d, *J* = 8 Hz, 2H), 7.90 (s, 1H), 8.49 (dd, *J* = 4.5, 1.5 Hz, 2H), 8.70 (s, 1H); MS (CI) *m/z* 400 (M + 1). Anal. (C₁₉H₁₃Cl₂N₅O) C, H, N.

2-Amino-6-(2,6-dichlorophenyl)-8-(3-dimethylamino-propyl)-8*H*-pyrido[2,3-*d*]pyrimidin-7-one (**14**). To a suspension of NaH (60% in mineral oil, 50 mg) in 8 mL of DMF was added **4** (319 mg, 1.04 mmol). The mixture was heated at 70 °C for 1.5 h resulting in a clear solution. In a second flask containing NaH (60% in mineral oil, 68 mg) in 6 mL of DMF was added 3-dimethylaminopropyl chloride hydrochloride (248 mg, 1.56 mmol). This suspension was stirred at room temperature for 30 min, then heated at 70 °C for 10 min, and added to the solution of the sodium salt of **4**. The resultant suspension was heated at 70 °C for 3 h, then cooled to room temperature, and filtered, washing with EtOAc. The filtrate was concentrated, and EtOAc and hexane were added. The resulting solid was collected by filtration to provide 216 mg (53%) of **14**; mp 136–141 °C; ¹H NMR (DMSO-*d*₆) δ 1.76 (pent, *J* = 7 Hz, 2H), 2.11 (s, 6H), 2.26 (t, *J* = 7 Hz, 2H), 4.29 (t, *J* = 7 Hz, 2H), 7.39 (br s, 2H), 7.44 (t, *J* = 8 Hz, 1H), 7.57 (d, *J* = 8 Hz, 2H), 7.77 (s, 1H), 8.64 (s, 1H); MS (CI) *m/z* 394 (M + 1). Anal. (C₁₈H₁₆Cl₂N₅O) C, H, N.

2-Amino-8-(3-benzoyloxypropyl)-6-(2,6-dichlorophenyl)-8*H*-pyrido[2,3-*d*]pyrimidin-7-one (**15**): yield 71%; mp undefined (softens at 63 °C); ¹H NMR (DMSO-*d*₆) δ 1.94 (pent, *J* = 7 Hz, 2H), 3.50 (t, *J* = 7 Hz, 2H), 4.36 (t, *J* = 7 Hz, 2H),

4.42 (s, 2H), 7.23–7.34 (complex m, 5H), 7.40 (br s, 2H), 7.44 (t, $J = 8$ Hz, 1H), 7.56 (d, $J = 8$ Hz, 2H), 7.76 (s, 1H), 8.63 (s, 1H); MS (CI) m/z 457 ($M + 1$). Anal. ($C_{23}H_{20}Cl_2N_4O_2 \cdot 0.2EtOAc$) Calcd: C, 60.44; H, 4.60; N, 11.85. Found: C, 60.90; H, 4.77; N, 11.91.

2-Amino-6-(2,6-dichlorophenyl)-8-(3-hydroxypropyl)-8H-pyrido[2,3-*d*]pyrimidin-7-one (16). A solution of 15 (379 mg, 0.83 mmol) in 75 mL of acetic acid was treated with increasing amounts of 5–20% palladium on carbon over 12 h. The catalyst was filtered off, and the filtrate was concentrated. The residue was partitioned between EtOAc and 1 N NaOH. The organic layer was dried over $MgSO_4$, filtered, and concentrated. Flash chromatography gave 94 mg (31%) of 16: mp undefined (softens at 98 °C); 1H NMR (DMSO- d_6) δ 1.78 (pent, $J = 7$ Hz, 2H), 3.45 (dt, $J = 7, 5.5$ Hz, 2H), 4.31 (t, $J = 7$ Hz, 2H), 4.46 (t, $J = 5.5$ Hz, 1H), 7.42 (br s, 2H), 7.44 (t, $J = 8$ Hz, 1H), 7.57 (d, $J = 8$ Hz, 2H), 7.78 (s, 1H), 8.65 (s, 1H); MS (CI) m/z 367 ($M + 1$). Anal. ($C_{18}H_{14}Cl_2N_4O_2 \cdot 0.2 H_2O$) C, H, N.

8-Methyl-2-methylsulfanyl-6-phenyl-8H-pyrido[2,3-*d*]pyrimidin-7-ylideneamine (18). To a solution of 17 (1.50 g, 8.18 mmol) and phenylacetone nitrile (1.2 mL, 10.36 mmol) in 10 mL of DMF was added K_2CO_3 (6.60 g, 47.83 mmol). The mixture was heated at 125 °C for 1.5 h. The hot mixture was filtered, and the solid was washed with DMF. Water was added to the filtrate, and the black solid was removed by filtration. The crystals that formed in the filtrate were collected to give 449 mg (19%) of 18: mp 150–151 °C; 1H NMR (DMSO- d_6) δ 2.51 (s, 3H), 3.57 (s, 3H), 7.28 (s, 1H), 7.37–7.52 (m, 6H), 8.48 (s, 1H); MS (CI) m/z 283 ($M + 1$). Anal. ($C_{15}H_{14}N_4S$) C, H, N.

8-Methyl-2-methylsulfanyl-6-phenyl-8H-pyrido[2,3-*d*]pyrimidin-7-one (20). A mixture of 18 (745 mg, 2.64 mmol) and 5.5 mL of Ac_2O was heated at reflux for 30 min. The cooled solution was mixed with diethyl ether and concentrated. The solid was washed with diethyl ether to provide 718 mg (84%) of the *N*-acetylamine derivative which was refluxed with concentrated HCl for 5 min. The resulting solid was collected and washed with water and 2-propanol to give 512 mg (87%) of 20 that was not purified.

8-Methyl-2-methylsulfonyl-6-thiophene-3-yl-8H-pyrido[2,3-*d*]pyrimidin-7-one (22). To a solution of 20 (206 mg, 0.73 mmol) in 30 mL of $CHCl_3$ was added 50–60% *m*-chloroperbenzoic acid (600 mg, 1.74–2.09 mmol). The solution was stirred at room temperature for 2 h, then washed with saturated $NaHCO_3$, dried over $MgSO_4$, and concentrated. EtOAc was added to the residue followed by hexane. The solid was collected and washed with 4:1 hexane/EtOAc to give 139 mg (60%) of 22: mp 191–193 °C; 1H NMR (DMSO- d_6) δ 3.44 (s, 3H), 3.68 (s, 3H), 7.39–7.47 (m, 3H), 7.69 (d, $J = 2$ Hz, 2H), 8.26 (s, 1H), 9.26 (s, 1H); MS (CI) m/z 316 ($M + 1$). Anal. ($C_{15}H_{13}N_3O_3S$) C, H, N.

8-Methyl-6-phenyl-2-phenylamino-8H-pyrido[2,3-*d*]pyrimidin-7-one (24). A mixture of 22 (174 mg, 0.55 mmol) and aniline (440 mg, 5.30 mmol) was heated at 175 °C for 10 min. The reaction was cooled to room temperature and directly purified by flash chromatography eluting with EtOAc to give 62 mg (34%) of 24: mp 210–211 °C; 1H NMR (DMSO- d_6) δ 3.68 (s, 3H), 7.04 (t, $J = 7$ Hz, 1H), 7.34–7.39 (m, 3H), 7.44 (d, $J = 7$ Hz, 2H), 7.68 (d, $J = 7$ Hz, 2H), 7.85 (d, $J = 8$ Hz, 2H), 8.03 (s, 1H), 8.85 (s, 1H), 10.16 (s, 1H); MS (CI) m/z 329 ($M + 1$). Anal. ($C_{20}H_{16}N_4O \cdot 0.25H_2O$) C, H, N.

8-Methyl-6-phenyl-2-(pyridin-4-ylamino)-8H-pyrido[2,3-*d*]pyrimidin-7-one (26). A mixture of 22 (146 mg, 0.46 mmol) and 4-aminopyridine (262 mg, 2.78 mmol) was heated at 175 °C for 10 min. The reaction was cooled to room temperature and directly purified by flash chromatography eluting with EtOAc to give 77 mg (51%) of 26: mp 244–245 °C; HPLC purity >99%, column VYDAC C_{18} 218TP54, mobile phase gradient 10–76% 0.1% TFA in acetonitrile to 0.1% TFA in water; 1H NMR (DMSO- d_6) δ 3.67 (s, 3H), 7.31–7.42 (m, 3H), 7.63 (t, $J = 7$ Hz, 2H), 7.78 (d, $J = 5$ Hz, 2H), 8.03 (s, 1H), 8.38 (d, $J = 5$ Hz, 2H), 8.88 (s, 1H), 10.49 (s, 1H); MS (CI) m/z 330 ($M + 1$). Anal. ($C_{18}H_{15}N_5O$) Calcd: C, 69.29; H, 4.59; N, 21.26. Found: C, 66.79; H, 5.05; N, 19.04.

2-[4-(2-Diethylaminoethoxy)phenylamino]-8-methyl-6-phenyl-8H-pyrido[2,3-*d*]pyrimidin-7-one (28). A mixture of 22 (197 mg, 0.63 mmol) and 4-(2-diethylaminoethoxy)aniline (260 mg, 1.25 mmol) was heated at 175 °C for 5 min. The reaction was cooled to room temperature, and the residue was purified by flash chromatography eluting with 5% MeOH in $CHCl_3$ to give 30 mg (11%) of 28: mp 157–158 °C; 1H NMR (DMSO- d_6) δ 0.98 (t, $J = 7$ Hz, 6H), 2.55 (q, $J = 7$ Hz, 4H), 2.77 (t, $J = 6$ Hz, 2H), 3.66 (s, 3H), 4.00 (t, $J = 6$ Hz, 2H), 6.94 (d, $J = 9$ Hz, 2H), 7.36–7.45 (m, 3H), 7.67–7.72 (m, 4H), 8.00 (s, 1H), 8.80 (s, 1H), 10.00 (s, 1H); MS (CI) m/z 444 ($M + 1$). Anal. ($C_{28}H_{28}N_5O_2 \cdot 0.5H_2O$) C, H, N.

8-Methyl-2-methylsulfanyl-6-thiophene-3-yl-8H-pyrido[2,3-*d*]pyrimidin-7-ylideneamine (19). To a solution of 17 (10.0 g, 54.6 mmol) and 3-thiopheneacetone nitrile (13.52 g, 109.2 mmol) in 100 mL of DMF was added K_2CO_3 (37.7 g, 273.0 mmol). The mixture was heated at 125 °C for 1.5 h. The hot mixture was filtered, and the solid was washed with EtOAc. The solid that appeared in the filtrate was collected and washed with 10% EtOAc in hexane followed by acetone to give 10.87 g (69%) of 19: mp 146–147 °C; 1H NMR (DMSO- d_6) δ 2.57 (s, 3H), 3.64 (s, 3H), 7.27–7.28 (m, 1H), 7.41 (s, 1H), 7.71 (s, 1H), 7.76–7.78 (m, 2H), 8.53 (s, 1H); MS (CI) m/z 289 ($M + 1$). Anal. ($C_{13}H_{12}N_4S_2$) C, H, N.

8-Methyl-2-methylsulfanyl-6-thiophene-3-yl-8H-pyrido[2,3-*d*]pyrimidin-7-one (21). A mixture of 19 (7.39 g, 25.7 mmol) and 50 mL of Ac_2O was heated at reflux for 15 min. The cooled solution was mixed with 200 mL of diethyl ether and concentrated. The solid was washed with diethyl ether to provide 6.27 g (74%) of the *N*-acetylamine derivative which was refluxed with concentrated HCl for 5 min. The resulting solid was collected, washed with water, and purified by flash chromatography eluting with CH_2Cl_2 to give 2.99 g (55%) of 21: mp 176–177 °C; 1H NMR (DMSO- d_6) δ 2.63 (s, 3H), 3.70 (s, 3H), 7.64–7.70 (m, 2H), 8.35–8.36 (m, 1H), 8.40 (s, 1H), 8.91 (s, 1H); MS (CI) m/z 290 ($M + 1$). Anal. ($C_{13}H_{11}N_3OS_2$) C, H, N.

8-Methyl-2-methylsulfonyl-6-thiophene-3-yl-8H-pyrido[2,3-*d*]pyrimidin-7-one (23). To a solution of 21 (2.01 g, 6.95 mmol) in 150 mL of $CHCl_3$ was added 50–60% *m*-chloroperbenzoic acid (5.50 g, 16.0–19.2 mmol). The solution was stirred at room temperature for 1 h, then washed with saturated $NaHCO_3$, dried over $MgSO_4$, and concentrated. The solid was purified by flash chromatography eluting with CH_2Cl_2 to give 1.81 g (81%) of 23: 1H NMR (DMSO- d_6) δ 3.50 (s, 3H), 3.76 (s, 3H), 7.70–7.76 (m, 2H), 8.48–8.49 (m, 1H), 8.58 (s, 1H), 9.28 (s, 1H); MS (CI) m/z 322 ($M + 1$).

8-Methyl-2-phenylamino-6-thiophene-3-yl-8H-pyrido[2,3-*d*]pyrimidin-7-one (25). A mixture of 23 (214 mg, 0.67 mmol) and aniline (400 mg, 4.82 mmol) was heated at 150 °C for 15 min. The reaction was cooled to room temperature and directly purified by flash chromatography eluting with 5% MeOH in $CHCl_3$ to give 67 mg (30%) of 25: mp 237–238 °C; 1H NMR (DMSO- d_6) δ 3.70 (s, 3H), 7.04 (t, $J = 7$ Hz, 1H), 7.36 (t, $J = 8$ Hz, 2H), 7.61–7.68 (m, 2H), 7.84 (d, $J = 8$ Hz, 2H), 8.27 (s, 1H), 8.30 (s, 1H), 8.83 (s, 1H), 10.15 (s, 1H); MS (CI) m/z 335 ($M + 1$). Anal. ($C_{18}H_{14}N_4OS \cdot 0.25H_2O$) C, H, N.

8-Methyl-2-(pyridin-4-ylamino)-6-thiophene-3-yl-8H-pyrido[2,3-*d*]pyrimidin-7-one (27). A mixture of 23 (190 mg, 0.59 mmol) and 4-aminopyridine (170 mg, 1.81 mmol) was heated at 175 °C for 5 min. The reaction was cooled to room temperature and directly purified by flash chromatography eluting with 5% MeOH in $CHCl_3$ to give 61 mg (31%) of 27: mp 310–311 °C; 1H NMR (DMSO- d_6) δ 3.74 (s, 3H), 6.69–7.63 (m, 2H), 7.84 (d, $J = 6$ Hz, 2H), 8.30–8.45 (m, 4H), 8.92 (s, 1H), 10.54 (s, 1H); MS (CI) m/z 336 ($M + 1$). Anal. ($C_{17}H_{13}N_5OS \cdot 0.25H_2O$) C, H, N.

2-[4-(2-Diethylaminoethoxy)phenylamino]-8-methyl-6-thiophene-3-yl-8H-pyrido[2,3-*d*]pyrimidin-7-one (29). A mixture of 23 (132 mg, 0.43 mmol) and 4-(2-diethylaminoethoxy)aniline (178 mg, 0.86 mmol) was heated at 175 °C for 5 min. The reaction was cooled to room temperature, and the residue was purified by flash chromatography eluting with 5% MeOH in $CHCl_3$ to give 40 mg (22%) of 29: mp 190–192 °C;

¹H NMR (DMSO-*d*₆) δ 0.98 (t, *J* = 7 Hz, 6H), 2.56 (q, *J* = 7 Hz, 4H), 2.77 (t, *J* = 6 Hz, 2H), 3.67 (s, 3H), 4.00 (t, *J* = 6 Hz, 2H), 6.94 (d, *J* = 9 Hz, 2H), 7.60–7.72 (m, 4H), 8.25 (t, *J* = 1 Hz, 1H), 8.27 (s, 1H), 8.78 (s, 1H), 9.99 (s, 1H); MS (CI) *m/z* 450 (M + 1). Anal. (C₂₄H₂₇N₅O₂S·H₂O) C, H, N.

2-Amino-8-methyl-6-thiophene-3-yl-8H-pyrido[2,3-*d*]pyrimidin-7-one (30). A mixture of 21 (170 mg, 0.59 mmol), ammonia, and MeOH was sealed in a glass tube and heated at 100 °C for 86 h. The solution was concentrated and purified by flash chromatography eluting with EtOAc. Recrystallization from EtOAc and hexane gave 115 mg (76%) of 30: mp 241–242 °C; ¹H NMR (DMSO-*d*₆) δ 3.59 (s, 3H), 7.33 (s, 2H), 7.58–7.64 (m, 2H), 8.21 (s, 1H), 8.22 (s, 1H), 8.63 (s, 1H); MS (CI) *m/z* 259 (M + 1). Anal. (C₁₂H₁₀N₄OS) C, H, N.

8-Methyl-2-methylamino-6-thiophene-3-yl-8H-pyrido[2,3-*d*]pyrimidin-7-one (31). A mixture of 21 (125 mg, 0.43 mmol), methylamine, and DMF was sealed in a glass tube and heated at 110 °C for 40 h. The solution was concentrated and the solid washed with diethyl ether to provide 61 mg (52%) of 31: mp 263–265 °C; ¹H NMR (DMSO-*d*₆) δ 2.92 (d, *J* = 4 Hz, 3H), 3.65 and 3.56 (two peaks, 3H), 7.58–7.65 (m, 2H), 7.83 (br s, 1H), 8.22 (s, 2H), 7.76 and 8.63 (two peaks, 1H); MS (CI) *m/z* 273 (M + 1). Anal. (C₁₃H₁₂N₄OS·0.25H₂O) C, H, N.

4-Methylamino-2-methylsulfanyl-pyrimidine-5-carbonitrile (32). Methylamine gas was bubbled through a 5 °C solution of 4-chloro-2-methylsulfanyl-pyrimidine-5-carbonitrile (32.0 g, 170 mmol) in 700 mL of diethyl ether. The reaction mixture was stirred at room temperature overnight. Additional methylamine gas was added, and the mixture stirred at room temperature overnight. The solids were collected and suspended in water. The suspension was filtered to provide 25.8 g (85%) of product. Recrystallization from EtOAc gave an analytical sample of 32: mp 189–190 °C; ¹H NMR (CDCl₃) δ 2.54 (s, 3H), 3.12 (d, *J* = 5 Hz, 3H), 5.50 (br s, 1H), 8.82 (s, 1H). Anal. (C₇H₈N₄S) C, H, N.

4-Methylamino-2-phenylamino-pyrimidine-5-carbonitrile (33). A suspension of 32 (10 g, 55.5 mmol) in 30 mL of aniline containing 4 drops of concentrated HCl was heated to 170 °C for 1 h. Upon removal of the heat source a solid rapidly formed. While the reaction mixture was still warm, 30 mL of MeOH was added. The thick mixture was cooled to room temperature and the solid collected washing with additional MeOH to provide 8.73 g (70%) of product. Recrystallization from EtOAc and MeOH gave an analytical sample of 33: mp 275–277 °C; ¹H NMR (DMSO-*d*₆) δ 2.91 (d, *J* = 4 Hz, 3H), 6.99 (t, *J* = 7.5 Hz, 1H), 7.29 (t, *J* = 8 Hz, 2H), 7.71 (br s, 1H), 7.76 (d, *J* = 8 Hz, 2H), 8.33 (s, 1H), 9.81 (br s, 1H). Anal. (C₁₂H₁₁N₅) C, H, N.

4-Methylamino-2-phenylamino-pyrimidine-5-carbaldehyde (34). To a 0 °C solution of 33 (4.5 g, 20.00 mmol) in 100 mL of THF was added 20 mL of a 1 M solution of diisobutylaluminum hydride in CH₂Cl₂. The ice bath was removed, and an additional 20 mL of a 1 M solution of diisobutylaluminum hydride in CH₂Cl₂ was added. After 30 min an additional 5 mL of a 1 M solution of diisobutylaluminum hydride in CH₂Cl₂ was added. After 30 min the reaction was quenched by the dropwise addition of 30 mL of MeOH. This mixture was partitioned between 200 mL of EtOAc and 140 mL of 1 N HCl. The layers were separated, and the acid layer was treated with 150 mL of 1 N NaOH and extracted with EtOAc. The organic layer was dried over MgSO₄, filtered, and concentrated to give 2.20 g of a 3:1 mixture of 34:33 by NMR. Purification by flash chromatography eluting with 1:1 hexane/EtOAc gave an analytical sample of 34: mp 210–212 °C; ¹H NMR (DMSO-*d*₆) δ 3.02 (d, *J* = 5 Hz, 3H), 7.01 (t, *J* = 7.5 Hz, 1H), 7.31 (t, *J* = 8 Hz, 2H), 7.83 (d, *J* = 8 Hz, 2H), 8.46 (s, 1H), 8.59 (br s, 1H), 9.55 (s, 1H), 9.97 (br s, 1H). Anal. (C₁₂H₁₃N₄O) C, H, N.

(7-Imino-8-methyl-6-thiophene-2-yl-7,8-dihydropyrido[2,3-*d*]pyrimidin-2-yl)phenylamine (35). To a suspension of NaH (21 mg of a 60% suspension of NaH in mineral oil) in 5 mL of 2-ethoxyethanol were added 2-thiopheneacetonitrile (53 μL, 0.50 mmol) and 34 (101 mg, 0.44 mmol). The reaction mixture was heated at reflux for 17 h. The solution was cooled

to room temperature, and the resulting precipitate was collected and washed with 1:1 hexane/EtOAc. The solid was purified by flash chromatography eluting with 10% MeOH in EtOAc to give 60 mg (41%) of 35: mp 241–242 °C; ¹H NMR (DMSO-*d*₆) δ 3.66 (s, 3H), 7.00 (t, *J* = 7 Hz, 1H), 7.21 (two peaks, 1H), 7.34 (t, *J* = 7 Hz, 2H), 7.44 (two peaks, 1H), 7.73–7.79 (m, 4H), 7.81 (two peaks, 1H), 8.52 (s, 1H), 9.92 (s, 1H); MS (CI) *m/z* 334 (M + 1). Anal. (C₁₈H₁₅N₅S) C, H, N.

8-Methyl-2-phenylamino-6-thiophene-2-yl-8H-pyrido[2,3-*d*]pyrimidin-7-one (37). A mixture of 35 (283 mg, 0.85 mmol) and 10 mL of Ac₂O was heated at reflux for 30 min. The solution was concentrated, and the resulting *N*-acetyl-imine derivative was dissolved in 10 mL of concentrated HCl and heated at reflux for 5 min. The reaction was cooled to room temperature, and the crude product was collected and purified by flash chromatography eluting with 5% MeOH in CHCl₃ to provide 74 mg (26%) of 37: mp 235–236 °C; ¹H NMR (DMSO-*d*₆) δ 3.72 (s, 3H), 7.04 (t, *J* = 7 Hz, 1H), 7.16 (d, *J* = 5 Hz, 1H), 7.36 (t, *J* = 8 Hz, 2H), 7.59 (d, *J* = 5 Hz, 1H), 7.84 (d, *J* = 8 Hz, 2H), 8.45 (s, 1H), 8.87 (s, 1H), 10.19 (s, 1H); MS (CI) *m/z* 335 (M + 1). Anal. (C₁₉H₁₄N₄OS·0.25H₂O) C, H, N.

[6-(Biphenyl-4-yl)-7-imino-8-methyl-8H-pyrido[2,3-*d*]pyrimidin-2-yl]phenylamine (36). To a suspension of NaH (110 mg of a 60% suspension of NaH in mineral oil) in 20 mL of 2-ethoxyethanol were added 4-biphenylacetonitrile (516 mg, 2.67 mmol) and 34 (538 mg, 2.36 mmol). The reaction mixture was heated at reflux for 17 h and then cooled to room temperature. The precipitate was collected and washed with 1:1 hexane/EtOAc to give 826 mg (87%) of 36: mp 259–260 °C; ¹H NMR (DMSO-*d*₆) δ 3.69 (s, 3H), 7.00 (t, *J* = 7 Hz, 1H), 7.32–7.41 (m, 5H), 7.42–7.54 (m, 4H), 7.75 (d, *J* = 8 Hz, 2H), 7.82 (d, *J* = 8 Hz, 4H), 8.51 (s, 1H), 9.89 (s, 1H); MS (CI) *m/z* 404 (M + 1). Anal. (C₂₆H₂₁N₅·0.3H₂O) C, H, N.

6-(Biphenyl-4-yl)-8-methyl-2-phenylamino-8H-pyrido[2,3-*d*]pyrimidin-7-one (38). A mixture of 36 (261 mg, 0.65 mmol) and 12 mL of Ac₂O was heated at reflux for 30 min. The solution was concentrated, and the resulting *N*-acetyl-imine derivative was dissolved in 10 mL of concentrated HCl and heated at reflux for 5 min. The reaction was cooled to room temperature, and the solid was collected and purified by flash chromatography eluting with 5% MeOH in CHCl₃ to provide 152 mg (58%) of 38: mp 263–264 °C; ¹H NMR (DMSO-*d*₆) δ 3.70 (s, 3H), 7.04 (t, *J* = 8 Hz, 1H), 7.35–7.41 (m, 3H), 7.49 (t, *J* = 8 Hz, 2H), 7.73–7.76 (m, 4H), 7.80–7.86 (m, 4H), 8.11 (s, 1H), 8.87 (s, 1H), 10.17 (s, 1H); MS (CI) *m/z* 405 (M + 1). Anal. (C₂₆H₂₀N₄O·0.80H₂O) C, H, N.

4-Ethylamino-2-methylsulfanyl-5-pyrimidinecarboxylate Ethyl Ester (40). To a room-temperature solution of 39 (10.00 g, 43.10 mmol) in 150 mL of THF was added Et₃N (18.5 mL, 133 mmol) followed by 9 mL of a 70% aqueous solution of ethylamine. The solution was stirred for 30 min, then concentrated, and partitioned between CHCl₃ and saturated aqueous NaHCO₃. The organic layer was dried over MgSO₄, filtered, and concentrated to provide 9.32 g (90%) of 40 as an oil: ¹H NMR (DMSO-*d*₆) δ 1.17 (t, *J* = 7 Hz, 3H), 1.30 (t, *J* = 7 Hz, 3H), 2.48 (s, 3H), 3.51 (dq, *J* = 7, 5 Hz, 2H), 4.27 (q, *J* = 7 Hz, 2H), 8.31 (t, *J* = 5 Hz, 1H), 8.53 (s, 1H); MS (CI) *m/z* 242 (M + 1). Anal. (C₁₀H₁₅N₃O₂S) C, H, N.

4-Ethylamino-2-methylsulfanyl-pyrimidine-5-carboxylic Acid Ethyl Ester (41). To a room-temperature solution of 40 (2.011 g, 8.34 mmol) in 70 mL of CHCl₃ was added (±)-*trans*-2-phenylsulfonyl-3-phenyloxaziridine (2.70 g, 10.34 mmol). The solution was stirred at room temperature for 7 h and then concentrated. The residue was purified by flash chromatography eluting with a gradient of EtOAc to 3% MeOH in EtOAc to provide 2.07 g (97%) of 41: mp 54–56 °C; ¹H NMR (DMSO-*d*₆) δ 1.18 (t, *J* = 7 Hz, 3H), 1.33 (t, *J* = 7 Hz, 3H), 2.84 (s, 3H), 3.55 (dq, *J* = 7, 5.5 Hz, 2H), 4.33 (q, *J* = 7 Hz, 2H), 8.59 (t, *J* = 5.5 Hz, 1H), 8.79 (s, 1H); MS (CI) *m/z* 258 (M + 1). Anal. (C₁₀H₁₅N₃O₃S) C, H, N.

4-Ethylamino-2-phenylamino-pyrimidine-5-carboxylic Acid Ethyl Ester (42). A solution of 41 (5.38 g, 20.9 mmol) in 4 mL of aniline was heated at 130 °C for 1 h. The solution was cooled to room temperature resulting in a solid; 20 mL of

1:1 hexane/EtOAc was added, and the white solid was collected by filtration providing 1.96 g (33%) of 42. The filtrate was concentrated and purified by flash chromatography eluting with 3:1 hexane/EtOAc to give an additional 257 mg of 42: mp 145–147 °C; ¹H NMR (DMSO-*d*₆) δ 1.21 (t, *J* = 7 Hz, 3H), 1.30 (t, *J* = 7 Hz, 3H), 3.52 (dq, *J* = 7, 5 Hz, 2H), 4.24 (q, *J* = 7 Hz, 2H), 6.98 (t, *J* = 7 Hz, 1H), 7.29 (t, *J* = 8 Hz, 2H), 7.79 (d, *J* = 8 Hz, 2H), 8.23 (t, *J* = 5 Hz, 1H), 8.56 (s, 1H), 9.77 (br s, 1H); MS (CI) *m/z* 287 (M + 1). Anal. (C₁₅H₁₈N₄O₂) C, H, N.

(4-Ethylamino-2-phenylamino-pyrimidin-5-yl)methanol (43). A solution of 42 (109 mg, 0.38 mmol) in 6 mL of THF was added dropwise to a room temperature suspension of LAH (35 mg, 0.92 mmol) in 5 mL of THF. After 25 min, an additional 30 mg of LAH was added and stirring was continued for 30 min. The reaction was carefully quenched with 120 μL of water, 200 μL of 15% NaOH, and 300 μL of water. After stirring for 1 h, the white precipitate was removed by filtration washing with EtOAc. The filtrate was concentrated and the residue purified by chromatography eluting with EtOAc to provide 36 mg (39%) of 43: mp 174–176 °C; ¹H NMR (DMSO-*d*₆) δ 1.19 (t, *J* = 7 Hz, 3H), 3.44 (dq, *J* = 7, 5.5 Hz, 2H), 4.28 (d, *J* = 5.5 Hz, 2H), 4.93 (t, *J* = 5.5 Hz, 1H), 6.53 (t, *J* = 5.5 Hz, 1H), 6.84 (t, *J* = 7 Hz, 1H), 7.21 (t, *J* = 8 Hz, 2H), 7.74 (s, 1H), 7.77 (d, *J* = 8 Hz, 2H), 8.93 (s, 1H); MS (CI) *m/z* 245 (M + 1). Anal. (C₁₃H₁₆N₄O) C, H, N.

4-Ethylamino-2-phenylamino-pyrimidine-5-carbaldehyde (44). To a solution of 43 (173 mg, 0.71 mmol) in 15 mL of CHCl₃ was added MnO₂ (600 mg, 6.89 mmol). After stirring at room temperature overnight the mixture was filtered through a pad of Celite, washing with CHCl₃. The filtrate was concentrated to give 170 mg (99%) of 44: mp 155–157 °C; ¹H NMR (DMSO-*d*₆) δ 1.21 (t, *J* = 7 Hz, 3H), 3.53 (m, 2H), 7.02 (q, *J* = 7 Hz, 1H), 7.31 (t, *J* = 8 Hz, 2H), 7.80 (d, *J* = 8 Hz, 2H), 8.47 (s, 1H), 8.65 (br s, 1H), 9.55 (s, 1H), 9.98 (br s, 1H); MS (CI) *m/z* 243 (M + 1). Anal. (C₁₃H₁₄N₄O) C, H, N.

(8-Ethyl-7-imino-6-phenyl-7,8-dihydropyrido[2,3-*d*]pyrimidin-2-yl)phenylamine (45). To a suspension of NaH (60% in mineral oil, 44 mg) in 5 mL of 2-ethoxyethanol was added phenylacetonitrile (120 μL, 1.08 mmol). After the mixture stirred for 5 min at room temperature, 44 (230 mg, 0.95 mmol) in 2 mL of 2-ethoxyethanol was added and the reaction mixture was heated at reflux for 5 h. The solution was cooled to room temperature, and the resulting precipitate was removed by filtration and washed with water to give 305 mg of product. Recrystallization from EtOAc provided 201 mg (62%) of 45: mp 193–195 °C; ¹H NMR (DMSO-*d*₆) δ 1.29 (t, *J* = 7 Hz, 3H), 4.47 (q, *J* = 7 Hz, 2H), 7.00 (t, *J* = 7 Hz, 1H), 7.23–7.54 (complex m, 9H), 7.82 (d, *J* = 7.5 Hz, 2H), 8.48 (s, 1H), 9.86 (s, 1H); MS (CI) *m/z* 342 (M + 1). Anal. (C₂₁H₁₉N) C, H, N.

8-Ethyl-6-phenyl-2-phenylamino-8H-pyrido[2,3-*d*]pyrimidin-7-one (47). 45 (182 mg, 0.53 mmol) was added to 2 mL of Ac₂O and heated at reflux for several min. The reaction was cooled to room temperature and concentrated. The residue was heated at reflux with 4 mL of concentrated HCl for several minutes. The reaction was cooled, and 10 mL of water was added resulting in the formation of a small amount of a precipitate that was removed by filtration and washed with water. The filtrate was made basic with 1 N NaOH, and the aqueous layer was extracted with EtOAc. The organic layer was washed with brine, dried over MgSO₄, and concentrated. The residue was combined with the solid obtained previously. Purification by flash chromatography eluting with a gradient of 1:1 hexane/EtOAc to EtOAc gave 92 mg (51%) of 47: mp 202–204 °C; ¹H NMR (DMSO-*d*₆) δ 1.30 (t, *J* = 7 Hz, 3H), 4.41 (q, *J* = 7 Hz, 2H), 7.04 (t, *J* = 7 Hz, 1H), 7.34–7.46 (complex m, 5H), 7.68 (d, *J* = 7.5 Hz, 2H), 7.85 (d, *J* = 7.5 Hz, 2H), 8.02 (s, 1H), 8.85 (s, 1H), 10.16 (s, 1H); MS (CI) *m/z* 343 (M + 1). Anal. (C₂₁H₁₈N₄O) C, H, N.

(8-Ethyl-7-imino-6-thiophene-3-yl-7,8-dihydropyrido[2,3-*d*]pyrimidin-2-yl)phenylamine (46). To a suspension of NaH (60% in mineral oil, 37 mg) in 5 mL of 2-ethoxyethanol was added 3-thiopheneacetonitrile (100 μL, 0.88 mmol) followed by 44 (180 mg, 0.74 mmol) in 2 mL of 2-ethoxyethanol.

The reaction was heated at reflux for 5 h resulting in a dark brown solution. The solution was cooled to room temperature, and 3 mL of water was added. The resulting precipitate was removed by filtration and washed with water. Recrystallization from EtOAc and MeOH gave 129 mg (51%) of 46: mp 223–225 °C; ¹H NMR (DMSO-*d*₆) δ 1.29 (t, *J* = 7 Hz, 3H), 4.47 (q, *J* = 7 Hz, 2H), 7.00 (t, *J* = 7 Hz, 1H), 7.26 (dd, *J* = 5, 1.5 Hz, 1H), 7.30–7.34 (m, 3H), 7.43 (s, 1H), 7.73–7.76 (m, 2H), 7.82 (d, *J* = 7.5 Hz, 2H), 8.47 (s, 1H), 9.87 (s, 1H); MS (CI) *m/z* 348 (M + 1). Anal. (C₁₉H₁₇N₅S) C, H, N.

8-Ethyl-2-phenylamino-6-thiophene-3-yl-8H-pyrido[2,3-*d*]pyrimidin-7-one (48). Compound was prepared from 46 (173 mg, 0.50 mmol) following the procedure used to prepare 47. The product was purified by flash chromatography, eluting with a gradient of 1:1 hexane/EtOAc to all EtOAc to give 47 mg (27%) of 48: mp 225–227 °C; ¹H NMR (DMSO-*d*₆) δ 1.31 (t, *J* = 7 Hz, 3H), 4.43 (q, *J* = 7 Hz, 2H), 7.04 (t, *J* = 7 Hz, 1H), 7.36 (t, *J* = 7.5 Hz, 2H), 7.62 (dd, *J* = 5, 3 Hz, 1H), 7.66 (dd, *J* = 5, 1 Hz, 1H), 7.84 (d, *J* = 7.5 Hz, 2H), 8.27 (dd, *J* = 3, 1 Hz, 1H), 8.29 (s, 1H), 8.83 (s, 1H), 10.15 (s, 1H); MS (CI) *m/z* 349 (M + 1). Anal. (C₁₉H₁₈N₅OS) C, H, N.

8-Ethyl-2-methylsulfanyl-6-phenyl-8H-pyrido[2,3-*d*]pyrimidin-7-ylideneamine (50). To a solution of 49 (570 mg, 2.89 mmol) and phenylacetonitrile (355 mg, 3.03 mmol) in 5 mL of DMF was added K₂CO₃ (1.94 g, 14.0 mmol). The mixture was heated at 125 °C for 17 h. The hot mixture was filtered and washed with EtOAc. The filtrate was concentrated and the resulting solid purified by flash chromatography eluting with a gradient of 1:1 hexane/EtOAc to EtOAc. Recrystallization from EtOAc and hexane gave 196 mg (23%) of 50: mp 120–121 °C; ¹H NMR (DMSO-*d*₆) δ 1.24 (t, *J* = 7 Hz, 3H), 2.58 (s, 3H), 4.45 (q, *J* = 7 Hz, 2H), 7.35 (s, 1H), 7.43–7.55 (m, 6H), 8.56 (s, 1H); MS (CI) *m/z* 297 (M + 1). Anal. (C₁₆H₁₆N₄S) C, H, N.

8-Ethyl-2-methylsulfanyl-6-phenyl-8H-pyrido[2,3-*d*]pyrimidin-7-one (51). A mixture of 50 (156 mg, 5.27 mmol) and 12 mL of Ac₂O was heated at reflux for 30 min. Diethyl ether was added, and the mixture was concentrated to give a solid that was washed with diethyl ether to provide 730 mg (41%) of the *N*-acetylamine derivative. This material was refluxed with concentrated HCl for 5 min. The resulting pale yellow solid was filtered off and washed with water and then diethyl ether to give 550 mg (92%) of 51: mp 155–157 °C; ¹H NMR (DMSO-*d*₆) δ 1.27 (t, *J* = 7 Hz, 3H), 2.63 (s, 3H), 4.42 (q, *J* = 7 Hz, 2H), 7.40–7.48 (m, 3H), 7.68–7.71 (m, 2H), 8.13 (s, 1H), 8.94 (s, 1H); MS (CI) *m/z* 298 (M + 1).

8-Ethyl-2-methylsulfonyl-6-phenyl-8H-pyrido[2,3-*d*]pyrimidin-7-one (52). To a solution of 51 (1.83 g, 6.16 mmol) in 150 mL of CHCl₃ was added 50–60% *m*-chloroperoxybenzoic acid (4.88 g, 14.2–17.0 mmol). The solution was stirred at room temperature for 2 h, then washed with saturated NaHCO₃, dried over MgSO₄, and concentrated. The solid was purified by flash chromatography eluting with a gradient of 1:1 hexane/EtOAc to EtOAc to give 1.07 g (53%) of 52: mp 201–202 °C; ¹H NMR (DMSO-*d*₆) δ 1.30 (t, *J* = 7 Hz, 3H), 3.50 (s, 3H), 4.45 (q, *J* = 7 Hz, 2H), 7.46–7.53 (m, 3H), 7.73–7.75 (m, 2H), 8.31 (s, 1H), 9.32 (s, 1H); MS (CI) *m/z* 330 (M + 1). Anal. (C₁₈H₁₅N₃O₃S) C, H, N.

8-Ethyl-6-phenyl-2-phenylamino-8H-pyrido[2,3-*d*]pyrimidin-7-one (47). A mixture of 52 (119 mg, 0.36 mmol) and aniline (301 mg, 3.63 mmol) was heated at 175 °C for 10 min. The reaction was cooled to room temperature and the residue purified by flash chromatography eluting with EtOAc to give 50 mg (40%) of 47: mp 195–196 °C; ¹H NMR (DMSO-*d*₆) δ 1.31 (t, *J* = 7 Hz, 3H), 4.41 (q, *J* = 7 Hz, 2H), 7.04 (t, *J* = 7 Hz, 1H), 7.34–7.39 (m, 3H), 7.44 (t, *J* = 7 Hz, 2H), 7.68 (d, *J* = 7 Hz, 2H), 7.84 (d, *J* = 7 Hz, 2H), 8.02 (s, 1H), 8.85 (s, 1H), 10.16 (s, 1H); MS (CI) *m/z* 343 (M + 1). Anal. (C₂₁H₁₈N₄O·0.25H₂O) C, H, N.

8-Ethyl-6-phenyl-2-(pyridin-4-ylamino)-8H-pyrido[2,3-*d*]pyrimidin-7-one (53). A mixture of 52 (150 mg, 0.46 mmol) and 4-aminopyridine (289 mg, 3.07 mmol) was heated at 175 °C for 1 h. The reaction was cooled to room temperature and the residue purified by flash chromatography eluting with

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EtOAc to give 70 mg (45%) of **53**: mp 265–268 °C; ^1H NMR ($\text{DMSO}-d_6$) δ 1.27 (t, $J = 7$ Hz, 3H), 4.39 (q, $J = 7$ Hz, 2H), 7.31–7.41 (m, 3H), 7.64 (d, $J = 8$ Hz, 2H), 7.77 (d, $J = 6$ Hz, 2H), 8.02 (s, 1H), 8.38 (d, $J = 6$ Hz, 2H), 8.88 (s, 1H), 10.49 (s, 1H); MS (CI) m/z 344 ($M + 1$); HPLC purity 98%, column VYDAC C₁₈ 218TP54, mobile phase gradient 10–76% 0.1% TFA in acetonitrile to 0.1% TFA in water.

2-[4-(2-Diethylaminoethoxy)phenylamino]-8-ethyl-6-phenyl-8H-pyrido[2,3-*d*]pyrimidin-7-one (54). A mixture of **52** (129 mg, 0.39 mmol) and 4-(2-diethylaminoethoxy)aniline (163 mg, 0.78 mmol) was heated at 175 °C for 5 min. The reaction was cooled to room temperature and the residue purified by flash chromatography eluting with 5% MeOH in CHCl_3 . The fractions containing product were collected, concentrated, and washed with 1:1 hexane/EtOAc. The filtrate was concentrated, and a minimum amount of EtOAc and a large amount of hexane were added to yield a solid which was collected to give 37 mg (21%) of **54**: mp 125–126 °C; ^1H NMR ($\text{DMSO}-d_6$) δ 0.98 (t, $J = 7$ Hz, 6H), 1.29 (t, $J = 7$ Hz, 3H), 2.56 (q, $J = 7$ Hz, 4H), 2.76 (t, $J = 6$ Hz, 2H), 4.01 (t, $J = 6$ Hz, 2H), 4.38 (q, $J = 7$ Hz, 2H), 6.94 (d, $J = 9$ Hz, 2H), 7.34–7.82 (m, 7H), 7.99 (s, 1H), 8.80 (s, 1H), 10.00 (s, 1H); MS (CI) m/z 458 ($M + 1$). Anal. ($\text{C}_{27}\text{H}_{31}\text{N}_5\text{O}_2 \cdot 0.5\text{H}_2\text{O}$) C, H, N.

Recombinant Tyrosine Kinases and Assays.⁴⁶ **1. In Vivo Chemotherapy.** Immune-deficient mice were housed in microisolator cages within a barrier facility on a 12-h light/dark cycle and received food and water ad libitum. Animal housing was in accord with AAALAC guidelines. All experimental protocols involving animals were approved by the Institutional Animal Care and Use Committee. The C6 rat glioma, the NIH 3T3 fibroblast line transfected with human PDGF ($\beta\beta$), and the DU-145 prostatic adenocarcinoma were maintained by serial passage in nude mice. Nude mice were also used as tumor hosts for test agent evaluations against these tumor models. In each experiment for anticancer activity evaluation, test mice (6–8/treatment group) weighing 18–22 g were randomized and implanted with tumor fragments in the region of the right axilla on day 0 (at least 6 mice/group and 12 in the control). Animals were treated with test compound on the basis of average cage weight on the days indicated in the table. Compound **54** was delivered as a solution in water. Compound dosing solution was prepared for 5 days at a time. Host body weight change data are reported as the maximum treatment-related weight loss in these studies and are included as a gross indicator of therapy-related toxicity. Calculation of tumor growth inhibition (% T/C), tumor growth delay ($T - C$), and net logs of tumor cell kill was performed as described previously.^{55–57} A positive net cell kill indicates that the tumor burden at the end of therapy was less than at the beginning of therapy. A negative net log cell kill indicates that the tumor grew during treatment. Net cell kills near 0 indicate no tumor growth during therapy.

2. Oral Pharmacokinetic Profiles in Rat. Oral pharmacokinetics were evaluated in three male Wistar rats following a single 5 mg/kg oral gavage dose of **53** and **54**. The rats were prepared in 17% PEG 400/83% methylcellulose (0.5%). Heparinized plasma samples were collected via a jugular vein cannula from 0 to 48 h. Calibration curves containing standards were prepared in methanol. Blank rat plasma (200 μL) was spiked with 20 μL of standards and extracted randomized with study samples. Following liquid-liquid extraction with ethyl ether, the organic layer was dried under nitrogen and reconstituted with 100 μL water/acetonitrile (50%/50% v/v). Samples were analyzed on an LC-MS/MS instrument with electrospray positive ion mode (VG Quattro II triple quadrupole). The assay lower limit of quantitation was 10 ng/mL. Pharmacokinetic parameter values were estimated by noncompartmental analysis of individual plasma concentration–time data. Maximum plasma concentrations (C_{max}) and times for these to occur (t_{max}) were recorded as observed. Apparent terminal elimination rate constants (λ_z) were estimated as the absolute value of the slope of the least-squares linear regression of the log-linear terminal phase of concentration–time profiles. Apparent terminal

elimination half-life values ($t_{1/2}$) were calculated from elimination-rate constants as $t_{1/2} = 0.693/\lambda_z$. Area under plasma concentration–time curve $\text{AUC}(0-\infty)$ values were calculated from time 0 to the time of last detectable concentration (t_{last}) using the trapezoidal rule and were extrapolated to infinity.

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4. The viscosities of dilute solutions of amino acids of long dipole moments increase with the length of the hydrocarbon chain, according to the equation $(\eta/\eta_0 - 1) = (0.052 + 0.10 \times n_{CH_2}) C = 2.5 K\phi$, where ϕ is the volume fraction occupied by the solute, C its concentration per liter of solution, and V its molal volume. K is defined by this equation and represents deviation from the behavior of a large ideal spherical solute molecule.

5. The comparable equation for aqueous solutions of aliphatic acids and amides is $(\eta/\eta_0 - 1) = (0.035 + 0.076 \times n_{CH_2}) = 2.5 K\phi$.

6. Measurements upon aliphatic amides, amino acids and peptides even up to viscosities more than three times that of water are given by the equation $(\eta/\eta_0 - 1) = 2.5 K\phi + (2.5 K\phi)^{2.8}$.

7. The sodium salts of the amino acids are all more viscous than the free amino acids, behave much as do the sodium salts of aliphatic acids and have the same viscosity at the same volume fraction. The above equation holds for the salts of amino and aliphatic acids up to solutions as concentrated as 10% with K equal to 2.3.

8. Those proteins which obey Poiseuille's law and are approximately spherical have roughly the same viscosity at the same volume fraction as amino acids and peptides. The higher viscosities of protein salts than of isoelectric proteins may be compared with the higher viscosity of the sodium salts of the amino acids and with the higher viscosity of the sodium salts of citric acid.

BOSTON, MASS.

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[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, YALE UNIVERSITY]

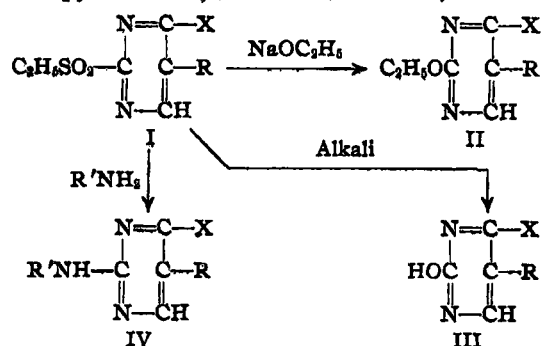
Researches on Pyrimidines. CXLIX. Reactions of Some 2-Ethylsulfonylpyrimidines

BY JAMES M. SPRAGUE¹ AND TREAT B. JOHNSON

A study of the action of chlorine on some ethylmercaptopyrimidines has resulted recently in the preparation of a series of new ethylsulfonylpyrimidine compounds.² These are the first representatives of this type to be described in the pyrimidine literature. In an attempt to establish the structure of these interesting compounds, it was observed that an ethoxyl group was easily substituted for an ethylsulfonyl group by the action of alcoholic alkali upon 2-ethylsulfonyl-4-ethoxy-5-methylpyrimidine.² This has led to a more detailed study of the behavior of the ethylsulfonylpyrimidines toward such reagents as sodium ethoxide, alkali, ammonia and aniline.

It has been found that the ethylsulfonyl group when substituted in a pyrimidine molecule behaves in a manner analogous to the corresponding chloropyrimidines. Thus this group may be replaced easily by an ethoxyl group when the ethylsulfonylpyrimidine (I) is treated with a cold alcoholic solution of sodium ethoxide. Consequently, it was not possible to prepare an ethoxysulfone derivative (I, X = OC₂H₅) from a chlorosulfone (I, X = Cl) by reaction with sodium ethoxide; instead the ethylsulfonyl group and chloro-

rine were simultaneously replaced by ethoxyl. 2,4-Diethoxy-5-methylpyrimidine (II, X = OC₂H₅, R = CH₃) was formed from both 2-ethylsulfonyl-4-ethoxy-5-methyl (I, X = OC₂H₅, R = CH₃) and 2-ethylsulfonyl-4-chloro-5-methylpyrimidine (I, X = Cl, R = CH₃). Likewise, 2,4-diethoxy-5-bromopyrimidine was obtained from 2-ethylsulfonyl-4-ethoxy-5-bromo- (I, X = OC₂H₅, R = Br) and from 2-ethylsulfonyl-4-chloro-5-bromopyrimidine (I, X = Cl, R = Br).

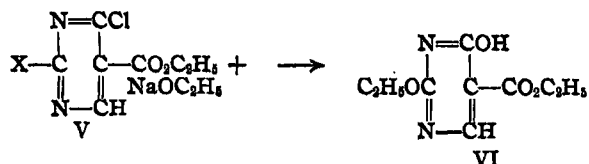


The aminosulfone, 2-ethylsulfonyl-4-amino-5-carbethoxypyrimidine (I, X = NH₂, R = CO₂-C₂H₅) reacts with sodium ethoxide to give 2-ethoxy-4-amino-5-carbethoxypyrimidine (II, X = NH₂, R = CO₂-C₂H₅). Also, when 2-ethylsulfonyl-4-chloro-5-carbethoxypyrimidine (V, X

(1) Sterling Professorship of Chemistry Research Assistant (1935-36).

(2) Sprague and Johnson, *THIS JOURNAL*, **57**, 2252 (1935).

= C₂H₅SO₂) was allowed to react with an excess of sodium ethoxide in alcoholic solution, a 2-ethoxy-4-oxy-5-carbethoxypyrimidine (VI) was obtained. The same compound (VI) was also obtained from 2,4-dichloro-5-carbethoxypyrimidine (V, X = Cl) and an excess of sodium ethoxide. To establish the position of the ethoxy group in compound VI,



attempts were made to produce a pyrimidine of this structure (VI) by the removal of the amino group in 2-ethoxy-4-amino-5-carbethoxypyrimidine (II, X = NH₂, R = CO₂C₂H₅) by action of nitrous acid. However, these attempts were not successful. Likewise the reverse procedure, converting (VI) into 2-ethoxy-4-amino-5-carbethoxypyrimidine by proceeding through the corresponding 4-chloro compound, was not successful on the quantity of pyrimidine available for the work. A product of indefinite composition was obtained. Consequently at present the exact structure of VI is not established with certainty. However, the reaction of V (X = C₂H₅SO₂ or Cl) with excess sodium ethoxide is apparently similar to that reported by Hilbert and Jansen.³ They observed the formation of 2-ethoxy-4-oxy-5-bromopyrimidine from 2,4-dichloro-5-bromopyrimidine and 2-oxy-4-ethoxypyrimidine from 2,4-dichloropyrimidine in presence of an excess of sodium ethoxide in alcohol. Thus the influence of a negative substituent in position 5 of the pyrimidine cycle is exhibited in this type of exchange. Since 2,4-dichlorocarbethoxypyrimidine (V) has the strongly negative carbethoxy group at position 5 of the pyrimidine ring the structure VI is tentatively assigned to the monoethoxy compound formed in the above reaction with sodium alcoholate.

On acid hydrolysis, the ethylsulfonyl group is replaced by hydroxyl and sulfur dioxide is evolved.³ Under these conditions an ethoxyl (or a chlorine) group in position 4 (I, X = OC₂H₅) is simultaneously replaced by an hydroxyl group. However, if alkali instead of acid is used for this reaction an ethoxyl group in position 4 of the pyrimidine cycle may be left intact. Thus, by the action of alkali upon 2-ethylsulfonyl-4-ethoxypyrimidines, the corresponding 2-oxy-4-ethoxypyrimidines (III, X = OC₂H₅, R = H, CH₃ or Br) were obtained. Of these compounds Hilbert and Jansen had previously prepared 2-oxy-4-ethoxypyrimidine (III, X = OC₂H₅, R = H). However, by their procedure, they obtained 2-ethoxy-4-oxy-5-bromopyrimidine rather than the isomeric 2-oxy-4-ethoxy-5-bromopyrimidine (III, X = OC₂H₅, R = Br) which we have now obtained from the corresponding ethylsulfonylpyrimidine.

The 2-ethylsulfonyl group is also replaceable by an amino or phenylamino group through the reaction with alcoholic ammonia or aniline. As in the chloropyrimidines, this reaction with ammonia and aniline emphasizes the effect of structure upon the activity of the ethylsulfonyl group. 2-Ethylsulfonyl-4-amino-5-carbethoxypyrimidine (I, X = NH₂, R = CO₂C₂H₅) with alcoholic ammonia gave 2,4-diamino-5-carbethoxypyrimidine. 2-Anilino-4-ethoxy-5-methylpyrimidine was obtained by the reaction of aniline on 2-ethylsulfonyl-4-ethoxy-5-methylpyrimidine. However, this ethylsulfonylpyrimidine was unreactive toward alcoholic ammonia at 110 or 130° and was recovered unchanged. This suggests that an amino ethylsulfonylpyrimidine would be obtained by the action of alcoholic ammonia on a chloroethylsulfonylpyrimidine (I, X = Cl). This was found to be true. 2-Ethylsulfonyl-4-amino-5-methylpyrimidine was obtained from the corresponding 4-chloropyrimidine (I, X = Cl, R = CH₃) and alcoholic ammonia at 100°.

Attempts to replace the ethylsulfonyl group by hydrogen through action of reducing agents have thus far been unsuccessful.

Experimental Part

2,4-Dimethoxy-5-methylpyrimidine.—A solution of 2.2 g. of 2-ethylsulfonyl-4-chloro-5-methylpyrimidine in 30 cc. of absolute ethanol was added with cooling to a solution of 0.57 g. of sodium in 25 cc. of dry ethanol. The mixture was allowed to stand overnight at room temperature and the alcohol removed in a vacuum below 40° (bath temperature). Water was added and the reaction product extracted with petroleum ether. After drying over calcium chloride the ether extract was evaporated. The residue (1.2 g., m. p. 35.5–36.5°) was identical with an authentic sample⁴ of 2,4-diethoxy-5-methylpyrimidine.

Likewise under similar conditions 2-ethylsulfonyl-4-ethoxy-5-methylpyrimidine (1.0 g.) with an alcoholic solution of sodium (0.15 g.) gave 2,4-diethoxy-5-methylpyrimidine (0.5 g.).

2,4-Diethoxy-5-bromopyrimidine.—2-Ethylsulfonyl-4-chloro-5-bromopyrimidine (1.2 g.) in dry ethanol

(3) Hilbert and Jansen, *THIS JOURNAL*, **56**, 134 (1934); **57**, 552 (1935).

(4) Johnson and Schmidt-Nickels, *ibid.*, **52**, 4514 (1930).

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(20 cc.) was added to a solution of sodium (0.24 g.) in alcohol (20 cc.). On working up as described above 0.85 g. of 2,4-diethoxy-5-bromopyrimidine (m. p. 72.5–73.5°) was obtained. This gave no depression of melting point when mixed with 2,4-diethoxy-5-bromopyrimidine prepared as described by Hilbert and Jansen.⁵

Under the same conditions 0.5 g. of 2-ethylsulfonyl-4-ethoxy-5-bromopyrimidine with 0.07 g. of sodium gave 0.32 g. of the same diethoxypyrimidine.

2-Ethoxy-4-amino-5-carbethoxypyrimidine.—A solution of 3.9 g. of 2-ethylsulfonyl-4-amino-5-carbethoxypyrimidine in 130 cc. of dry ethanol was added to a cold solution of 0.53 g. of sodium in 30 cc. of dry ethanol. After standing overnight at room temperature the alcohol was removed in a vacuum below 40°. The solid residue was extracted thoroughly with hot benzene. On evaporation the benzene extract gave 2.1 g. of crystalline product, m. p. 104–105.5°. This may be recrystallized from an alcohol-water mixture or benzene, m. p. 105–105.5°.

*Anal.*⁶ Calcd. for $C_9H_{10}O_5N_2$: C, 51.16; H, 6.21; N, 19.90. Found: C, 51.31; H, 6.47; N, 19.97.

Attempts to determine the ethoxyl content of this compound by the Vieback and Schwappach method as described by Clark⁶ were unsuccessful. The values obtained ranged from 31 to 40% (calcd. 42.77) for one to five hours' duration. Likewise, 2,4-diamino-5-carbethoxypyrimidine gave low values, 16–20% (calcd. 24.74%) for duration of one to two and one-half hours.

On evaporating on a steam-bath with concentrated hydrochloric acid this pyrimidine was converted into 2-oxo-4-amino-5-carbethoxypyrimidine,⁷ m. p. 276° (dec.).

Anal. Calcd. for $C_7H_8O_4N_2$: N, 22.95. Found: N, 22.88.

The 2-Ethoxy-4-oxo-5-carbethoxypyrimidine.—(1) One gram of 2-ethylsulfonyl-4-chloro-5-carbethoxypyrimidine in 22 cc. of dry ethanol was added to a solution of 0.24 g. of sodium in 15 cc. of dry ethanol with cooling. After standing at room temperature overnight, the precipitated sodium chloride was filtered off and the alcohol removed in a vacuum below 40°. The residue was dissolved in about 10 cc. of water, and, after washing with ether, acidified with acetic acid, whereupon a solid separated (m. p. 184–185°). This was recrystallized from water and separated in the form of needles; yield 0.4 g., m. p. 185–186°.

Anal. Calcd. for $C_9H_{10}O_5N_2$: N, 13.20; OC_2H_5 , 42.45. Found: N, 13.14; OC_2H_5 , 42.00.

(2) To a solution of 1.15 g. of sodium in 30 cc. of absolute ethanol, 3.6 g. of 2,4-dichloro-5-carbethoxypyrimidine⁸ in 30 cc. of dry ethanol was slowly added with cooling in ice. After addition was complete, the reaction was allowed to stand overnight (sixteen hours). The alcohol was removed below 35° in a vacuum, and the residue dissolved in 25 cc. of water, filtered and extracted with ether. The ether extract, after washing with water, was dried with calcium chloride and evaporated. The residue (0.62 g., m. p. 33–35°) was recrystallized from an alcohol-water mixture, m. p. 33.5–34°. The results of a nitrogen analysis

agreed with the calculated for 2,4-diethoxy-5-carbethoxypyrimidine.

Anal. Calcd. for $C_{11}H_{12}O_4N_2$: N, 11.66. Found: N, 11.68.

The aqueous solution on acidification with 2 cc. of acetic acid gave a precipitate which after drying weighed 2.20 g., m. p. 186°. This was recrystallized from water, m. p. 186–186.5°. It was shown by mixed melting point and analysis to be identical with the pyrimidine obtained from the chlorosulfone above.

Anal. Calcd. for $C_9H_{10}O_5N_2$: N, 13.20; OC_2H_5 , 42.45. Found: N, 13.28; OC_2H_5 , 42.34.

2 - Oxy - 4 - ethoxypyrimidine.—2 - Ethylsulfonyl - 4 - ethoxypyrimidine (0.3 cc.) was boiled with 10% sodium hydroxide solution (1–2 cc.) until solution was complete. The solution was cooled in ice and acidified with acetic acid. The precipitate obtained here was recrystallized from water or an alcohol-ethyl acetate mixture and melted at 164–165°. A mixed melting point with a sample prepared by the method of Hilbert and Jansen (m. p. 164–165°) showed no depression. It gave the Wheeler and Johnson cytosine color test.⁹

2-Oxy-4-ethoxy-5-bromopyrimidine.—One-half gram of 2-ethylsulfonyl-4-ethoxy-5-bromopyrimidine was boiled with 10 cc. of 10% sodium hydroxide until solution was complete (about five minutes). The solution was then cooled and acidified with acetic acid. The dried precipitate (0.3 g.) was recrystallized from water, m. p. 234°. This gave the Wheeler and Johnson color test for cytosine.⁹

Anal. Calcd. for $C_8H_7O_2N_2Br$: N, 12.79; OC_2H_5 , 20.57. Found: N, 12.78; OC_2H_5 , 20.24.

2-Oxy-4-ethoxy-5-methylpyrimidine.—One gram of 2-ethylsulfonyl-4-ethoxy-5-methylpyrimidine with 30 cc. of sodium hydroxide on acidification gave 0.5 g. of product. This was recrystallized from water, m. p. 212–213°.

Anal. Calcd. for $C_7H_{10}O_2N_2$: N, 18.17; OC_2H_5 , 29.23. Found: N, 18.18; OC_2H_5 , 29.15.

2,4 - Diamino - 5 - carbethoxypyrimidine.—2 - Ethylsulfonyl-4-amino-5-carbethoxypyrimidine (1.3 g.) was heated at 110° for one and one-half hours with 16 cc. of alcoholic ammonia, saturated at 0°. On cooling, large needle-formed crystals separated, yield 0.90 g. This was recrystallized from alcohol, m. p. 204–205°.

Anal. Calcd. for $C_7H_{10}O_2N_4$: N, 30.76. Found: N, 30.50.

2-Aniline-4-ethoxy-5-methylpyrimidine.—Freshly distilled aniline, 1.8 cc., and 1.15 g. of 2-ethylsulfonyl-4-ethoxy-5-methylpyrimidine were heated in an atmosphere of carbon dioxide for two to three hours at 130°. During the reaction a solid separated and the odor of sulfur dioxide was noticeable. At the end of the reaction, hot ethyl acetate was added and the solid filtered off. The filtrate was treated with norite and evaporated. The residue solidified and was triturated with cold 50% alcohol and filtered. This gave 0.45 g., m. p. 118–119°. However, a further quantity may be obtained from the alcoholic filtrate. This was recrystallized from alcohol-water mixture; m. p. 120.5–121°. It was found later that this reaction could be carried out at the temperature of the steam-bath.

(5) We are indebted to Dr. Frank Stodola of this Laboratory for the carbon and hydrogen analyses of this compound.

(6) Clark, *J. Assoc. Official Agr. Chem.*, 15, 136 (1932).

(7) Wheeler and Johns, *Am. Chem. J.*, 38, 594 (1907).

Anal. Calcd. for $C_{10}H_{10}ON_2$: N, 18.33; OC_2H_5 , 19.66. Found: N, 18.14; OC_2H_5 , 19.50.

On evaporating on a steam-bath with concentrated hydrochloric acid and making alkaline with ammonia, this compound gave 2-anilino-4-oxy-5-methoxypyrimidine, m. p. 254–255° from alcohol. This same compound was obtained for comparison by heating 2-ethylmercapto-4-oxy-5-methylpyrimidine with aniline on a steam-bath until all the ethylmercaptan was evolved. Benzene was added and the solid removed by filtering. This was recrystallized from alcohol, m. p. 254–255°.

Anal. Calcd. for $C_{11}H_{11}ON_2$: N, 20.88. Found: N, 20.70.

2 - Ethylsulfonyl - 4 - amino - 5 - methylpyrimidine.—Two grams of 2-ethylsulfonyl-4-chloro-5-methylpyrimidine was heated with 25 cc. of ethanol, saturated with dry ammonia at 0°, for two hours at 100°. The alcohol was then evaporated and the residue extracted with hot ethyl acetate. The ethyl acetate solution was concentrated to a small volume and cooled. There was obtained 0.9 g. of crystals, m. p. 128–130°. A further quantity was obtained from the filtrate (0.13 g.). This was recrystallized from ethyl acetate, m. p. 136.5–136.5°.

Anal. Calcd. for $C_7H_{11}O_2N_2S$: N, 20.88; S, 15.94. Found: N, 20.90; S, 16.18.

On hydrolysis² with hydrochloric acid this sulfone evolved sulfur dioxide. The aqueous solution on evaporation gave 5-methylcytosine hydrochloride.³ This was recrystallized from an alcohol-concentrated hydrochloric acid solution by addition of ether, m. p. 288–290° (dec.).

Anal. Calcd. for $C_5H_7O_2N_3 \cdot HCl$: N, 26.00; Cl, 21.95. Found: N, 25.80; Cl, 22.21.

Summary

In this paper is described the behavior of several 2-ethylsulfonylpyrimidines when allowed to interact with sodium alcoholate, alkali, ammonia and aniline. The ethylsulfonyl group of the pyrimidine reacts in all these changes in a similar manner as a halogen atom being easily replaced by alkoxy, hydroxy and amino groups.

(8) Wheeler and Johnson, *Am. Chem. J.*, 31, 598 (1904).

NEW HAVEN, CONNECTICUT

RECEIVED DECEMBER 26, 1935

Calculation of the Solubility of a Mixture of Hydrogen and Nitrogen in Water at 25° in the Pressure Range of 50 to 1000 Atmospheres

BY JACOB KIELLAND¹

Introduction

R. Wiebe and V. L. Gaddy² have recently published experimental data concerning the solubility of a 76.42:23.58 hydrogen-nitrogen mixture in water at 25°, and at high pressure. It was found that the solubility of the mixture could be calculated to within a few per cent. from the values of the pure constituents, by multiplying the mole fraction of each separate gas by its own solubility at a pressure equal to the total pressure of the mixture, and then adding these values together.

However, on examining the solubility of each one of the constituents it will be found that there is a certain amount of difference between the values observed and those calculated. As far as the nitrogen is concerned, there is a deviation of about 13% at 800 to 1000 atm., Wiebe and Gaddy's own analyses of the composition of the gas in the water phase being employed (see Table I).

Discussion

The difference between the calculated and observed solubilities—assuming that there is no

systematical error in measurement—may be explained by the existence of deviations from the fugacity rule³ of Lewis and Randall, which is the basis of method of calculation, and states

$$\bar{f}_i = \frac{\partial f}{\partial n_i} = N_i f_i$$

or, in other words, that the activity coefficient of a gas is the same at constant pressure, irrespective of whether the gas occurs alone or in mixtures. According to A. R. Merz and C. W. Whittaker,⁴ however, this rule does not apply to hydrogen-nitrogen mixtures at the pressures in question in the present case. From the work carried out by them, the values for γ_i/γ_i^0 or $\bar{f}_i/N_i f_i^0$, given in Table II, columns 2 and 3, have been taken.

In the water phase, where the action of the water molecules on the activity coefficients must be assumed to be of greater importance than the interaction of the separate gases, the latter process will not be taken into consideration. The results obtained prove this assumption to be perfectly justified.

It must be pointed out that the solubility correction for deviations from the Lewis and Randall

(1) Chemical Engineer, Norsk Hydro-Elektrisk Kvaeststofaktieselskab, Oslo, Norway.

(2) Wiebe and Gaddy, *THIS JOURNAL*, 57, 1487 (1935).

(3) Cf. Newton and Dodge, *Ind. Eng. Chem.*, 27, 578 (1935).

(4) Merz and Whittaker, *THIS JOURNAL*, 50, 1522 (1928).

prisms, m. p. 265–267° (yield, 5.1 g.) (Found: C, 47.0; H, 6.0; N, 13.5; Cl', 13.3. $C_{21}H_{20}N_4Br \cdot 2HCl \cdot 2H_2O$ requires C, 46.9; H, 6.0; N, 13.0; Cl', 13.2%). The *dipicrate* crystallised from β -ethoxyethanol as yellow laminae, m. p. 224–225° (Found: C, 44.7; H, 4.3; N, 16.6. $C_{21}H_{20}N_4Br \cdot 2C_6H_3O_7N_3$ requires C, 44.7; H, 3.6; N, 17.3%).

(b) 4-Chloro-2- β -diethylaminoethylamino-6-methylpyrimidine (6.1 g.), 6-bromo- β -naphthylamine (5.5 g.), water (25 c.c.), and 10N-hydrochloric acid (2.5 c.c.) were refluxed for 1 hour. From the clear solution 4-(6'-bromo- β -naphthylamino)-2- β -diethylaminoethylamino-6-methylpyrimidine dihydrochloride crystallised on cooling. It was filtered off and dissolved in hot water, and the solution decolorised with charcoal and filtered. Addition of hydrochloric acid to the filtrate reprecipitated the product, which was filtered off, dried, and crystallised from alcohol. It formed colourless needles, m. p. 266–268° undepressed by admixture with material made by method (a) (yield, 9.9 g.).

4-(6'-Bromo- β -naphthylamino)-2- γ -diethylaminopropylamino-6-methylpyrimidine (XIII; R = $[CH_2]_3 \cdot NEt_2$).—4-Chloro-2- γ -diethylaminopropylamino-6-methylpyrimidine (6.4 g.), 6-bromo- β -naphthylamine (5.5 g.), water (25 c.c.), and 10N-hydrochloric acid (2.2 c.c.) were refluxed for one hour. The product, which separated on cooling, was filtered off, dissolved in water and reprecipitated with hydrochloric acid. After being dried, the *dihydrochloride* (4787) was crystallised from alcohol; colourless needles, m. p. 259–261° (yield, 11.2 g.) (Found in material dried at 100°: C, 47.4; H, 6.5; N, 13.0; Cl', 13.3. $C_{22}H_{22}N_4Br \cdot 2HCl \cdot 2H_2O$ requires C, 47.9; H, 6.2; N, 12.7; Cl', 12.9%).

4-(6'-Bromo- β -naphthylamino)-2- γ -dibutylaminopropylamino-6-methylpyrimidine (XIII; R = $[CH_2]_3 \cdot NBu^a$).—4-Chloro-2- γ -dibutylaminopropylamino-6-methylpyrimidine (7.8 g.) and 6-bromo- β -naphthylamine (5.5 g.) were added to water (25 c.c.) containing 10N-hydrochloric acid (2.5 c.c.) and the mixture refluxed for one hour. On adding more hydrochloric acid and cooling 4-(6'-bromo- β -naphthylamino)-2- γ -dibutylaminopropylamino-6-methylpyrimidine *dihydrochloride* (4788) separated and was filtered off. It was purified by dissolving in water, reprecipitating with hydrochloric acid and crystallising the dried product from alcohol-ethyl acetate; it was thus obtained as colourless needles, m. p. 250–252° (yield, 8.8 g.) (Found: C, 51.6; H, 6.7; N, 11.4; Cl', 11.7. $C_{24}H_{28}N_4Br \cdot 2HCl \cdot 2H_2O$ requires C, 51.4; H, 6.9; N, 11.5; Cl', 11.7%).

4-(6'-Bromo- β -naphthylamino)-2- δ -diethylamino- α -methylbutylamino-6-methylpyrimidine (XIII; R = $CHMe \cdot [CH_2]_3 \cdot NEt_2$).—4-Chloro-2- δ -diethylamino- α -methylbutylamino-6-methylpyrimidine (9.5 g.), 6-bromo- β -naphthylamine (7.4 g.), water (50 c.c.), and 10N-hydrochloric acid (3.3 c.c.) were refluxed for 2 hours. The resulting clear solution was cooled, made alkaline with sodium hydroxide, and extracted with chloroform. The chloroform extract was shaken with 5% acetic acid, and the acetic acid extract separated and added to excess sodium hydroxide. The precipitated oil was extracted with chloroform, and the solution dried (K_2CO_3) and evaporated. The oil was characterised as its *dipicrate*, which separated from β -ethoxyethanol as yellow prisms, m. p. 240–242° (Found: C, 46.6; H, 4.3; N, 16.4. $C_{24}H_{28}N_4Br \cdot 2C_6H_3O_7N_3$ requires C, 46.6; H, 4.1; N, 16.6%). The *dihydrochloride* (5063), prepared by evaporating to dryness under reduced pressure a solution of the base in 2N-hydrochloric acid, crystallised from alcohol-ethyl acetate as colourless laminae, m. p. 260–262° (Found: C, 53.4; H, 6.3; N, 12.5; Cl', 12.5. $C_{24}H_{28}N_4Br \cdot 2HCl$ requires C, 53.0; H, 6.3; N, 12.9; Cl', 13.1%).

4-(6'-Bromo- β -naphthylamino)-2- γ -piperidinopropylamino-6-methylpyrimidine (XIII; R = $[CH_2]_3 \cdot N < CH_2 > \cdot CH_2$).—A mixture of 4-chloro-2- γ -piperidinopropylamino-6-methylpyrimidine (5.4 g.), 6-bromo- β -naphthylamine (4.45 g.), water (40 c.c.), and 10N-hydrochloric acid (2 c.c.) was refluxed for 1 hour. From the clear solution the product began to separate after about 10 minutes. After adding more hydrochloric acid to make the mixture acid to Congo red and cooling, the product was filtered off, dried, and crystallised from alcohol giving the *dihydrochloride* (5041) as colourless elongated prisms, m. p. 288–289° (yield, 10.3 g.) (Found: C, 48.8; H, 6.0; N, 12.2; Cl', 13.2. $C_{23}H_{28}N_4Br \cdot 2HCl \cdot 2H_2O$ requires C, 49.0; H, 6.0; N, 12.4; Cl', 12.6%).

4-(6'-Bromo- β -naphthylamino)-2- γ -n-butylaminopropylamino-6-methylpyrimidine (XIII; R = $[CH_2]_3 \cdot NHBu^a$).—Crude 4-chloro-2- γ -n-butylaminopropylamino-6-methylpyrimidine (6.4 g.), 6-bromo- β -naphthylamine (5.5 g.), water (25 c.c.), and 10N-hydrochloric acid (2.5 c.c.) were refluxed for 2 hours, and the solution cooled and made alkaline with sodium hydroxide. The liberated base was extracted with chloroform and this in turn extracted with 5% acetic acid. The acetic acid extract was made alkaline with sodium hydroxide and the base isolated with chloroform. It formed an oil which did not crystallise but was characterised as its *dipicrate*, which separated from β -ethoxyethanol in yellow prisms, m. p. 219–220° (Found: C, 45.3; H, 4.1; N, 16.9. $C_{23}H_{28}N_4Br \cdot 2C_6H_3O_7N_3$ requires C, 45.3; H, 3.6; N, 17.1%). The *dihydrochloride* (5152), prepared by dissolving the base in warm 2N-hydrochloric acid and cooling, was filtered off and dried. It crystallised from alcohol as colourless prisms, m. p. 301–303° (Found: C, 50.8; H, 5.6; N, 13.9; Cl', 12.8. $C_{23}H_{28}N_4Br \cdot 2HCl$ requires C, 51.3; H, 5.8; N, 13.6; Cl', 13.8%).

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74. Synthetic Antimalarials. Part VII. 2-Arylamino-4-dialkylaminoalkylamino-pyrimidines. Variation of Substituents in the 5- and the 6-Position.

By F. H. S. CURD, (Miss) D. N. RICHARDSON, and F. L. ROSE.

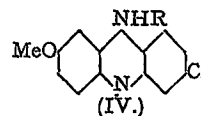
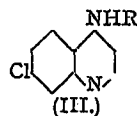
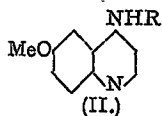
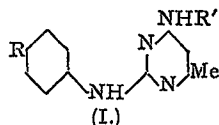
The studies of 2-arylamino-4-aminoalkylamino-6-methylpyrimidines as antimalarials (Curd and Rose, this vol., p. 343; Curd, Davis, and Rose, *ibid.*, p. 351) have been extended to compounds having (a) no substituent in the 6-position, (b) a 6-phenyl group, and (c) various substituents in the 5-position (with and without a 6-methyl group).

The formal similarity of compounds of type (I) to riboflavin (see previous papers) has been further explored and an attempt has been made to increase their activity by the synthesis of structures capable not only of riboflavin antagonism but also possibly of interference with nucleoside synthesis on the basis of the hypothesis (Part III, Hull, Lovell, Openshaw, Payman, and Todd, this vol., p. 357) that pyrimidine derivatives bearing a substituent in position 5 and an aminoalkylamino group in position 4 might be capable of functioning in this way.

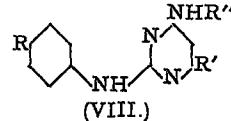
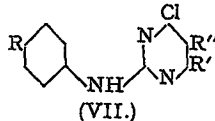
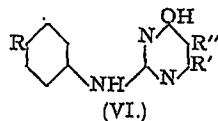
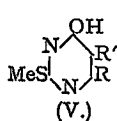
In our initial study of 2-arylamino-4-dialkylaminoalkylamino-6-methylpyrimidines (I; R = Cl, OMe, R' = NH-alkylene-N(alkyl)₂) as antimalarials (Part I, this vol., p. 343) and the later more detailed examination of the effect of different substituents in the arylamino group and of variations in the dialkylaminoalkylamino residue (Part II, this vol., p. 351; Part V, *ibid.*, p. 366) the 6-methyl group was invariably present. The

original reason for this was that such compounds were more accessible than those unsubstituted in the 6-position, and not because the 6-methyl group was thought to have any chemotherapeutic significance.

It has been suggested (Part V, *loc. cit.*) that quinoline derivatives of types (II) and (III; R = dialkylaminoalkyl) may, because of their relationship to mepacrine (IV; R = CHMe·[CH₂]₃·NEt₃), act in a similar manner to this drug and therefore to the compounds of type (I), by interference with a riboflavin-containing enzyme important for the survival of the malaria parasite. The introduction of a methyl group into the 2-position of compounds of type (II) appears to have a dystherapeutic effect (Magidson and Rubtsov, *J. Gen. Chem. Russ.*, 1937, 7, 1896; Krichevski *et al.*, *J. Microbiol. Epidemiol. and Immunobiol. U.S.S.R.*, 1935, 14, 642). According to D.R.P. 683,692 the same is true of compounds of type (III), and by analogy it seemed possible that removal

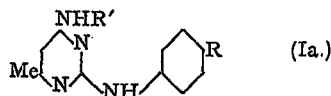
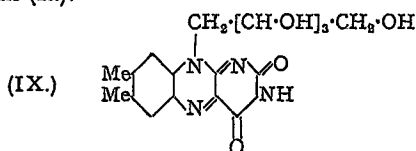


of the 6-methyl group in type (I) might lead to increased antimalarial activity. In order to investigate this point we prepared 2-*p*-chloroanilino-4-β-diethylaminoethylaminopyrimidine (VIII; R = Cl, R' = H, R'' = [CH₂]₃·NEt₃) and the corresponding diethylaminopropylamino derivative (VIII; R = Cl, R' = H, R'' = [CH₂]₃·NEt₃) by condensing *p*-chloroaniline with 4-hydroxy-2-methylthiopyrimidine (V; R = R' = H) (Wheeler and Merriam, *Amer. Chem. J.*, 1903, 29, 478) to give 2-*p*-chloroanilino-4-hydroxypyrimidine (VI; R = Cl, R' = R'' = H) followed by conversion, with phosphoryl chloride, into the chloropyrimidine (VII; R = Cl, R' = R'' = H) and condensation of this with β-diethylaminoethylamine and γ-diethylaminopropylamine respectively. The antimalarial activity of these compounds was much lower than that of the corresponding 6-methyl derivatives. Similar low antimalarial activity was found with 2-*p*-toluidino-4-β-diethylaminoethylaminopyrimidine (VIII; R = Me, R' = H, R'' = [CH₂]₃·NEt₃).



The importance of the 6-methyl group in (I; R = Cl, R' = dialkylaminoalkyl) having been demonstrated we were led to examine the effect of replacing the 6-methyl group by a variety of other groups. This will be dealt with more fully in a later communication, but we record here the synthesis of 2-*p*-chloroanilino-4-β-diethylaminoethylamino-6-phenylpyrimidine (VIII; R = Cl, R' = Ph, R'' = [CH₂]₃·NEt₃) from 4-hydroxy-2-methylthio-6-phenylpyrimidine (V; R = Ph, R' = H) via (VI; R = Cl, R' = Ph, R'' = H) and the chloropyrimidine (VII; R = Cl, R' = Ph, R'' = H). The product was practically devoid of antimalarial activity.

Previously, in considering the formal resemblance of compounds of type (I) to riboflavin (IX), it has been supposed that the pyrimidine ring of (I) corresponds to the same ring of the vitamin, but it seemed possible that the observed antagonism between (I; R = Cl, R' = [CH₂]₃·NEt₃) and (IX) (Madinaveitia, *Biochem. J.*, in the press) might be accounted for by a correspondence of the pyrimidine ring of the former to the benzene ring of riboflavin thus correlating the 6-methyl group of (I) to the 6-methyl group of the vitamin. This is illustrated in (Ia).



On this basis the introduction of an additional methyl group into the 5-position of the pyrimidine ring of (I) to correspond to the 7-methyl group of riboflavin appeared to be of interest, but there were other reasons for a detailed study of the effect of introducing additional substituents into this position in (I).

An investigation of alkylpyrimidines carrying aminoalkylamino groups uncomplicated by anilino groups, carried out at Manchester University under Professor Todd and reported in Part III (this vol., p. 357), led to the discovery that 2-amino-4-aminoalkylamino-6-methylpyrimidines of type (X), corresponding to the anilino derivatives of type (I), were without antimalarial activity at tolerated doses, but that the introduction of alkyl substituents into the 5-position of the pyrimidine ring to give (XI; R = dialkylaminoalkyl, R' = alkyl) restored activity.

This dissimilarity between compounds of type (X) and type (I) and the fact that no antagonism could be demonstrated between (XI; R = CHMe·[CH₂]₃·NEt₃, R' = Me) and riboflavin using *Lactobacillus casei* suggested a mode of action for compounds of type (XI) different from that which is responsible for the activity of compounds of type (I), and the speculative hypothesis was advanced that compounds of the former type might be capable of interference with either the synthesis or the function of purine nucleosides, particularly those, such as adenosine, which are widely occurring coenzyme constituents.

If this hypothesis is correct then it seemed possible that the introduction of a substituent into the 5-position of the pyrimidine ring of compounds of type (I) might cause a potentiation of activity. Such compounds

TABLE I.

Antimalarial Activities.

The antimalarial activities were estimated using chicks infected with *P. gallinaceum*. For the significance of the symbols used to express activity reference should be made to Part I.

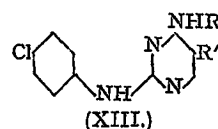
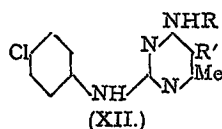
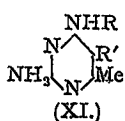
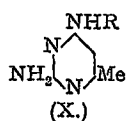
Ref. No.	Substance.	Dose, mg./kg.	Activity.
3780	2- <i>p</i> -Chloroanilino-4- γ -diethylaminopropylaminopyrimidine	160 80	+ —
3839	2- <i>p</i> -Chloroanilino-4- β -diethylaminoethylaminopyrimidine	160 80	+ to ++ —
3834	2- <i>p</i> -Toluidino-4- β -diethylaminoethylaminopyrimidine	120 80	$\pm$ $\pm$
4145	2- <i>p</i> -Chloroanilino-4- β -diethylaminoethylamino-6-phenylpyrimidine	160 80	$\pm$ —
3687	2- <i>p</i> -Chloroanilino-4- γ -diethylaminopropylamino-5 : 6-dimethylpyrimidine	160 80 40	++ ++ + to ++
3903	2- <i>p</i> -Chloroanilino-4- β -diethylaminoethylamino-5 : 6-dimethylpyrimidine	40 20	+ $\pm$
4410	2- <i>p</i> -Chloroanilino-4- γ -dimethylaminopropylamino-5 : 6-dimethylpyrimidine	40 20	++ +
4065	2- <i>p</i> -Chloroanilino-4- β -diethylaminoethylamino-6-methyl-5-ethylpyrimidine	120 80 40	+ to ++ + $\pm$
4119	2- <i>p</i> -Chloroanilino-4- β -dimethylaminoethylamino-6-methyl-5-ethylpyrimidine	80 40	+ —
4120	2- <i>p</i> -Chloroanilino-4- γ -diethylaminopropylamino-6-methyl-5-ethylpyrimidine	80 40	+ —
4118	2- <i>p</i> -Chloroanilino-4- γ -dimethylaminopropylamino-6-methyl-5-ethylpyrimidine	80 40	++ +
4396	2- <i>p</i> -Chloroanilino-4- δ -diethylamino- α -methylbutylamino-6-methyl-5-ethylpyrimidine	160 120 80	+ — —
4209	2- <i>p</i> -Chloroanilino-4- β -diethylaminoethylamino-5-benzyl-6-methylpyrimidine	160 40	+ to ++ +
4231	2- <i>p</i> -Chloroanilino-4- γ -diethylaminopropylamino-5-benzyl-6-methylpyrimidine	160 80	+ to ++ —
4238	2- <i>p</i> -Chloroanilino-4- γ -dimethylaminopropylamino-5-benzyl-6-methylpyrimidine	160 80	+ $\pm$
4253	2- <i>p</i> -Chloroanilino-4- δ -diethylamino- α -methylbutylamino-5-benzyl-6-methylpyrimidine	160 80	— —
3563	5-Bromo-2- <i>p</i> -chloroanilino-4- γ -diethylaminopropylamino-6-methylpyrimidine	400 80	+ —
4343	2- <i>p</i> -Chloroanilino-4- β -diethylaminoethylamino-5 : 6-cyclohexenopyrimidine	200 120 80 40	++ + + —
4356	2- <i>p</i> -Chloroanilino-4- γ -diethylaminopropylamino-5 : 6-cyclohexenopyrimidine	80 40	+ to ++ +
4355	2- <i>p</i> -Chloroanilino-4- γ -dimethylaminopropylamino-5 : 6-cyclohexenopyrimidine	80 40 20 10	++ ++ + to ++ +
4557	2- <i>p</i> -Chloroanilino-4- γ -dimethylaminopropylamino-5 : 6-cyclopentenopyrimidine	80 40	+ to ++ —
3815	2- <i>p</i> -Chloroanilino-4- β -diethylaminoethylamino-5-methylpyrimidine	80 40	— —
4146	2- <i>p</i> -Chloroanilino-4- β -diethylaminoethylamino-5-phenylpyrimidine	160 80	$\pm$ —
4208	2- <i>p</i> -Chloroanilino-4- γ -diethylaminopropylamino-5-phenylpyrimidine	80 40	— —
4064	2- <i>p</i> -Anisidino-4- β -diethylaminoethylamino-5-phenylpyrimidine	160 80	— —
4260	2- <i>p</i> -Chloroanilino-4- β -diethylaminoethylamino-5-phenoxy pyrimidine	160 80 40	$\pm$ — —
2666	2- <i>p</i> -Chloroanilino-4- β -diethylaminoethylamino-6-methylpyrimidine	80 40	++ +
3711	2- <i>p</i> -Chloroanilino-4- γ -dimethylaminopropylamino-6-methylpyrimidine	40 20	++ +

should still retain the property of acting as riboflavin antagonists and if they were capable also of purine nucleoside antagonism they might be expected to possess a greater activity against an enzyme system of which riboflavin-adenosine-dinucleotide is a constituent than a compound capable only of interference with either the

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riboflavin or the adenosine portion. It also appeared possible that a compound of type (XII) might have a wider range of antagonistic activity and be capable of interference not only with riboflavin-containing enzymes but also with other enzyme systems having a purine nucleoside as a coenzyme constituent.

A series of compounds of type (XII) has therefore been prepared, and the following have been introduced as substituents into the 5-position: methyl, ethyl, benzyl, bromine. Disubstitution in the 5- and the 6-position has also taken the form of an additional 5- or 6-membered *cyclopenteno*- or *cyclohexeno*-ring. In nearly every case more than one dialkylaminoalkyl group has been tried. The compounds prepared are listed in Table I, which gives an indication of the antimalarial activities. For comparison the activities of two compounds of type (I) are included. Full biological details will be published and discussed in detail elsewhere, but no compound of type (XII) has shown a significantly greater activity than the corresponding compound of type (I) without the 5-substituent.



It then occurred to us that an investigation of compounds of type (XIII; R' = alkyl) might give some information on the mode of action of type (XII). It has been mentioned above that compounds of type (VIII; R' = H) are much less active than those of type (VIII; R' = Me). If then the introduction of a substituent into position 5 of (VIII; R = H) to give (XIII) conferred on these compounds the power to act in the same manner as those of type (XI) it might be anticipated that in type (XIII; R' = alkyl) the activity might be largely restored as compared with (VIII; R = H). 2-*p*-Chloroanilino-4- β -diethylaminoethylamino-5-methylpyrimidine (XIII; R = [CH₂]₂·NEt₂, R' = Me) was therefore prepared but had no activity. Low activity was also characteristic of several other compounds of this type, but with different substituents in the 5-position, which were prepared: 2-*p*-chloroanilino-4- β -diethylaminoethylamino-5-phenylpyrimidine (XIII; R = [CH₂]₂·NEt₂, R' = Ph), the corresponding *p*-anisidino-derivative, 2-*p*-chloroanilino-4- γ -diethylamino-propylamino-5-phenylpyrimidine (XIII; R = [CH₂]₃·NEt₂, R' = Ph), and 2-*p*-chloroanilino-4- β -diethylamino-ethylamino-5-phenoxy-pyrimidine (XIII; R = [CH₂]₂·NEt₂, R' = OPh).

Compounds of types (XII) and (XIII) were made by the now well established method from a 4-hydroxy-2-alkylthiopyrimidine (V) appropriately substituted in the 5- and 6-positions through the intermediate stages (VI) and (VII). The majority of the 4-hydroxy-2-alkylthiopyrimidines of type (V) used were known compounds but a few are described now for the first time. 5-Bromo-4-hydroxy-2-methylthio-6-methylpyrimidine was prepared by bromination of 4-hydroxy-2-methylthio-6-methylpyrimidine in acetic acid. 4-Hydroxy-2-methylthio-5:6-cyclopentenopyrimidine and 4-hydroxy-2-methylthio-5:6-cyclohexenopyrimidine were prepared by condensation of ethyl cyclopentanone-2-carboxylate and ethyl cyclohexanone-2-carboxylate respectively with *S*-methylisothiourea in an aqueous medium. The yields were low owing to side reactions the products of which were not investigated, but in the latter case the yield was considerably improved by condensing the ethyl cyclohexanone-2-carboxylate with thiourea and methylating the resulting 4-hydroxy-2-thiol 5:6-cyclohexenopyrimidine with methyl sulphate and alkali.

EXPERIMENTAL.

2-*p*-Chloroanilino-4-hydroxypyrimidine (VI; R = Cl, R' = R'' = H).—4-Hydroxy-2-methylthiopyrimidine (Wheeler and Merriam, *loc. cit.*) (18 g.), *p*-chloroaniline (32 g.), and β -ethoxyethanol (50 c.c.) were refluxed with stirring for 26 hours. The solid material which separated was filtered off after cooling, washed with hot alcohol (100 c.c.), and dried (yield, 21.5 g.). Crystallisation from acetic acid gave 2-*p*-chloroanilino-4-hydroxypyrimidine as thin colourless prisms, m. p. 242–244° (Found: N, 19.0. C₁₀H₈ON₂Cl requires N, 19.0%).

4-Chloro-2-*p*-chloroanilinopyrimidine (VII; R = Cl, R' = R'' = H).—2-*p*-Chloroanilino-4-hydroxypyrimidine (21.5 g.) and phosphoryl chloride (55 c.c.) were refluxed for 15 minutes. The excess of phosphoryl chloride was removed under reduced pressure and the solid residue added to ice. The resulting suspension was made alkaline with ammonia and, after stirring for 1 hour, the crude product was collected and washed with water. It crystallised from alcohol in clusters of colourless prisms, m. p. 124° (yield, 16.0 g.) (Found: C, 50.2; H, 2.6; N, 17.0; Cl, 29.4. C₁₀H₇N₃Cl₂ requires C, 50.0; H, 2.9; N, 17.5; Cl, 29.6%).

2-*p*-Chloroanilino-4-hydroxy-6-phenylpyrimidine (VI; R = Cl, R' = Ph, R'' = H).—4-Hydroxy-2-methylthio-6-phenylpyrimidine (Wheeler and Merriam, *loc. cit.*) (8.2 g.) and *p*-chloroaniline (12 g.) were stirred and heated at 130–140° for 6 hours. The cooled melt was ground and then refluxed with alcohol (75 c.c.) for 1½ hours. The product was filtered off, washed with alcohol, and dried (yield, 9.7 g.). It crystallised from β -ethoxyethanol as colourless needles, m. p. 312–313° (Found: C, 64.1; H, 4.0; N, 14.0. C₁₈H₁₃ON₂Cl requires C, 64.8; H, 4.0; N, 14.1%).

4-Chloro-2-*p*-chloroanilino-6-phenylpyrimidine (VII; R = Cl, R' = Ph, R'' = H).—The above hydroxy compound (9.3 g.) and phosphoryl chloride (30 c.c.) were refluxed for 1½ hours. After removing the excess of phosphoryl chloride under reduced pressure the residue was added to crushed ice and made alkaline with ammonia. After standing for 1 hour the crude product was filtered off, washed with water, and crystallised from alcohol, giving 4-chloro-2-*p*-chloroanilino-6-phenylpyrimidine as colourless prisms, m. p. 166–168° (yield, 6.1 g.) (Found: N, 13.5; Cl, 22.3. C₁₈H₁₁N₃Cl₂ requires N, 13.3; Cl, 22.5%).

2-*p*-Chloroanilino-4-hydroxy-5:6-dimethylpyrimidine (VI; R = Cl, R' = R'' = Me).—4-Hydroxy-2-ethylthio-5:6-dimethylpyrimidine (Wheeler and Merriam, *loc. cit.*) (15 g.) and *p*-chloroaniline (26 g.) were heated at 130–140° for 9 hours with stirring. The cooled and ground reaction mixture was refluxed with alcohol (150 c.c.) for 1 hour and filtered, and the undissolved residue washed with alcohol and dried (yield, 20.85 g.). When purified by crystallisation from aqueous acetic acid the compound had m. p. 270–272° (Found: C, 57.8; H, 5.2; N, 16.5. C₁₂H₁₂ON₂Cl requires C, 57.7; H, 4.8; N, 16.9%).

4-Chloro-2-p-chloroanilino-5 : 6-dimethylpyrimidine (VII; R = Cl, R' = R'' = Me).—2-p-Chloroanilino-4-hydroxy-5 : 6-dimethylpyrimidine (20 g.) and phosphoryl chloride (60 c.c.) were refluxed for 1½ hours and the reaction mixture worked up as in the case of previous chloropyrimidines. Purified by crystallisation from alcohol the product formed colourless prisms, m. p. 161—162° (yield, 11.4 g.) (Found : C, 53.2; H, 4.1; N, 15.5. C₁₃H₁₁N₃Cl₂ requires C, 53.7; H, 4.1; N, 15.7%).

2-p-Chloroanilino-4-hydroxy-6-methyl-5-ethylpyrimidine (VI; R = Cl, R' = Me, R'' = Et).—Prepared from 4-hydroxy-2-methylthio-6-methyl-5-ethylpyrimidine (*idem, ibid.*) (18.4 g.) and p-chloroaniline (32 g.) exactly as described above for the corresponding 5-methyl compound, the compound formed clusters of colourless needles from β-ethoxyethanol, m. p. 246—247° (Found : C, 59.1; H, 5.3; N, 15.9. C₁₃H₁₃ON₂Cl requires C, 59.2; H, 5.3; N, 15.9%) (yield, 24.95 g.).

4-Chloro-2-p-chloroanilino-6-methyl-5-ethylpyrimidine (VII; R = Cl, R' = Me, R'' = Et).—2-p-Chloroanilino-4-hydroxy-6-methyl-5-ethylpyrimidine (13.05 g.) and phosphoryl chloride (40 c.c.) were refluxed for 1½ hours and the reaction mixture worked up in the usual way. The chloropyrimidine crystallised from alcohol as stout colourless needles, m. p. 128—130° (yield, 11.4 g.) (Found : N, 14.9; Cl, 25.3. C₁₃H₁₃N₃Cl₂ requires N, 14.9; Cl, 25.2%).

2-p-Chloroanilino-4-hydroxy-5-benzyl-6-methylpyrimidine (VI; R = Cl, R' = Me, R'' = CH₂Ph).—4-Hydroxy-2-ethylthio-5-benzyl-6-methylpyrimidine (Wheeler and McFarland, *Amer. Chem. J.*, 1909, 42, 101) (13.08 g.) and p-chloroaniline (17.5 g.) heated at 130—140° for 6 hours and worked up as described above for this type of compound gave the hydroxy-pyrimidine (yield, 15.55 g.) which crystallised from β-ethoxyethanol as colourless needles, m. p. 258—260° (Found : C, 65.8; H, 4.9; N, 12.9. C₁₈H₁₆ON₂Cl requires C, 66.4; H, 4.9; N, 12.9%).

4-Chloro-2-p-chloroanilino-5-benzyl-6-methylpyrimidine (VII; R = Cl, R' = Me, R'' = CH₂Ph), prepared from the above hydroxy compound (29.55 g.) and phosphoryl chloride (90 c.c.), crystallised from alcohol as colourless tables, m. p. 124—125° (yield, 25.5 g.) (Found : N, 12.3; Cl, 20.5. C₁₈H₁₆N₃Cl₂ requires N, 12.2; Cl, 20.6%).

5-Bromo-4-hydroxy-2-methylthio-6-methylpyrimidine (V; R = Me, R' = Br).—4-Hydroxy-2-methylthio-6-methylpyrimidine (45 g.) was dissolved in acetic acid (1,500 c.c.) and bromine (54 g.) added gradually with stirring at room temperature. The precipitate was collected and washed with water. It was then stirred with dilute ammonia, filtered off, and purified by crystallisation from β-ethoxyethanol from which the compound separated as colourless thick prisms, m. p. 254—256° (decomp.) (yield, 41 g.) (Found : N, 11.8; Br, 34.5. C₈H₈ON₂BrS requires N, 11.9; Br, 34.0%).

5-Bromo-2-p-chloroanilino-4-hydroxy-6-methylpyrimidine (VI; R = Cl; R' = Me; R'' = Br).—5-Bromo-4-hydroxy-2-methylthio-6-methylpyrimidine (11.75 g.), p-chloroaniline (12.75 g.), and β-ethoxyethanol (25 c.c.) were stirred and heated at 120—130° for 26 hours. The product was filtered off, washed well with alcohol, and dried (yield, 11.75 g.). It crystallised from dimethylformamide-water in colourless blunt-ended needles, m. p. 267—269° (decomp.) (Found : C, 42.5; H, 3.2; N, 13.6. C₁₁H₉ON₂ClBr requires C, 42.0; H, 2.9; N, 13.4%).

4-Chloro-5-bromo-2-p-chloroanilino-6-methylpyrimidine (VII; R = Cl, R' = Me, R'' = Br).—The above hydroxy-pyrimidine (20 g.) and phosphoryl chloride (45 c.c.) were refluxed for 3 hours. Excess of phosphoryl chloride was then removed under reduced pressure and the residue added to ice with stirring. After making alkaline with ammonia, the chloro compound was extracted with ether, and the extract dried and evaporated. Crystallisation of the residue from alcohol gave the chloropyrimidine as practically colourless prisms, m. p. 176—178° (Found : N, 12.5; 1 mg. = 1.376 mg. Ag halides. C₁₁H₉N₃Cl₂Br requires N, 12.6%; 1 mg. = 1.428 mg. Ag halides).

4-Hydroxy-2-methylthio-5 : 6-cyclohexenopyrimidine. (a) Ethyl cyclohexanone-2-carboxylate (29 g.) was added to a solution of S-methylisothiourea sulphate (24 g.) in water (150 c.c.) containing potassium hydroxide (12 g.). The ester dissolved immediately and a solid was precipitated. After standing overnight this was filtered off and washed with water. It was then dissolved in sodium hydroxide solution, and the solution treated with charcoal, filtered, and acidified with acetic acid. The dried precipitate was crystallised first from β-ethoxyethanol and then from alcohol, giving 4-hydroxy-2-methylthio-5 : 6-cyclohexenopyrimidine as colourless needles, m. p. 220—222° (yield, 4.7 g.) (Found : C, 55.3; H, 5.8; N, 14.0. C₈H₁₀ON₂S requires C, 55.1; H, 6.1; N, 14.3%).

(b) Thiourea (16 g.) and ethyl cyclohexanone-2-carboxylate (34 g.) were added to a solution of sodium (12 g.) in methanol (300 c.c.) and refluxed for 5½ hours. The mixture was then evaporated to dryness under reduced pressure, and the residue dissolved in water, treated with decolourising charcoal, and filtered. The filtrate was acidified with acetic acid and the precipitated product collected, washed with water, and dried. Crystallisation from β-ethoxyethanol gave 4-hydroxy-2-thiol-5 : 6-cyclohexenopyrimidine as colourless flat prisms, m. p. 314—320° (decomp.) (yield, 20.8 g.) (Found : C, 52.8; H, 5.2; N, 15.4. C₈H₁₀ON₂S requires C, 52.75; H, 5.5; N, 15.4%).

4-Hydroxy-2-thiol-5 : 6-cyclohexenopyrimidine (18.2 g.) was dissolved in 10% potassium hydroxide solution (50 c.c.) and methyl sulphate (15 g.) added to the solution in small portions, with shaking. The precipitate was filtered off, washed with water, and dried. Crystallisation from alcohol gave 4-hydroxy-2-methylthio-5 : 6-cyclohexenopyrimidine, m. p. 220—222° undepressed in admixture with material made by method (a) (yield, 11.15 g.).

2-p-Chloroanilino-4-hydroxy-5 : 6-cyclohexenopyrimidine. 4-Hydroxy-2-methylthio-5 : 6-cyclohexenopyrimidine (14.8 g.) and p-chloroaniline (24 g.) were heated at 130—140° for 6 hours with stirring in the initial stages. Methylthiol was evolved and a homogeneous melt gradually solidified. After cooling this was ground and refluxed with alcohol (200 c.c.) for 1½ hours. The mixture was then cooled, filtered, and the residue washed with alcohol and dried (yield, 19.4 g.). The compound separated from β-ethoxyethanol as clusters of colourless prisms, m. p. 284—287° with previous darkening (Found : C, 60.6; H, 5.0; N, 15.1. C₈H₁₀ON₂Cl requires C, 61.0; H, 5.1; N, 15.2%).

4-Chloro-2-p-chloroanilino-5 : 6-cyclohexenopyrimidine. The above hydroxy compound (18.4 g.) and phosphoryl chloride (56 c.c.) were refluxed for 1½ hours and the reaction mixture worked up as in the case of previous chloropyrimidines. The product separated from alcohol as colourless plates, m. p. 137—138° (yield, 13.84 g.) (Found : N, 14.5; Cl, 24.1. C₁₄H₁₂N₃Cl₂ requires N, 14.3; Cl, 24.2%).

4-Hydroxy-2-methylthio-5 : 6-cyclopentenopyrimidine. S-Methylisothiourea sulphate (33 g.) and ethyl cyclopentanone-2-carboxylate (40 g.) were added to a solution of potassium hydroxide (15 g.) in water (100 c.c.). The ester quickly dissolved and the mixture was left for 2 days. The solid material was then collected and dissolved in sodium hydroxide solution, and the solution treated with charcoal. After filtration the solution was acidified with acetic acid and the precipitate filtered off, washed with water, and dried. Crystallisation from β-ethoxyethanol gave 4-hydroxy-2-methylthio-5 : 6-cyclopentenopyrimidine (3.5 g.) as colourless prisms, m. p. 270—272° (Found : C, 52.6; H, 5.2; N, 15.3. C₈H₁₀ON₂S requires C, 52.8; H, 5.5; N, 15.4%).

2-p-Chloroanilino-4-hydroxy-5 : 6-cyclopentenopyrimidine. 4-Hydroxy-2-methylthio-5 : 6-cyclopentenopyrimidine (6 g.) and p-chloroaniline (11 g.) were intimately mixed and heated at 130—140° for 24 hours. The product was powdered, boiled with alcohol (100 c.c.) for 3 hours, and filtered. The insoluble material was crystallised from β-ethoxyethanol-water giving the hydroxy-pyrimidine as colourless needles, m. p. 244—246° with previous darkening (yield, 6.35 g.) (Found : C, 60.1; H, 4.7; N, 15.7. C₁₃H₁₂ON₂Cl requires C, 59.7; H, 4.6; N, 16.1%).

4-Chloro-2-p-chloroanilino-5 : 6-cyclopentenopyrimidine, prepared from the above hydroxy compound (5.8 g.) and phosphoryl chloride (18 c.c.), crystallised from alcohol as colourless laminæ, m. p. 151—152° (yield, 3.95 g.) (Found : N, 14.7; Cl, 25.2. C₁₃H₁₁N₃Cl₂ requires N, 15.0; Cl, 25.4%).

2-p-Chloroanilino-4-hydroxy-5-methylpyrimidine (VI; R = Cl, R' = H, R'' = Me).—4-Hydroxy-2-methylthio-5-

TABLE II.

2-p-Chloroamolino-4-dialkylaminoalkylaminopyrimidines. Variation of the Substituents in the 5- and the 6-Position.

Substituent at—					Analysis.								
5.	6.	4.	Derivative.	M. p.	Formula.	Found, %				Required, %			
						C.	H.	N.	Cl.	C.	H.	N.	Cl.
H	H	NH·[CH ₂] ₂ ·NEt ₂	—	71—72°	C ₁₄ H ₂₂ N ₆ Cl	60.0	6.8	22.0	—	60.1	6.9	21.9	—
H	H	NH·[CH ₂] ₂ ·NEt ₂	Dihydrochloride	237—238	C ₁₄ H ₂₂ N ₆ Cl, 2HCl, 1.5H ₂ O	45.6	7.4	16.1	17.2	45.8	6.4	16.7	16.9
			Oil	—	—	—	—	—	—	—	—	—	—
			Dipicrate	218—220	C ₁₇ H ₂₄ N ₆ Cl, 2C ₆ H ₃ O ₇ N ₂	44.1	3.8	18.8	—	44.0	3.8	19.5	—
H	Ph	NH·[CH ₂] ₂ ·NEt ₂	Dihydrochloride	208—210	C ₁₄ H ₂₂ N ₆ Cl, 2HCl, 2H ₂ O	46.1	6.7	15.8	16.7	46.1	6.8	15.8	16.05
			Oil	—	—	—	—	—	—	—	—	—	—
			Dihydrochloride	277—279	C ₂₂ H ₂₆ N ₆ Cl, 2HCl	56.0	5.6	15.2	15.2	56.3	6.0	14.9	15.15
Me	Me	NH·[CH ₂] ₂ ·NEt ₂	—	100—102	C ₁₄ H ₂₂ N ₆ Cl	62.0	7.6	20.4	—	62.2	7.5	20.1	—
			Dihydrochloride	270—272	C ₁₄ H ₂₂ N ₆ Cl, 2HCl, H ₂ O	49.1	6.8	16.0	16.0	49.3	6.8	16.0	16.2
Me	Me	NH·[CH ₂] ₂ ·NEt ₂	—	104—106	C ₁₄ H ₂₂ N ₆ Cl	63.7	8.1	19.3	—	63.1	7.7	19.4	—
			Dihydrochloride	277—279	C ₁₄ H ₂₂ N ₆ Cl, 2HCl, H ₂ O	49.8	7.0	15.1	—	50.4	7.1	15.5	—
			(decomp.)	—	—	—	—	—	—	—	—	—	—
Me	Me	NH·[CH ₂] ₂ ·NMe ₂	—	116—118	C ₁₄ H ₂₂ N ₆ Cl	60.9	7.0	20.6	—	61.2	7.2	21.0	—
			Dihydrochloride	238—240	C ₁₄ H ₂₂ N ₆ Cl, 2HCl, H ₂ O	47.8	6.4	16.2	16.9	48.1	6.6	16.5	16.7
Et	Me	NH·[CH ₂] ₂ ·NEt ₂	—	92—94	C ₁₄ H ₂₂ N ₆ Cl	63.8	7.7	19.2	—	63.1	7.7	19.4	—
			Dihydrochloride	258—260	C ₁₄ H ₂₂ N ₆ Cl, 2HCl, 0.5H ₂ O	51.2	6.9	15.8	15.8	51.4	7.0	15.8	16.05
Et	Me	NH·[CH ₂] ₂ ·NMe ₂	—	115—116	C ₁₄ H ₂₂ N ₆ Cl	60.6	7.1	20.4	—	61.2	7.2	21.0	—
			Dihydrochloride	262	C ₁₄ H ₂₂ N ₆ Cl, 2HCl, H ₂ O	47.9	6.6	16.6	16.5	48.1	6.6	16.5	16.75
Et	Me	NH·[CH ₂] ₂ ·NEt ₂	—	108—109	C ₁₄ H ₂₂ N ₆ Cl	63.7	7.7	18.3	—	63.9	8.0	18.6	—
			Dihydrochloride	272—274	C ₁₄ H ₂₂ N ₆ Cl, 2HCl, H ₂ O	51.3	7.3	15.0	14.7	51.4	7.3	15.0	15.2
Et	Me	NH·[CH ₂] ₂ ·NMe ₂	—	126—128	C ₁₄ H ₂₂ N ₆ Cl	62.6	7.5	19.7	—	62.2	7.5	20.1	—
			Dihydrochloride	244—246	C ₁₄ H ₂₂ N ₆ Cl, 2HCl, 1.5H ₂ O	48.1	6.7	15.0	16.1	48.3	6.9	15.6	15.9
Et	Me	NH·CHMe·[CH ₂] ₂ ·NEt ₂	—	Oil	—	—	—	—	—	—	—	—	—
			Dihydrochloride	231—233	C ₂₂ H ₂₆ N ₆ Cl, 2HCl, 1.5H ₂ O	52.8	7.7	14.0	14.3	52.4	7.7	13.9	14.1
CH ₂ ·Ph	Me	NH·[CH ₂] ₂ ·NEt ₂	—	114—116	C ₂₄ H ₂₈ N ₆ Cl	67.8	6.9	16.4	—	68.0	7.1	16.5	—
			Dihydrochloride	255—256	C ₂₄ H ₂₈ N ₆ Cl, 2HCl, 0.5H ₂ O	56.8	6.3	14.0	14.0	57.0	6.5	13.8	14.0
CH ₂ ·Ph	Me	NH·[CH ₂] ₂ ·NEt ₂	—	104—105	C ₂₂ H ₂₆ N ₆ Cl	68.7	7.2	16.6	—	68.6	7.3	16.0	—
			Dihydrochloride	274—276	C ₂₂ H ₂₆ N ₆ Cl, 2HCl	58.6	6.5	13.4	13.7	58.8	6.7	13.7	13.9
			(decomp.)	—	—	—	—	—	—	—	—	—	—
CH ₂ ·Ph	Me	NH·[CH ₂] ₂ ·NMe ₂	—	112—114	C ₂₂ H ₂₆ N ₆ Cl	67.4	6.7	17.0	—	67.4	6.8	17.1	—
			Dihydrochloride	254—256	C ₂₂ H ₂₆ N ₆ Cl, 2HCl, H ₂ O	55.2	5.9	14.1	14.3	55.1	6.4	14.0	14.2
CH ₂ ·Ph	Me	NH·CHMe·[CH ₂] ₂ ·NEt ₂	—	Oil	—	—	—	—	—	—	—	—	—
			Dihydrochloride	242—244	C ₂₄ H ₂₈ N ₆ Cl, 2HCl, 0.5H ₂ O	59.5	6.7	13.2	13.2	59.2	7.1	12.8	13.0
Br	Me	NH·[CH ₂] ₂ ·NEt ₂	—	94—96	C ₁₄ H ₂₂ N ₆ ClBr	50.1	5.9	16.0	—	50.6	5.9	16.4	—
			Dihydrochloride	238—240	C ₁₄ H ₂₂ N ₆ ClBr, 2HCl, 0.5H ₂ O	42.8	6.0	13.7	13.4	42.5	5.5	13.8	14.0
5 : 6-cycloHexeno		NH·[CH ₂] ₂ ·NEt ₂	—	132—134	C ₂₀ H ₂₈ N ₆ Cl	64.3	7.0	18.3	—	64.3	7.5	18.7	—
			Dihydrochloride	224—226	C ₂₀ H ₂₈ N ₆ Cl, 2HCl, 2H ₂ O	49.4	6.8	14.8	14.5	49.7	7.0	14.5	14.7
5 : 6-cycloHexeno		NH·[CH ₂] ₂ ·NEt ₂	—	138—140	C ₂₁ H ₃₀ N ₆ Cl	65.1	7.7	18.4	—	65.0	7.7	18.1	—
			Dihydrochloride	202—204	C ₂₁ H ₃₀ N ₆ Cl, 2HCl, 1.5H ₂ O	51.3	6.8	14.7	14.7	51.7	7.2	14.4	14.6
: 6-cycloHexeno		NH·[CH ₂] ₂ ·NMe ₂	—	125—127	C ₁₆ H ₂₂ N ₆ Cl	63.6	7.1	19.5	—	63.4	7.2	19.5	—
			Dihydrochloride	290	C ₁₆ H ₂₂ N ₆ Cl, 2HCl	53.3	6.3	16.1	16.4	52.7	6.5	16.2	16.4
			(decomp.)	—	—	—	—	—	—	—	—	—	—
5 : 6-cycloPenteno		NH·[CH ₂] ₂ ·NMe ₂	—	142—144	C ₁₅ H ₂₂ N ₆ Cl	62.8	7.4	20.8	—	62.5	6.9	20.3	—
Me	H	NH·[CH ₂] ₂ ·NEt ₂	—	106—108	C ₁₇ H ₂₄ N ₆ Cl	61.4	7.4	21.0	—	61.2	7.2	21.0	—
			Dihydrochloride	269	C ₁₇ H ₂₄ N ₆ Cl, 2HCl	50.0	6.5	17.2	17.0	50.2	6.4	17.2	17.5
			(decomp.)	—	—	—	—	—	—	—	—	—	—
Ph	H	NH·[CH ₂] ₂ ·NEt ₂	—	152—153	C ₂₂ H ₂₆ N ₆ Cl	67.1	6.4	17.3	—	66.8	6.6	17.7	—
			Dihydrochloride	264—266	C ₂₂ H ₂₆ N ₆ Cl, 2HCl	56.6	6.0	14.8	15.2	56.35	6.0	14.9	15.2
Ph	H	NH·[CH ₂] ₂ ·NEt ₂	—	155—156	C ₂₂ H ₂₆ N ₆ Cl	67.0	6.5	16.9	—	67.4	6.8	17.1	—
			Dihydrochloride	264—266	C ₂₂ H ₂₆ N ₆ Cl, 2HCl	56.7	6.3	14.4	14.8	57.2	6.2	14.5	14.7
OPh	H	NH·[CH ₂] ₂ ·NEt ₂	—	84—85	C ₂₂ H ₂₆ ON ₆ Cl	64.2	6.2	17.0	—	64.2	6.3	17.0	—

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methylpyrimidine (Wheeler and Merriam, *loc. cit.*) (20.8 g.) and *p*-chloroaniline (34 g.) were boiled in β -ethoxyethanol (50 c.c.) for 48 hours and the mixture worked up as described above for 2-*p*-chloroanilino-4-hydroxypyrimidine. The compound crystallised from acetic acid as colourless thin plates, m. p. 266–267° (yield, 26 g.) (Found: C, 55.6; H, 4.5; N, 17.7. $C_{11}H_{10}ON_2Cl$ requires C, 56.06; H, 4.2; N, 17.8%).

4-Chloro-2-*p*-chloroanilino-5-methylpyrimidine (VII; R = Cl, R' = H, R'' = Me), prepared by refluxing 2-*p*-chloroanilino-4-hydroxy-5-methylpyrimidine (21 g.) and phosphoryl chloride (63 c.c.) for $\frac{1}{2}$ hour and working up in the usual way, crystallised from β -ethoxyethanol as colourless prisms, m. p. 153–159° (yield, 13.9 g.) (Found: N, 16.0; Cl, 28.4. $C_{11}H_9N_2Cl_2$ requires N, 16.5; Cl, 28.0%).

2-*p*-Chloroanilino-4-hydroxy-5-phenylpyrimidine (VI; R = Cl, R' = H, R'' = Ph).—4-Hydroxy-2-ethylthio-5-phenylpyrimidine (Wheeler and Bristol, *Amer. Chem. J.*, 1906, 33, 460) (19.44 g.) and *p*-chloroaniline (29.9 g.) were heated at 130–140° for 6 hours, with stirring until the melt solidified. When cold, it was ground, refluxed with alcohol (150 c.c.) for $\frac{1}{2}$ hours, and filtered off. The compound was practically insoluble in all the usual organic solvents and was purified by dissolution in sodium hydroxide solution and reprecipitation with acetic acid followed by boiling with β -ethoxyethanol. It then had m. p. 328–330° (Found: C, 64.0; H, 4.1; N, 14.6. $C_{16}H_{13}ON_2Cl$ requires C, 64.5; H, 4.0; N, 14.1%).

4-Chloro-2-*p*-chloroanilino-5-phenylpyrimidine (VII; R = Cl, R' = H, R'' = Ph), prepared by the action of phosphoryl chloride (60 c.c.) on the preceding hydroxy compound (20 g.) for $\frac{1}{2}$ hour followed by working up in the usual way, crystallised from alcohol as colourless elongated prisms, m. p. 133–134° (yield, 15.35 g.) (Found: N, 13.1; Cl, 22.1. $C_{16}H_{11}N_3Cl_2$ requires N, 13.3; Cl, 22.5%).

2-*p*-Anisidino-4-hydroxy-5-phenylpyrimidine (VI; R = OMe, R' = H, R'' = Ph).—4-Hydroxy-2-ethylthio-5-phenylpyrimidine (17.4 g.) and *p*-anisidine (18.45 g.) were boiled in β -ethoxyethanol (60 c.c.) for 21 hours. After cooling, the crystalline product was filtered off, washed well with hot alcohol, and dried (yield, 19.1 g.). It separated from acetic acid as colourless laminæ, m. p. 271–272° (Found: C, 69.3; H, 5.0; N, 14.5. $C_{17}H_{15}O_2N_2$ requires C, 69.6; H, 5.1; N, 14.3%).

4-Chloro-2-*p*-anisidino-5-phenylpyrimidine (VII; R = OMe, R' = H, R'' = Ph), from 2-*p*-anisidino-4-hydroxy-5-phenylpyrimidine (17.15 g.) with phosphoryl chloride (45 c.c.), crystallised from alcohol as colourless needles, m. p. 152° (yield, 11.65 g.) (Found: N, 13.4; Cl, 11.4. $C_{17}H_{14}ON_2Cl$ requires N, 13.5; Cl, 11.4%).

2-*p*-Chloroanilino-4-hydroxy-5-phenoxyypyrimidine (VI; R = Cl, R' = H, R'' = OPh).—4-Hydroxy-2-ethylthio-5-phenoxyypyrimidine (Johnson and Heyl, *Amer. Chem. J.*, 1907, 37, 628) (11.6 g.) and *p*-chloroaniline (16 g.) were heated at 130–140° for 28 hours. The residue (solid when cold) was worked up as before giving the hydroxypyrimidine which, after crystallisation from β -ethoxyethanol, had m. p. 240–242° (yield, 14.3 g.) (Found: C, 60.9; H, 3.8; N, 13.1. $C_{16}H_{12}O_2N_2Cl$ requires C, 61.2; H, 3.8; N, 13.4%).

4-Chloro-2-*p*-chloroanilino-5-phenoxyypyrimidine (VII; R = Cl, R' = H, R'' = OPh).—2-*p*-Chloroanilino-4-hydroxy-5-phenoxyypyrimidine (9.45 g.) and phosphoryl chloride (30 c.c.) were refluxed for 30 minutes. The resulting clear solution was poured on ice and the mixture made alkaline with ammonia. After stirring for 2½ hours the supernatant liquid was decanted. The oily residue was dissolved in alcohol, the solution made alkaline with ammonia and poured into water, and the precipitate collected. The compound crystallised from aqueous alcohol as colourless prisms, m. p. 112–113° (yield, 5.3 g.) (Found: C, 58.1; H, 3.7; N, 12.6. $C_{16}H_{11}ON_2Cl_2$ requires C, 57.8; H, 3.3; N, 12.65%).

4-Dialkylaminoalkylaminopyrimidines.—The chloropyrimidine and the dialkylaminoalkylamine (1.25 mols.) were heated at 125–135° for 8 hours with stirring. The resulting melt was dissolved in hot dilute hydrochloric acid and the solution made alkaline with sodium hydroxide. The base was then extracted with ether or chloroform and the extract shaken several times with 5% acetic acid. Alternatively, the solvent was removed by distillation and the residue extracted with 5% acetic acid. The base was liberated from the combined acetic acid extracts by the addition of sodium hydroxide and again taken into ether or chloroform. Evaporation of the dried solution left the base as a solid, or as an oil which usually crystallised when triturated with light petroleum (b. p. 40–60°). The substances which could not be obtained crystalline in this way, notably those containing the 3-diethylamino- α -methylbutyl side chain, were converted into their dihydrochlorides. The solid bases were purified by crystallisation from light petroleum. For biological testing the dihydrochlorides were usually prepared by dissolving the bases in hot 2*N*-hydrochloric acid and allowing the solution to cool, whereupon the salt normally crystallised out and was collected and dried. If the hydrochloride failed to separate on cooling the solution was evaporated to dryness under reduced pressure at 50–60° and the evaporation repeated with alcohol or alcohol-benzene to remove water and adhering hydrochloric acid; it was then purified by crystallisation from alcohol, alcohol-ethyl acetate, or alcohol-acetone.

In addition to the substances contained in Table II the following were prepared by the same general method:

2-*p*-Toluidino-4- β -diethylaminoethylaminopyrimidine, prepared from 4-chloro-2-*p*-toluidinopyrimidine (Johnson and Storey, *Amer. Chem. J.*, 1908, 40, 143) and β -diethylaminoethylamine, gave a dihydrochloride which crystallised from alcohol-acetone as colourless prisms, m. p. 220–221° (Found: C, 52.1; H, 7.3; N, 17.5; Cl, 18.3. $C_{17}H_{25}N_5 \cdot 2HCl \cdot H_2O$ requires C, 52.3; H, 7.4; N, 17.9; Cl, 18.2%).

2-*p*-Anisidino-4- β -diethylaminoethylamino-5-phenylpyrimidine, crystallised from light petroleum (b. p. 100–120°) in clusters of colourless prisms, m. p. 158–159° (Found: C, 70.2; H, 7.4; N, 18.0. $C_{23}H_{25}ON_4$ requires C, 70.6; H, 7.4; N, 17.9%). The dihydrochloride separated from alcohol-ethyl acetate as crystals, m. p. 209–211° (Found: C, 56.4; H, 6.2; N, 14.5; Cl, 15.3. $C_{23}H_{25}ON_4 \cdot 2HCl \cdot 1.5H_2O$ requires C, 56.2; H, 6.9; N, 14.3; Cl, 14.5%).

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[Received, October 17th, 1945.]

75. The Anhydrides of Polyhydric Alcohols. Part II. Derivatives of 1:2:5:6-Dianhydro Mannitol.

By L. F. WIGGINS.

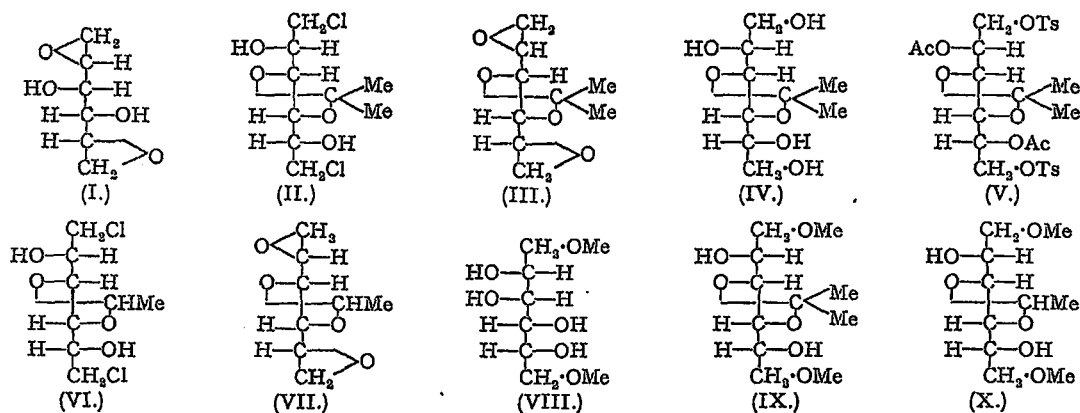
By the action of cold sodium methoxide on 1:6-dichloro 3:4-monoacetone mannitol and 1:6-dichloro 3:4-ethylidene mannitol the monoacetone and the monoethylidene derivative of 1:2:5:6-dianhydro mannitol have been obtained. These compounds were extremely reactive and suffered ring fission under mild alkaline or acid conditions.

The constitutions of these compounds have been proved and some products of ring fission have been described.

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It is well known that ethylene oxidic ring compounds, as exemplified by ethylene oxide itself, glycidol, or ethylene oxide anhydro bodies of the sugar series, are extremely reactive. Thus in the last class the ethylene oxidic ring can react additively with alkaline reagents such as sodium methoxide, ammonia, or hydrazine (see for example Peat and Wiggins, *J.*, 1938, 1810; Robertson and Whitehead, *J.*, 1940, 319) and with acidic reagents such as halogen acids (cf. Müller, *Ber.*, 1934, 67, 421; Peat and Wiggins, *loc. cit.*; Robertson and Dunlop, *J.*, 1938, 472).

It was envisaged that if difunctional substances of this type could be prepared then they would probably undergo ring scission with other difunctional compounds such as diamines or dibasic acids to give polymeric materials of some value. Such a compound would be 1:2-5:6-dianhydro mannitol (I) which possesses at each extremity of the molecule an ethylene oxide anhydro ring which would be available for reaction with suitable difunctional compounds. Although it has not yet been found possible to prepare 1:2-5:6-dianhydro mannitol itself yet two derivatives of this substance, namely 3:4-monoacetone and 3:4-monoethylidene 1:2-5:6-dianhydro mannitol, have been obtained and it is with the preparation and proof of the constitution of these compounds that this paper is chiefly concerned.



1:6-Dichloro mannitol (Siwoliboff, *Annalen*, 1886, 233, 368; Haworth, Heath, and Wiggins, *J.*, 1944, 155) gives on acetonisation three products consisting of (i) 1:6-dichloro monoacetone mannitol obtained in 70% yield, (ii) 1:6-dichloro diacetone mannitol (m. p. 100–102°), and (iii) liquid isomeric 1:6-dichloro diacetone mannitol. The compounds (ii) and (iii) are new and probably differ in respect of the orientation of their acetone groups, which are probably attached in one case to adjacent carbon atoms and in the other to alternate carbon atoms. The monoacetone derivative (i) was first prepared by Micheel (*Annalen*, 1932, 496, 77) who assigned to it, without proof, the structure 1:6-dichloro 3:4-monoacetone mannitol (II). In the course of the following argument this contention is proved. On treatment of (II) with sodium methoxide a liquid was obtained, the analytical figures for which showed it to possess two anhydro rings and to be a dianhydro monoacetone mannitol. The anhydro rings in this compound were extremely labile and it underwent ring scission on boiling for a few hours with aqueous sodium hydroxide. In this way a product was obtained which after benzylation in pyridine solution gave the 1:2:5:6-tetrabenzoyl 3:4-monoacetone mannitol of Brigl and Grüner (*Ber.*, 1933, 66, 933). Therefore the acetone group in Micheel's 1:6-dichloro monoacetone mannitol and in the dianhydro compound derived from it must certainly be attached to C₃ and C₄, and the former compound is correctly represented by (II). It is clear therefore that in the dianhydro compound C₁, C₂, C₅, and C₆ must be involved in anhydro ring formation. This being so, since the dianhydro compound is produced by hydrolysis of the halogen atoms attached to C₁ and C₆, each anhydro ring must also involve either C₁ or C₆ and therefore two possible structures for the anhydride are apparent: (a) 1:2-5:6-dianhydro 3:4-monoacetone mannitol, and (b) 1:5-2:6-dianhydro 3:4-monoacetone mannitol. The first structure involves rings of the ethylene oxide type which are characterised by their greater activity, and the second involves rings of the pyran type which are considerably more stable. The actual properties of the dianhydro body strongly suggest (a) as the correct structure. Moreover atomic models show that (a) can exist but that it is impossible to build up structure (b). Therefore the correct structure of the dianhydro compound must be that of 1:2-5:6-dianhydro 3:4-monoacetone mannitol represented by (III).

A second route to the synthesis of 1:2-5:6-dianhydro 3:4-monoacetone mannitol has been worked out and involves the following steps. Mannitol is readily converted into triacetone mannitol by the original method of Fischer (*Ber.*, 1895, 28, 1167) and, on partial hydrolysis with 70% acetic acid, 3:4-monoacetone mannitol (IV) is obtained in 80% yield (see Wiggins, this vol., p. 13). Tosylation followed by acetylation gave 1:6-ditosyl 2:5-diacyetyl 3:4-monoacetone mannitol (V) which on treatment with four equivalents of sodium methoxide in the cold gave 1:2-5:6-dianhydro 3:4-monoacetone mannitol identical in respect of refractive index, b. p., and optical inactivity with the substance obtained from 1:6-dichloro 3:4-monoacetone mannitol.

A second derivative of 1:2-5:6-dianhydro mannitol, namely the 3:4-monoethylidene derivative, has been prepared as follows. 1:6-Dichloro mannitol on treatment with paraldehyde and a few drops of concentrated

Note 1 Handbook

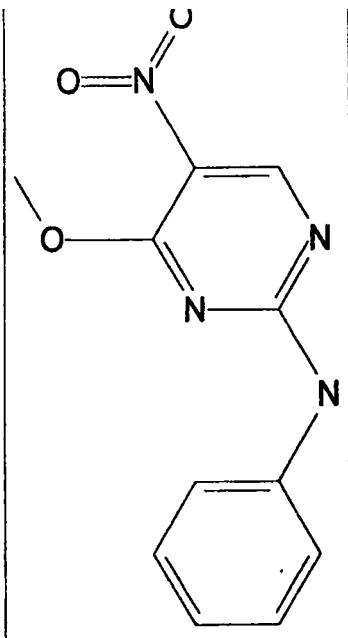
Ref. 1 675158, LitLink ; Journal; Naito; Inoue; CPBTAL; Chem.Pharm.Bull.; 6; 1958; 338, 341.

Melting Point

VALUE (MP) C	Solvent (.SOL)	Note	Ref.
150	ethanol	1	1

Note 1 Handbook

Ref. 1 675158, LitLink ; Journal; Naito; Inoue; CPBTAL; Chem.Pharm.Bull.; 6; 1958; 338, 341.



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Substance

Beilstein Registry Number	249340
Beilstein Preferred RN	99845-42-8
CAS Registry Number	99845-42-8
Chemical Name	(4-methoxy-5-nitro-pyrimidin-2-yl)-phenyl-amine
Autoname	(4-methoxy-5-nitro-pyrimidin-2-yl)-phenyl-amine
Molecular Formula	C ₁₁ H ₁₀ N ₄ O ₃
Molecular Weight	246.22
Lawson Number	29675, 14131, 289
Compound Type	heterocyclic
Constitution ID	189396
Tautomer ID	267873
Beilstein Reference	4-25-00-03358
Entry Date	1988/06/27
Update Date	1992/05/13

Field Availability List 1-2 of 2

Code	Field Name	Occ.
<u>CPD</u>	Crystal Property Description	1
<u>MP</u>	Melting Point	1

Crystal Property Description

Colour & Other Properties	hellgelb
Note 1	Handbook

Ref. 1 675158, LitLink ; Journal; Naito; Inoue; CPBTAL; Chem.Pharm.Bull.; 6; 1958; 338, 341.

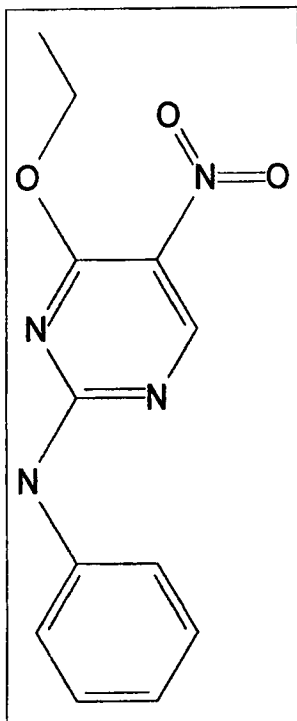
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Melting Point

VALUE (MP) C	Solvent (.SOL)	Note	Ref.
183	methanol	1	1

Note 1 Handbook

Ref. 1 675158, LitLink ; Journal; Naito; Inoue; CPBTAL; Chem.Pharm.Bull.; 6; 1958; 338, 341.



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Substance

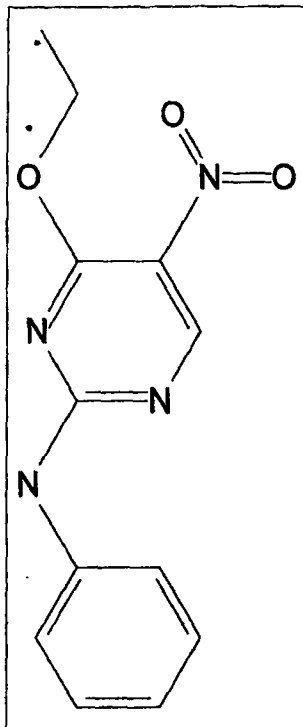
Beilstein Registry Number	265505
Beilstein Preferred RN	100517-98-4
CAS Registry Number	100517-98-4
Chemical Name	(4-ethoxy-5-nitro-pyrimidin-2-yl)-phenyl-amine
Autoname	(4-ethoxy-5-nitro-pyrimidin-2-yl)-phenyl-amine
Molecular Formula	C ₁₂ H ₁₂ N ₄ O ₃
Molecular Weight	260.25
Lawson Number	29675, 14131, 298
Compound Type	heterocyclic
Constitution ID	200880
Tautomer ID	273513
Beilstein Reference	4-25-00-03358
Entry Date	1988/06/27
Update Date	1992/05/13

Field Availability List 1-2 of 2

Code	Field Name	Occ.
<u>CPD</u>	Crystal Property Description	1
<u>MP</u>	Melting Point	1

Crystal Property Description

Colour & Other Properties gelb



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Substance

Beilstein Registry Number	265505
Beilstein Preferred RN	100517-98-4
CAS Registry Number	100517-98-4
Chemical Name	(4-ethoxy-5-nitro-pyrimidin-2-yl)-phenyl-amine
Autoname	(4-ethoxy-5-nitro-pyrimidin-2-yl)-phenyl-amine
Molecular Formula	C ₁₂ H ₁₂ N ₄ O ₃
Molecular Weight	260.25
Lawson Number	29675, 14131, 298
Compound Type	heterocyclic
Constitution ID	200880
Tautomer ID	273513
Beilstein Reference	4-25-00-03358
Entry Date	1988/06/27
Update Date	1992/05/13

Field Availability List 1-2 of 2

Code	Field Name	Occ.
<u>CPD</u>	Crystal Property Description	1
<u>MP</u>	Melting Point	1

Crystal Property Description

Colour & Other Properties	gelb
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Note 1 Handbook

Ref. 1 675158, LitLink ; Journal; Naito; Inoue; CPBTAL; Chem.Pharm.Bull.; 6; 1958; 338, 341.

Melting Point

VALUE (MP) C	Solvent (.SOL)	Note	Ref.
150	ethanol	1	1

Note 1 Handbook

Ref. 1 675158, LitLink ; Journal; Naito; Inoue; CPBTAL; Chem.Pharm.Bull.; 6; 1958; 338, 341.

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**SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO
DIRECCIÓN DE NUEVAS CREACIONES**

Radicación No. 03-112613

Clasificación Internacional (IPC): A61K 31/505, A61K 31/506, C07D 239/48

Concepto Técnico No: 0286

Patente de Invención ☒ Patente de Modelo de Utilidad ☐

Vía Nacional ☐

Vía PCT (fase nacional) ☒ Cap. I ☐ Cap. II ☒

No. Sol. Internacional PCT/EP02/05669

Fecha de Presentación Internal. 23 de Mayo de 2002

Como resultado del examen de fondo, realizado con fundamento en el artículo 48 de la Decisión 486 de la Comisión de la Comunidad Andina, se le comunica:

1. Documentos estudiados

Descripción: Folios: 139 a 321 como se radicó inicialmente

Reivindicaciones: Folios 604 a 626 como se radicó inicialmente
Folios 664 a 681 modificadas
Folios 709 a 745 modificadas

Dibujos: Folio 627 como se radicó inicialmente

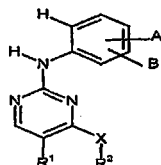
Prioridad: Folios: 372 a 432
29 de Mayo de 2001, Alemania, DE 10127581.1

Respuesta del solicitante: Folios: 652 a 662, Folios 704 a 708 y 806 a 838

2. Objeto de la invención

Título de la solicitud: **"PIRIMIDINAS INHIBIDORAS DE CDK, SU PREPARACIÓN Y USO COMO MEDICAMENTOS"**

El objeto de la presente solicitud de patente de invención consiste en obtener compuestos derivados de la pirimidina según fórmula general I con propiedades inhibidoras de las quinasas, dependientes de ciclina, y sus medicamentos.



Fórmula General I

Expediente: 03-112613

El problema planteado en esta solicitud es la necesidad de obtener compuestos derivados de la pirimidina, los cuales poseen efecto inhibitorio sobre las quinasas dependientes de ciclina (CDK) en cantidades nanomoleculares.

Los compuestos derivados de pirimidina presentados en esta solicitud, se usan para el tratamiento de diferentes enfermedades como por ejemplo cáncer, tumores sólidos y leucemia, y enfermedades autoinmunes entre otros.

Para solucionar este problema técnico la solicitud en estudio, el solicitante proporciona compuestos derivados de la pirimidina según fórmula general I, sus medicamentos y sus usos

Clasificación Internacional de Patentes (IPC.): A61K 31/505, A61K 31/506, C07D 239/48, C07D 239/47, 239/34, 239/42

3. De la solicitud de Patente

Descripción (artículo 28 D. 486)

La descripción cumple con el requisito de claridad o comprensión de acuerdo al art. 28 de la Decisión 486.

Reivindicaciones (artículo 30 D 486)

Realizadas las modificaciones por el solicitante al capítulo reivindicatorio, se encuentra que es claro y conciso y está lo suficientemente sustentado por la descripción, cumpliendo con el requisito de claridad, concisión y sustento en la descripción de acuerdo al art. 30 de la Decisión 486.

4. Unidad de invención (artículo 25 D 486)

La solicitud presentada cumple con el requisito de unidad de invención.

5. No es una invención (artículo 15 D 486 literal a)-f))

El capítulo reivindicatorio presentado por el solicitante corresponde a una invención.

6. Excepción a la patentabilidad (artículo 20 D486 literal a)-d))

El capítulo reivindicatorio presentado por el solicitante, es patentable.

7. Reivindicaciones relativas a un uso o a un segundo uso (artículos 14 y 21 D 486)

El capítulo reivindicatorio presentado por el solicitante, es patentable.

8. Análisis de la(s) Oposición(es): (artículo 43 D. 486):

No se presentaron oposiciones durante el estudio de patentabilidad de la solicitud presentada

9. Determinación del Estado de la Técnica

La fecha que se ha tenido en cuenta para determinar el Estado de la Técnica es:

- Fecha de presentación de la solicitud prioritaria: 29 de Mayo de 2001

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Considerando que la fecha de presentación de la primera solicitud de patente de la cual se reivindica prioridad es: 29/05/2001, no se encontraron documentos que divulguen los compuestos de la solicitud presentada

10. Análisis de patentabilidad (artículo 14 D486)

Novedad (artículo 16 D486)

Los compuestos presentados en la solicitud se consideran novedosos de acuerdo con las enseñanzas del estado de la técnica por la sustitución sulfonilo o sulfonamida en el anillo de fenilo en vez de nitrógeno, cadenas alquílicas, oxígeno, o azufre; y las sustituciones en 4 y 5 del anillo de pirimidina, como es un halógeno fijo y X como oxígeno o amino.

Nivel Inventivo (Art18 D486)

No se encontró dentro del estado de la técnica, tal como lo estipulado en el numeral 9, documento alguno que afecte el nivel inventivo.

La solicitud presentada posee nivel inventivo pues el solicitante resuelve el problema de obtener compuestos derivados pirimidinas los cuales poseen efecto inhibitorio sobre las quinasas dependientes de ciclina (CDK); las sustituciones nuevas no son obvias para el experto en la materia porque el análisis comparativo sobre el efecto de los compuestos en la línea celular MCF7 (Folio 707), evidencia su acción inhibitoria sobre las quinasas dependientes de ciclina CDK.

El problema técnico planteado en esta solicitud como obtener compuestos derivados de pirimidinas los cuales poseen efecto inhibitorio sobre las quinasas dependientes de ciclina (CDK); no sería solucionado fácilmente por el Experto en la Materia y queda plenamente resuelto en la solicitud presentada, demostrando que posee nivel inventivo de acuerdo con el artículo 18 de la Decisión 486.

12. Conclusión:

En consecuencia y por todo lo expuesto anteriormente, se recomienda **Conceder** la reivindicación: 1 a 3 (folios: 709-745).

Bogotá, D.C.,

Se le sugiere al solicitante modificar el título de la solicitud a "Derivados de pirimidinas, inhibidoras de CDK"

CONCEPTO TÉCNICO No. 0286	EXAMINADOR TÉCNICO Código No. 202070
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