



Trip Report for
“31st National Medicinal Chemistry Symposium”
University of Pittsburgh, Pittsburgh, Pennsylvania
June 15–19, 2008

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Abstract: *The 31st National Medicinal Chemistry Symposium was held at the University of Pittsburgh in Pittsburgh, Pennsylvania on June 15–19, 2008. The meeting highlighted some of the recent developments in the field of Medicinal Chemistry. The oral session topics included: Hepatitis C, Virtual Library Design and Screening, Molecular Approaches to Cancer Therapy, Inflammatory/Immunology, Late-Breaking/First-Time Disclosures and Biomarkers and Nucleic Acid Targeting Strategies. This report highlights selected material from the oral presentations as well as the posters presented.*

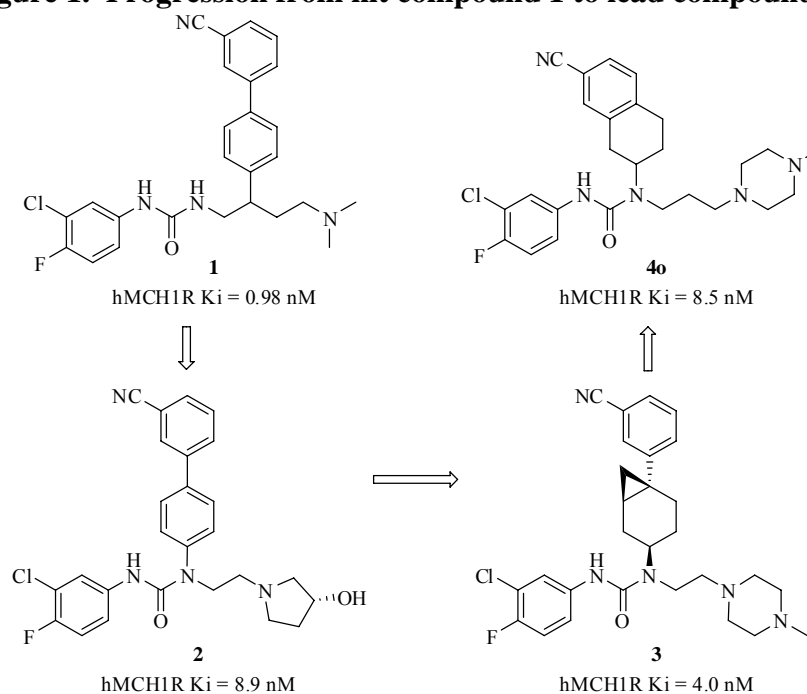
“Discovery of Tetralin Ureas as Potent MCH1R Antagonists”

Tao Guo, Pharmacopeia, Inc., San Diego, CA

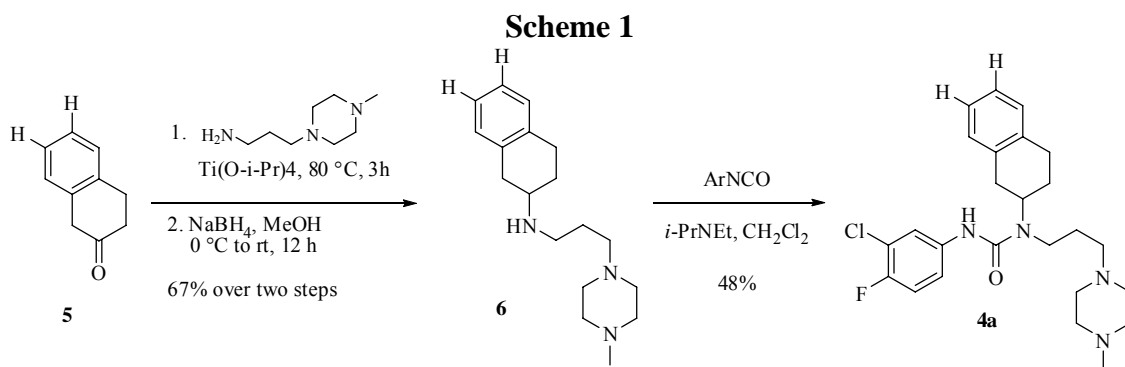
Obesity is a growing concern in industrialized countries throughout the world. In the United States, over 30% of adults are obese. Currently approved weight loss drugs such as orlistat and sibutramine suffer from limited efficacy and undesirable side effects that limit patient compliance, and therefore new medications are needed. Melanin concentrating hormone (MCH) is a 19 amino acid neuropeptide involved in the regulation of food intake and energy balance. Mice lacking MCH-1 receptors are lean and resistant to obesity. Hence, antagonism of the melanin concentrating hormone receptor 1 (MCHR1) shows promise as a therapeutic target for the treatment of obesity.

Researchers at Pharmacopeia, Inc. worked in collaboration with Schering-Plough to discover novel antagonists of MCHR1. A lead hopping approach was utilized, starting from biphenyl urea **1** (Figure 1). The biphenyl group was moved to one of the urea nitrogens to give compound **2**. One of the phenyl rings was replaced with a bicyclo[4.1.0]heptyl ring system to give compound **3**. Finally, the bicyclo/phenyl group was replaced with substituted tetralin to provide compound **4a**.

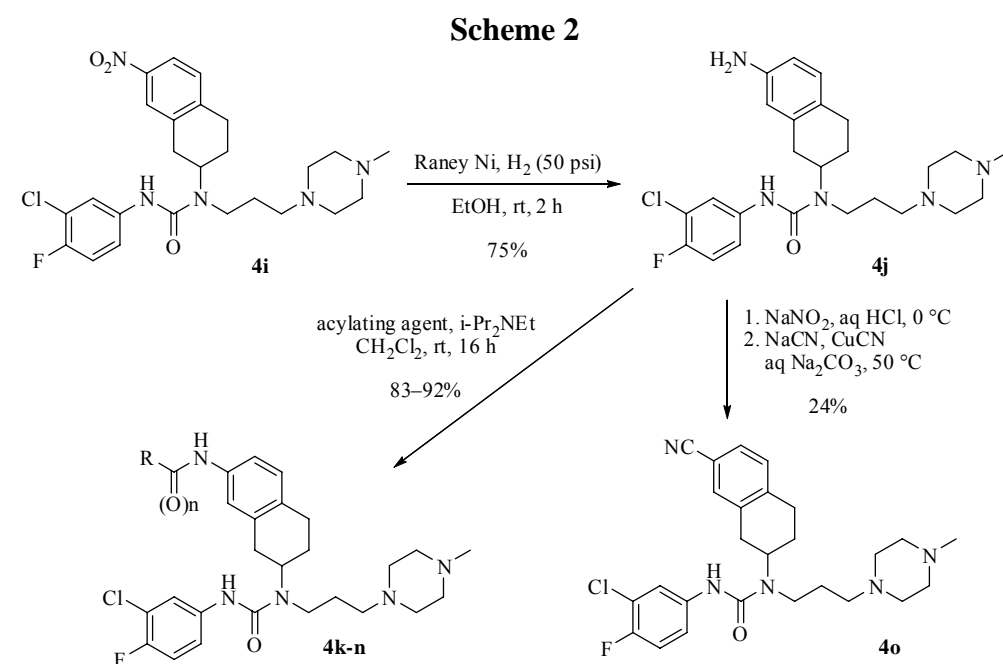
Figure 1. Progression from hit compound 1 to lead compound 4a.



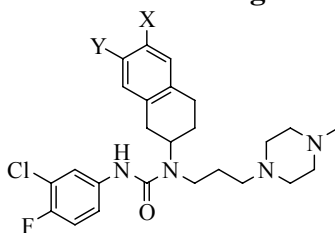
A variety of substituted tetralin derivatives were described. The synthesis of these compounds was accomplished as shown in Scheme 1 for compound **4a**. Starting from the desired β -tetralone, reductive amination installed the aminopropylpiperazine portion of the molecule. Treatment of this compound **6** with 3-chloro-4-fluorophenyl isocyanate gave the desired product **4a**.



Additional elaboration of tetralin derivative **4i** provided analogues **4j–4o** (Scheme 2).



The SAR of the substituted tetralin analogues is shown in Table 1. Substitution at the 6-position of the tetralin ring system (compounds **4b–4d**) was tolerated. Substitution at the 7-position with a sulfone or sulfonamide (compounds **4f–4h**) resulted in an approximately 3 to 8-fold loss in MCH1R binding affinity compared to unsubstituted analogue **4a**. However, substitution at the 7-position with a nitro, acetamide or cyano group (compounds **4i**, **4k** and **4o**, respectively) provided 30 to 60-fold enhancement in binding affinity.

Table 1. MCH1R Binding Affinity SAR

Compound	X	Y	hMCH1R Ki (nM)	Compound	X	Y	hMCH1R Ki (nM)
4a	H	H	380	4i	H	NO ₂	11
4b	Br	H	180	4j	H	NH ₂	260
4c	CN	H	340	4k	H	NHCOMe	6
4d	OMe	H	190	4l	H	NHCO- <i>i</i> -Pr	61
4e	H	OMe	92	4m	H	NHCONHEt	160
4f	H	SO ₂ Me	1300	4n	H	NHSO ₂ Me	96
4g	H	SO ₂ NHMe	3000	4o	H	CN	8.5
4h	H	SO ₂ NMe ₂	3000				

In vivo data was reported for the compounds **4k** and **4o** (Table 2). Compound **4o** demonstrated rat pharmacokinetics and mouse *ex vivo* MCH1R receptor binding consistent with progression to an *in vivo* efficacy model. However, no efficacy data was provided.

Table 2. Selected *in vivo* data for compounds 4k and 4o.

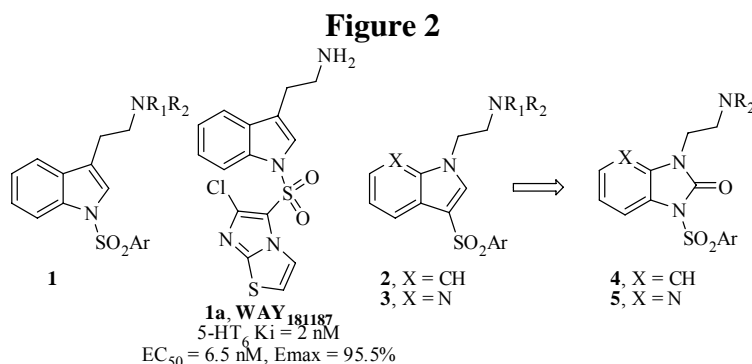
Compound	Rapid rat AUC _{0-6h} (10 mg/kg, po, ng•h/mL)	Mouse <i>ex vivo</i> MCH1R binding (30 mg/kg, po, 6h)
1k	195	23%
1o	642	84%

“1-(Arylsulfonyl)-3-aminoethyl-1,3-dihydro-2H-benzimidazol-2-one and 1-(Arylsulfonyl)-3-aminoethyl-1,3-dihydro-imidazo[4,5-*b*]pyridine-2-one as 5-HT₆ Receptor Ligands”

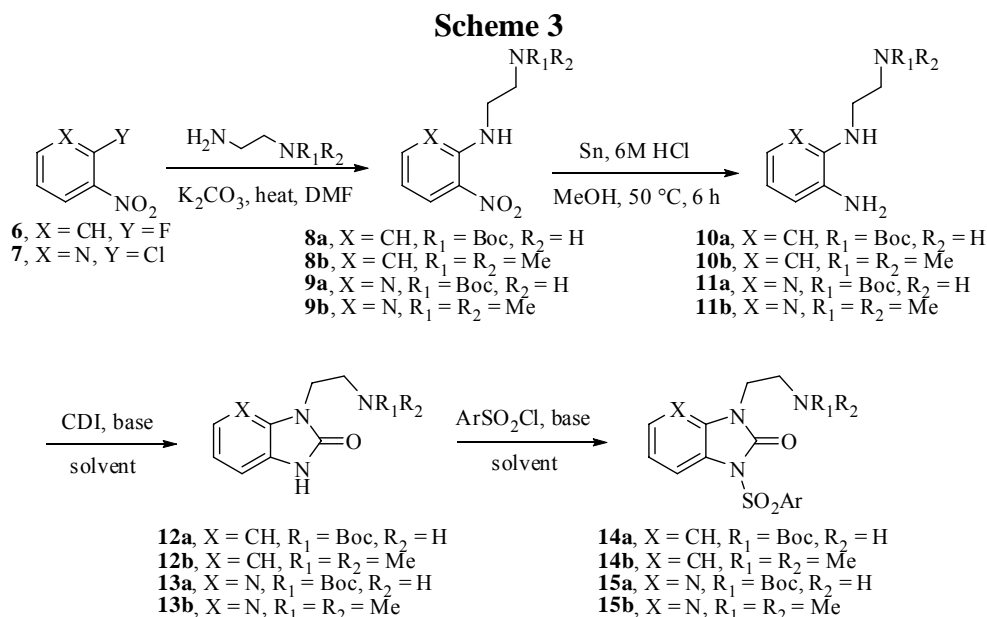
Derek Cole, Wyeth Research, Pearl River, NY

The 5-hydroxytryptamine-6 (5-HT₆) receptor is a serotonin receptor located almost exclusively in the central nervous system that has been implicated in learning and memory functions. A variety of selective 5-HT₆ antagonists have been reported in the literature, including compounds from Roche and SmithKline. However, the focus of this research was on the discovery of selective *agonists* as tools to better understand the effects of 5-HT₆ agonism *in vivo*. Previous studies from Wyeth had identified tryptamine-based 5-HT₆ modulators **1** with agonist activity, depending on the arylsulfonyl and other substituents (Figure 1). For example, compound **1a** (WAY-181187) was identified as a

selective 5-HT₆ agonist with activity *in vivo* in a schedule-induced polydipsia model for obsessive compulsive disorders. The “flipped” tryptamine-like analogues **2** were found to be relatively weak agonists, while the azaindole compounds **3** were generally full agonists. Based upon these results, the core was modified further to give 1,3-dihydro-2*H*-benzimidazol-2-one and 1,3-dihydro-imidazo[4,5-*b*]pyridin-2-one derivatives **4** and **5**, respectively.



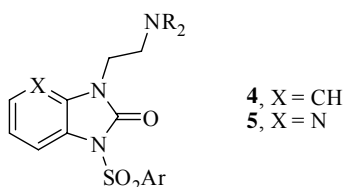
A variety of arylsulfonyl analogues of compounds **4** and **5** were prepared starting from 2-halo-1-nitrobenzenes **6** or **7** (Scheme 3). Halide displacement with primary amines gave compounds **8** and **9**. Tin-mediated nitro reduction followed by cyclization with carbonyldiimidazole gave benzimidazolone and imidazopyridinone derivatives **12** and **13**, respectively. Sulfonylation and optional deprotection provided a library of analogues of **4** and **5**.



Compounds **4** and **5** were tested for 5-HT₆ affinity in a [³H]-LSD radioligand binding assay. The results for a selected set of compounds are shown in Table 3. Despite the increased polarity of the benzimidazolone core relative to indole, compounds **4** retained 5-HT₆ affinity. Primary and tertiary amines showed comparable 5-HT₆ affinities. For example, compound **4a** had a 5-HT₆ K_i value of 23 nM, while the dimethylamine

derivative **4b** had a 5-HT₆ K_i value of 17 nM. In general 1,3-dihydro-imidazo[4,5-*b*]pyridine-2-one derivatives **5** showed improved 5-HT₆ affinity relative to benzimidazolones **4**. For example, 2-naphthyl derivative **5a** showed a 5-fold enhancement in affinity compared to benzimidazolone **4a**. The functional activity of selected analogues was determined in a cAMP cyclase assay. Many of the compound **4** derivatives showed partial agonist activity. However, several of the compound **5** derivatives such as **5c** and **5d** were full agonists. The most potent analogue of both series was partial agonist **5b**, with an EC₅₀ of 9.6 nM.

Table 3. 5-HT₆ binding and cyclase data.



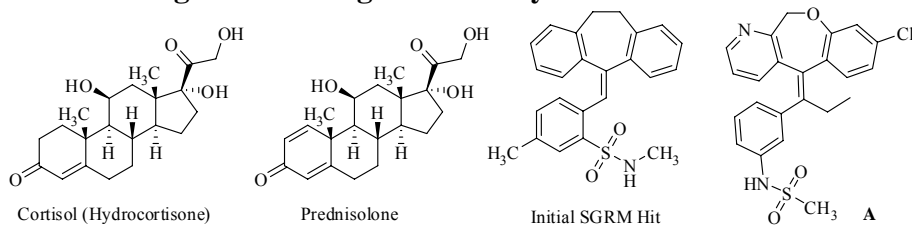
Compound	X	Ar	R	5-HT ₆ K _i (nM)	EC ₅₀ (nM)	E _{max} (%)
4a	CH	2-naphthyl	H	23 ± 3	NR	NR
4b	CH	2-naphthyl	Me	17 ± 2	141 ± 7 (ant)	66 ± 1
4c	CH	6-Cl-imidazothiazol-5-yl	H	5.6 ± 0.9	25 ± 3 (ag)	66 ± 1
4d	CH	6-Cl-imidazothiazol-5-yl	Me	20 ± 2	167 ± 46 (ag)	84 ± 2
4e	CH	5-Br-thien-2-yl	H	26 ± 4	NR	NR
5a	N	2-naphthyl	H	4.5 ± 0.7	40 ± 9 (ag)	55 ± 2
5b	N	6-Cl-imidazothiazol-5-yl	H	3.7 ± 0.4	9.6 ± 0.5 (ag)	72 ± 0.4
5c	N	5-Br-thien-2-yl	H	8.7 ± 1.2	116 ± 8 (ag)	100 ± 0
5d	N	2,5-Cl-thien-3-yl	H	22 ± 4	284 ± 100 (ag)	100 ± 0

“Preclinical Characterization of a Selective Glucocorticoid Receptor Modulator (SGRM)”

Michael Cogan, Eli Lilly & Company, Indianapolis, IN

The glucocorticoid receptor (GR) is part of the super-family of hormone-activated nuclear receptors and is present in almost every cell in the body. Cortisol, the natural ligand for the GR, is shown in Figure 3. Cortisol (also known as hydrocortisone) enters the nucleus of the cell and binds to the GR, which forms a dimer with another GR. The dimer associates with specific areas of DNA and attracts RNA polymerase and other transcription factors. This action regulates gene expression which controls the development, homeostasis and metabolism of the organism. Other hormones in this class are estrogen, testosterone, vitamin A and vitamin D.

Figure 3. Endogenous and synthetic SGRM's.



Cortisol has many physiological effects, regulating a range of processes from insulin production and immune responses to potassium excretions and memory retention. Pharmacologically, it is most often used as an anti-inflammatory. Topical formulations are available over the counter for allergic rashes, eczema, psoriasis and most skin conditions. Prednisolone (Figure 3) has 4 times the anti-inflammatory effects as cortisol and both can be used as injections into diseased joints to treat conditions such as gout and rheumatoid arthritis. Although these treatments are effective, the lists of side-effects are long and diverse and therefore cortisol injections are severely limited in their application for joint pain management.

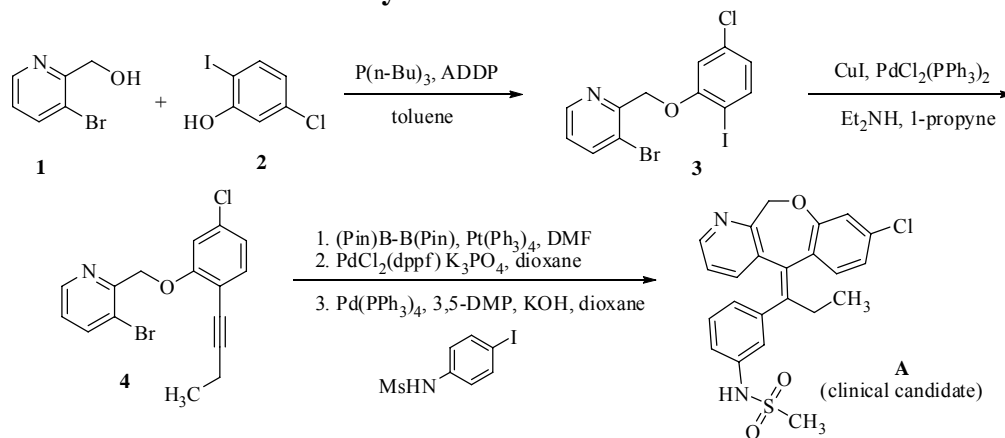
The researchers at Eli Lilly initiated their SRGM campaign with a high-throughput screening of the GAMPE panel of compounds against GR binding. The therapeutic endpoint is to have a small molecule for the treatment of rheumatoid arthritis and immune disorders. The initial hit from their screen was a tricyclic phenyl sulfonamide (Figure 3). The presenter, Michael Coglean, disclosed the initial hit but would not reveal how the SAR was developed. He did disclose the structure of the clinical candidate **A** (Figure 3).

The biological activity of **A** is documented in the Eli Lilly patent application PCT WO 2008008882. Compound **A** binds to the GR with a K_i of 0.35 nM and is at least 30-fold selective against other nuclear hormone receptors for mineralocorticoid (117 nM), androgen (16 nM) and progesterone (31 nM), and no binding to the estrogen receptor was observed. In cells **A** is a full, potent transrepressor of GR and has an EC_{90} of 4.8 and 30 nM for GR induced production of inflammatory mediators interleukin-6 (IL-6) and tumor necrosis factor- α (TNF- α), respectively. In mice models of inflammation, compound **A** exhibited an ED_{50} of 0.31 and 1.2 mg/kg for inhibition of IL-1 β production and carrageenan induced paw weight gain, respectively. These activities are roughly 5 times more potent than the activity reported for prednisolone. One major side effect of corticoid treatments is induced osteoporosis, and **A** displayed reduced ability to elicit GR-mediated bone effects. Particularly in mice, compound **A** induced little reduction in mouse serum osteocalcin compared to prednisolone at their respective ED_{50} 's.

The synthesis of compound **A** has been published by the discovery research department at Eli Lilly (*Org. Lett.* **2008**, *10*, 2701). Shown below in Scheme 4 is the route for the clinical candidate **A**. Starting with bromopyridyl methanol **1**, the ether intermediate **3** was formed with phenol **2** under Mitsunobu conditions. A regioselective Sonogashira coupling with the iodide was accomplished to give the ethyl alkyne **4**. The synthesis was completed by tandem intra- and intermolecular coupling reactions. First, the alkyne was subjected to diboration with bis(pinacolato)diboron, which gave the *syn*-diboration

product. The second step was to form the 7-membered ring with an intramolecular Suzuki reaction. Thirdly, the remaining pinacolatoboron moiety was displaced by the *para*-iodoaniline to give the final product. With this route, one could envision rapid SAR development by easily replacing any of the three aryl halide pieces.

Scheme 4. Synthesis of clinical candidate A.



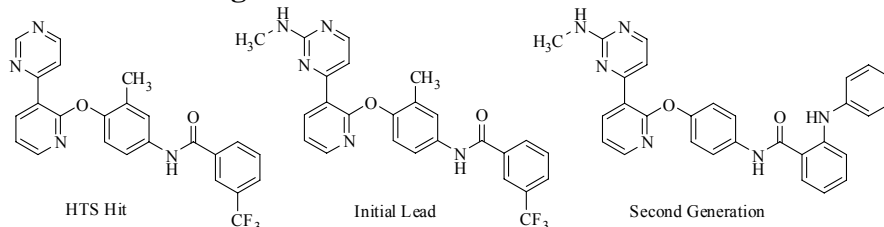
“Synthesis and SAR of Pyridinyl Anthranilamides as Potent, Selective, and Orally Bioavailable Aurora Kinase Inhibitor”

Phillip Olivieri, Amgen, Cambridge, MA

The Aurora kinases (A, B and C) are a family of serine/threonine kinases that play essential roles in cell division, including the regulation of chromosome segregation and cytokinesis. More specifically, Aurora A and B are required for correct function of centrosomes and attachment of the mitotic spindle to the centrosomes, respectively. Little is known about the function or role of Aurora C. The protein domains are organized similarly to other kinases with a highly conserved C-terminal ATP site and an N-terminal substrate binding site, which is specific to each kinase giving rise to its substrate specificity. Defects in the function and expression level of Aurora kinases A and/or B are highly associated with tumorigenesis and therefore, these kinases have generated substantial interest as targets for the treatment of cancer.

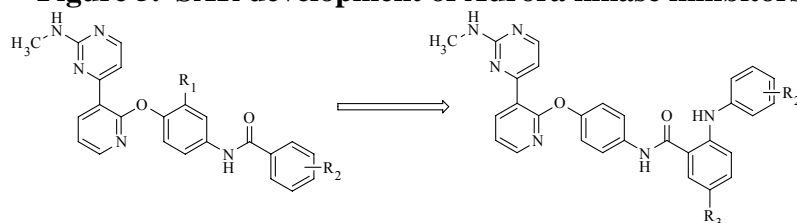
Researchers from Amgen, Inc. initiated an Aurora kinase campaign with a high throughput screening. The HTS hit (Figure 4) was a pyridinyl pyrimidine amide with an IC_{50} of 250 nM for Aurora A. Quickly, it was found that adding a methyl amine group at the 2-position of the pyrimidine increased the IC_{50} 10 fold to 19 nM, which gave their initial lead compound (Figure 4). X-Ray crystal structures revealed the key interactions of the initial lead with Aurora A. The methylamine and pyrimidine nitrogens form donating and accepting hydrogen bonds, respectively, with the hinge region of the kinase, which normally forms hydrogen bonds with the adenosine of ATP. The remaining interactions with the DFG loop are mainly hydrophobic. Actually, the initial lead forms an induced fit with the flexible DFG loop, generating a conformation not observed in the other pyrimidine analog co-crystal structures.

Figure 4. Aurora Kinase Inhibitors.



An investigation into the SAR was initiated by varying the substitution on the two phenyl rings (Figure 5). With a methyl group optimal at R₁, 3- and 4-substitutions at R₂ were tolerated while substitution at the 2-position was deleterious. This first effort reveals how the initial lead was obtained (Figure 4). Although these analogues possessed enzymatic activity, they exhibited no activity in cellular based assays. The researchers found that substitution of the terminal phenyl ring with an anthranilamide system was advantageous for cellular activity. The cells used in the assay were HeLa cells, which are cervical cancer cells. In the anthranilamide analogs, a range of groups are accepted at different positions at R₂ in the enzymatic assay. Bulky substituents at R₂ degraded cellular activity. Various small groups (H, F, OCH₃) at R₃ were tolerated.

Figure 5. SAR development of Aurora kinase inhibitors.



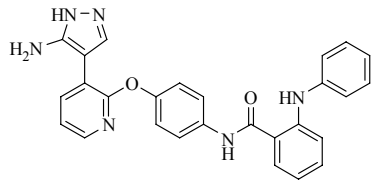
Enzyme IC ₅₀ (nM)				Enzyme IC ₅₀ (nM)		Cell EC ₅₀ (nM)
R ₁	R ₂	Aurora A	Aurora B	R ₂	R ₃	24 h ploidy
H	3-CF ₃	70	118	H	H	15
Me	3-CF ₃	19	66	H	H	89
Me	3-CF ₃ - 4-dimethylamine	55	80	H	F	20
H	3-Br	36	107	3-CF ₃	H	180
Me	3-pyrrole	22	99	4-F	H	28
Me	H	151	1053	4-F	F	38
Me	3-Cl	69	430	2-Cl	OMe	25
Me	4-Cl	50	335	3-F	H	28
Me	2-Cl	292	36000	4-Cl	H	23
				3-pyridyl	H	217
				2,3-dimethyl	H	58

The second generation compound (Figure 4) inhibits Aurora A and B with an IC₅₀ of 15 and 9 nM, respectively. The EC₅₀ against HeLa cells is 15 nM. With this promising profile, preliminary ADMET data was obtained for this compound. The *in vitro* metabolism was 466 and 72 μL/min/mg in rat and human, respectively. At a dose of 2 mg/kg *i.v.*, the clearance was 3.4 L/h/kg in rat. The half life was 1.8 h and %F was less

than 1. To conclude, the second generation compound suffers from high clearance and low bioavailability.

In order to address the poor ADMET profile, the researchers replaced the hinge binding pyrimidine portion. They exchanged the 2-methylamino-pyrimidine with an azaindole, pyrazole, oxazole, and substituted pyrimidines. All the substitutions retained activity except the oxazole. The 2-amino-pyrazole possessed the highest enzymatic activity and was therefore submitted to ADMET studies. The results are shown in Figure 6. Sacrificing some cellular potency, the new lead has an improved profile from the second generation. However, the new lead still has relatively poor bioavailability, suffering from short half-life, low %F, and low solubility.

Figure 6. New Lead compound and ADMET data.

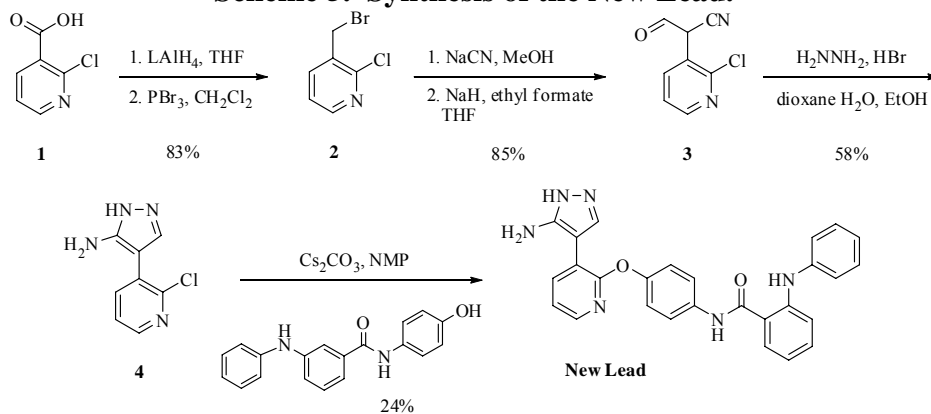


New Lead

Enzyme IC ₅₀ (nM)	AurA	7
	AurB	4
HeLa Cell EC ₅₀ (nM)	24 h 4N+	63
In vitro metabolism (μL/min/mg)	rat / human	82 / 43
Rat PK (male)	IV 2 mpk	
	CL (L/h/kg)	1.5
	t _{1/2} (h)	1.7
	PO 5 mpk	
	% F	9
Solubility (mg/mL)	HCl / PBS / SIF	0.007 / 0.001 / 0.032

Shown below in Scheme 5 is the synthetic route to the 2-amino-pyrazole lead. Starting with 2-chloronicotinic acid (**1**), reduction to the benzyl alcohol is followed by bromination to give intermediate **2**. After displacement of the bromide with a cyano group, a formyl group is added to give aldehyde **3**. Pyrazole ring formation is accomplished with hydrazine under acidic conditions yielding **4**. The 2-position chloride is displaced with the 4-hydroxyphenyl-benzamide under basic conditions to give the final compound.

Scheme 5. Synthesis of the New Lead.

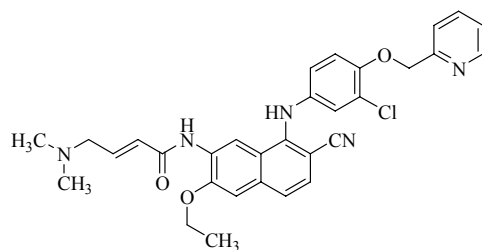


“Cancer Therapeutic Modalities with Irreversible Kinase Inhibitors”

Tarek S. Mansour, Wyeth Research

The central role of protein kinases in regulating all cellular signaling pathways that are associated with diseases have positioned them as druggable targets by small molecule inhibitors in oncology and nononcology indications, including autoimmune disease, metabolic and central nervous system disorders. In eukaryotic cells, the primary mechanism for regulating protein function is associated with a phosphorylation reaction of either a serine, threonine or tyrosine residue. High-throughput screening has established itself as a viable platform for kinase drug discovery. In general, most kinase inhibitors that advance in preclinical and clinical studies target the conserved adenosine triphosphate (ATP) binding site. However, recent studies highlighted the roles of reactive amino acids such as cysteine and lysine residues that form a covalent bond with the inhibitor, which renders these irreversible inhibitors with distinctive structural features as alternatives to ATP competitive ones.

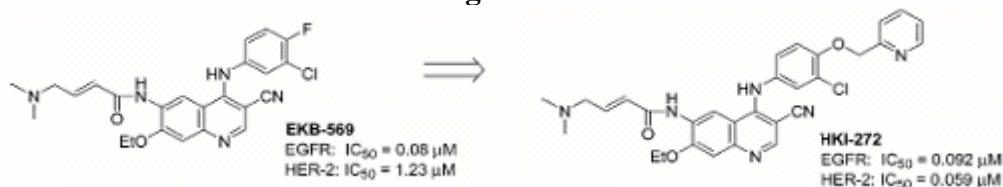
In oncology, irreversible kinase inhibition has been reported for the epidermal growth factor receptor (EGFR) and ErbB receptors, phosphoinositide 3'-kinase (PI3K) and protein kinase B (AKT). Irreversible inhibitors of these kinases such as EKI-785, EKB-569 (pelitinib), HKI-272 (neratinib), wortmannins and pyranonaphthoquinones are effective against several cancers. This presentation highlighted molecular insights into the inhibition of these important biological targets by irreversible inhibitors discovered at Wyeth. The discussion below will focus on the medicinal chemistry development of HKI-272 (**25o**).



HKI-272 (**25o**)

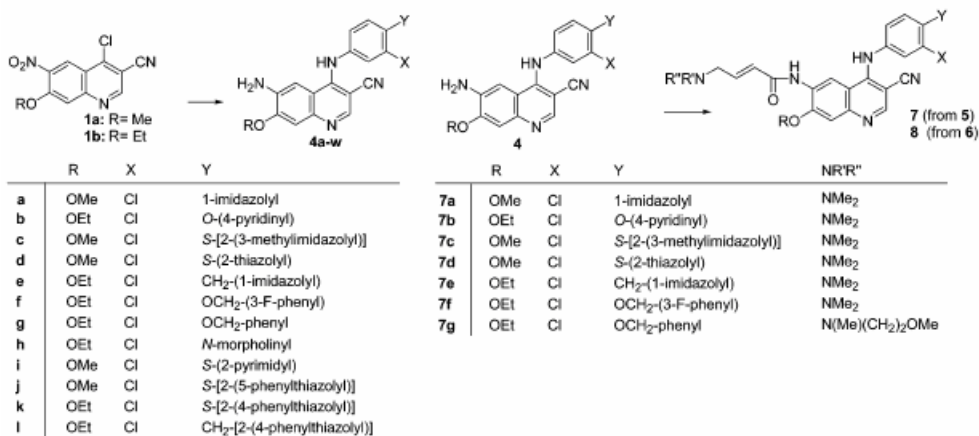
HKI-272 (**25o**) was developed as an irreversible inhibitor of human epidermal growth factor receptor-2 (HER-2) and epidermal growth factor receptor (EGFR) kinases (H.-R. Tsou, *et al.*, *J. Med. Chem.* **2005**, *48*, 1107-131). HKI-272 (**25o**) demonstrated enhanced activities for inhibiting HER-2 kinase and the growth of HER-2 positive cells compared to EKB-569, an EGFR kinase inhibitor also developed at Wyeth (Figure 7).

Figure 7

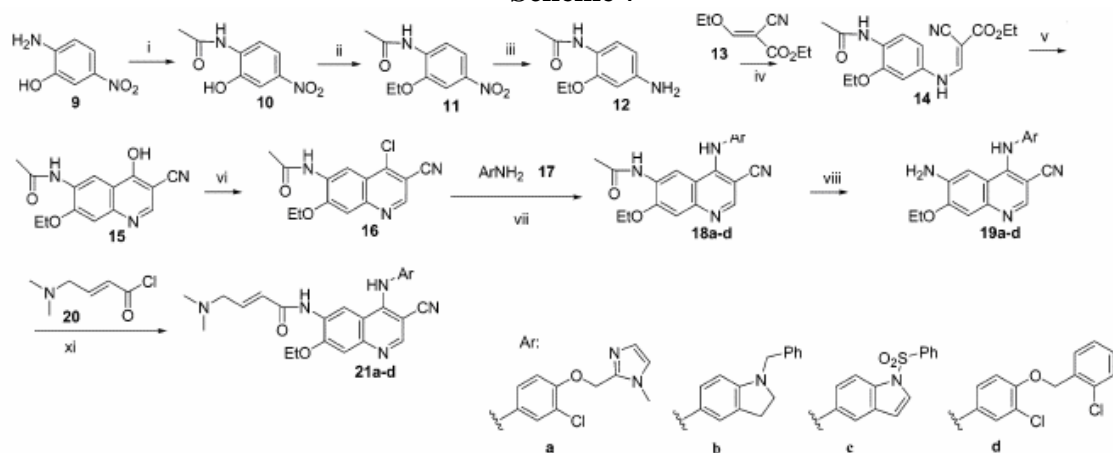


The synthetic routes that were used to prepare a majority of the analogues are shown in Schemes 6 through 8. They were mostly prepared by acylation of 6-amino-(4-aryl-amino)-quinoline-3-carbonitriles with unsaturated acid chlorides or by amination of 4-chloro-6-(crotonamido)-quinoline-3-carbonitriles with monocyclic or bicyclic anilines. The third route was developed to prepare a key intermediate, 6-acetoamido-4-chloroquinoline-3-carbonitrile, that involved a safer cyclization step. The researchers at Wyeth showed that attaching a large lipophilic group at the *para* position of the 4-(arylamino) ring resulted in improved potency for inhibiting HER-2 kinase.

Scheme 6



Scheme 7



^a (i) Ac₂O, HOAc, 60 °C; (ii) EtBr, DMF, K₂CO₃, 60 °C; (iii) H₂, Pd/C, THF; (iv) toluene, 90 °C; (v) Dowtherm, 250 °C; (vi) POCl₃, diglyme, 100 °C; (vii) pyridine hydrochloride, *n*-PrOH or methoxyethanol, reflux; (viii) aqueous HCl, reflux; (xi) CH₃CN, *N*-methyl-2-pyrrolidone or DMF, from 0 °C to room temperature.

Scheme 8

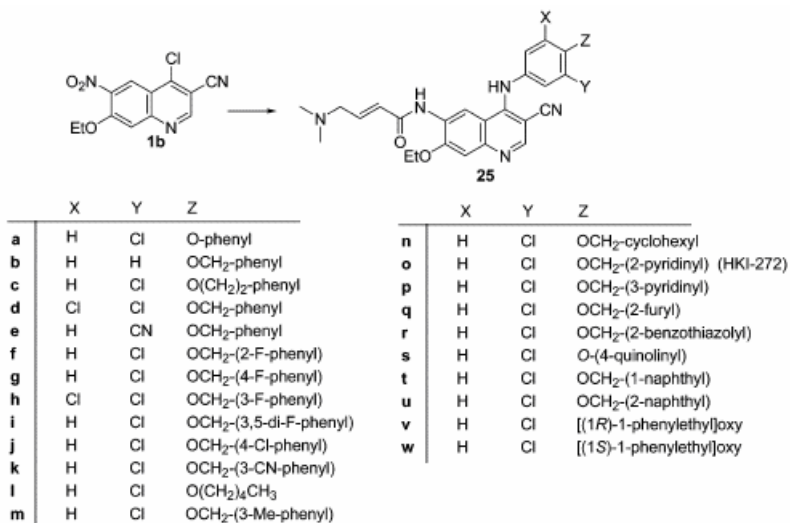
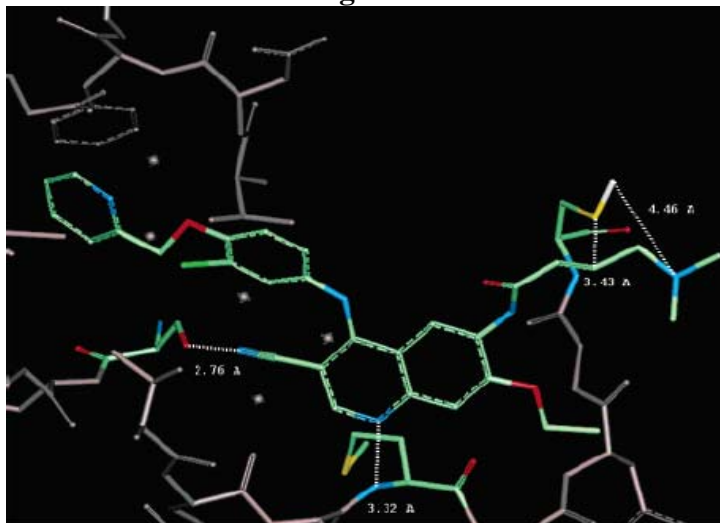


Figure 8 illustrates the importance of the basic dialkylamino group at the end of the Michael acceptor for activity, due to the intramolecular catalysis of the Michael addition (Cys 805 in the HER-2–HKI-272 (**25o**) complex).

Figure 9



HKI-272 (**25o**) is currently being evaluated in a Phase I/II study in combination with Paclitaxel in subjects with advanced tumors and breast cancer and a Phase II study evaluating HKI-272 in subjects with advanced breast cancer. Furthermore, a safety and tolerability study is being carried out (oral administration) with Japanese subjects having advanced solid tumors and in a Phase I/II study in combination with Vinorelbine in subjects with solid tumors and metastatic breast cancer. Additionally, HKI-272 is being evaluated in a Phase I study to determine the mass balance and metabolic distribution of orally administered compound, and in a separate Phase I study HKI-272 is being evaluated for cardiac repolarization (heart rhythms).

For further information on HKI-272 and the development of EKB-569, see: (a) S. K. Rabindran, *et al.*, *Cancer Research* **2004**, 64, 3958-3965. (b) X. Zhang, *et al.*, *J. Clinical Oncology* **2008**, 26, 1742-1751. (c) C. H. Yun, *et al.*, *Proc. Natl. Acad. Sci. USA* **2008**, 105, 2070-2075. (d) A. Wissner, *et al.*, *J. Med. Chem.* **2003**, 46, 49-63 and also www.clinicaltrials.gov for US-based clinical trials.

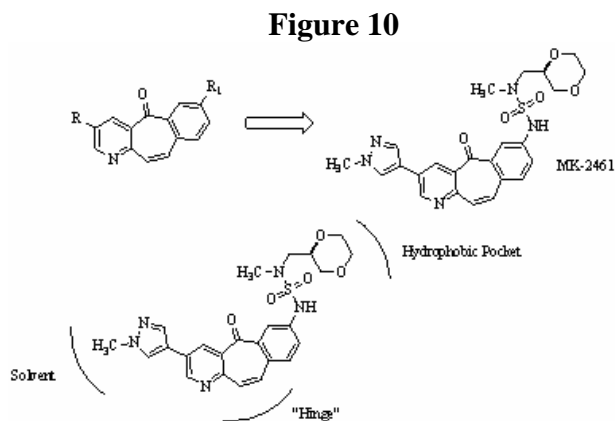
“Discovery of a Unique Class of c-Met Inhibitors for the Treatment of Cancer”

Jason D. Katz, Merck Laboratories

Dr. Katz discussed the unique class of compounds which contain the 5*H*-benzo[4,5]-cyclohepta[1,2-*b*]pyridine scaffold. These compounds are inhibitors of tyrosine kinases, in particular the receptor tyrosine kinase MET, and are useful in the treatment of cellular proliferative diseases, for example cancer, hyperplasias, restenosis, cardiac hypertrophy, immune disorders and inflammation.

Recently, members of the MET proto-oncogene family, a subfamily of receptor tyrosine kinases, have drawn special attention to the association between invasion and metastasis. The MET family, including MET (also referred to as c-Met) and RON receptors, can function as oncogenes like most tyrosine kinases. MET has been shown to be over expressed and/or mutated in a variety of malignancies. A number of MET activating mutations, many of which are located in the tyrosine kinase domain, have been detected in various solid tumors and have been implicated in invasion and metastasis of tumor cells. A number of reviews on MET and its function as an oncogene have recently been published.

This presentation focused on the first time disclosure of the development of MK-2461 within the Merck labs. The binding site for MK-2461 was described as having a hinge region, required for potency, and both a hydrophobic and solvent pocket that were thoroughly explored through SAR analysis.



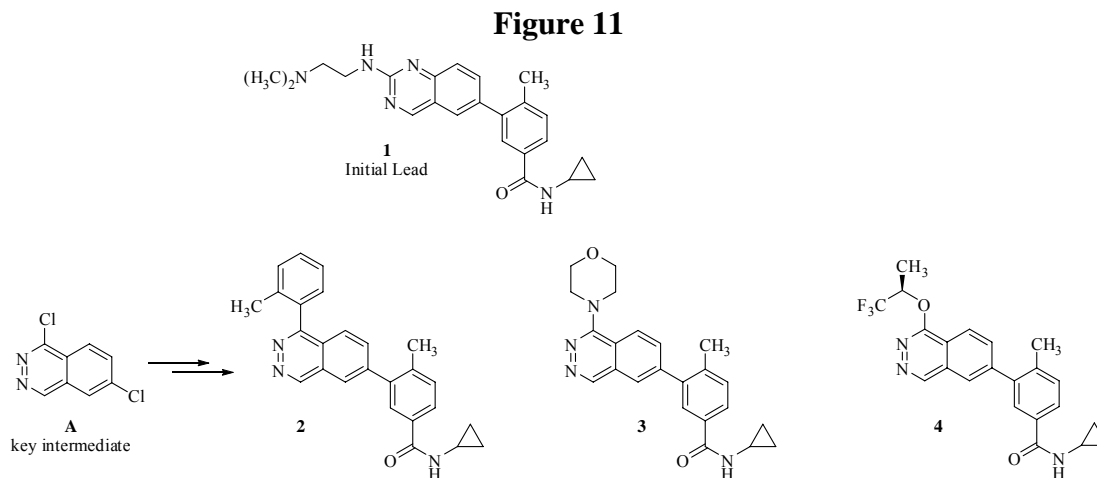
MK-2461 is currently in both phase I studies for patients with advanced cancer and in a phase I/II clinical trial in patients with advanced solid tumors both within the United States. For more information on this series, see: (a) WO Patent 2007/002258 A2. (b)

Cancer and Metastasis Review **2003**, 22, 309-325. (c) *Nature Reviews Molecular Cell Biology* **2003**, 4, 915-923. (d) *Nature Reviews Cancer* **2002**, 2, 289-300. (e) WO Patent 2008/008310 A2. (f) WO Patent 2007/050401 A2.

“Structure-based design Potent and Highly Selective p38 MAP Kinase Inhibitors”

Andrew S. Tasker, Amgen, Inc.

Amgen sought to develop a P38 Map kinase inhibitor for the treatment of rheumatoid arthritis (RA) utilizing a structure-based approach, which considered not only the structure of the target but that of other kinases to achieve maximal efficacy with minimal side effects. This research program set a high bar for activity and safety as there are already drugs in this area, namely Humira (Abbott) and Enbrel (Amgen/Wyeth). Amgen sought to apply their vast in-house knowledge of kinases and kinase screens to develop a potent, selective compound.



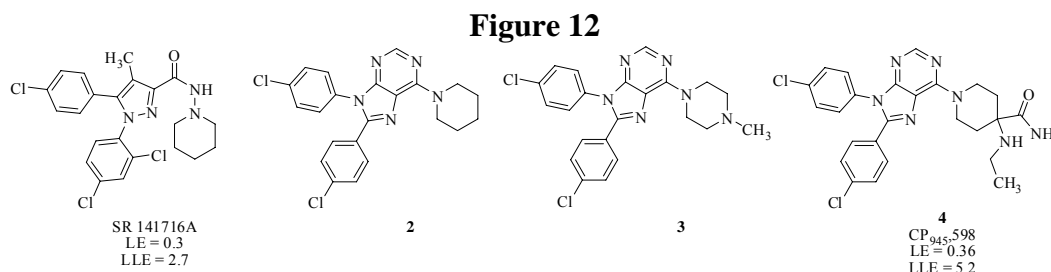
The initial Amgen lead compound **1** (Figure 11, WO 2006/039718) was active against collagen-induced RA with a dosage of 1 mg/kg, but had some problems with hERG activity and PK. X-Ray structures of **1** bound in the active site showed a good fit but revealed empty space above and below the compound. To exploit this unoccupied space, the medicinal chemists employed key intermediate **A** to generate new compounds on the order of **2**.

Optimization of the binding led to **3** which was potent, active ($ED_{50} = 0.06$ mg/kg), and had good hERG activity, but which required BID (1 mg/kg) dosing because of a short half-life: once a day dosing would have required 14 mg/kg dosing. Continued work led to the development of **4** which showed high bioavailability (>70%), very slight hERG activity, and allowed for a single dose daily at 0.8 mg/kg. Compound **4** has shown remarkable selectivity for P38 MAP kinase with an IC_{50} of 300 nM against one kinase and over 1000 nM for all other kinases screened. These compounds are covered in WO 2008/030466.

“Discovery of 1-[9-(4-chlorophenyl)-8-(2-chlorophenyl)-9H-purin-6-yl]-4-ethylaminopiperidine-4-carboxylic acid amine hydrochloride (CP-945,598): a novel, potent, and selective cannabinoid CB1 receptor antagonist”

David Griffith, Pfizer

This talk described the development of CP-945,598 which is currently in Phase III clinical studies for treatment of obesity. The Pfizer group began with the concept of generating constrained analogues of Sanofi compound SR 141716A. The medicinal chemistry group generated **3** (Figure 12) which showed good activity ($K_i = 5$ nM in rat and human) and better solubility than **2** but which had some hERG activity ($IC_{50} = 350$ nM). The addition of small polar groups to alleviate the hERG activity led to the development of CP-945,598 (**4**), which featured improved activity (Rat $K_i = 2.8$ nM, human $K_i = 0.8$ nM), better hERG activity ($IC_{50} > 3000$ nM), and a brain to plasma ratio of 0.1. Animal studies on CP945,598 demonstrated that the weight loss was due to fat consumption resulting from increased activity and not just due to decreased food intake.



During the course of the presentation, the use of ligand efficiency (LE) and ligand-lipophilicity efficiency (LLE) in evaluating potential lead compounds was discussed. LE and LLE have been widely used in fragment based drug discovery but are becoming more common in traditional medicinal chemistry programs. Ligand efficiency is useful for comparing potential lead compounds from several series and is defined as the binding energy divided by the number of heavy atoms (HAC (heavy atom count, all atoms in a molecule except for hydrogen atoms):

$$LE = -\Delta G/HAC \approx -RT \ln(IC_{50})/HAC$$

In evaluating lead compounds, given the preference that a drug candidate has a molecular weight less than 500 and IC_{50} of less than 10 nM, an LE value of at least 0.3 is required.

LLE is a revised approach which takes into consideration the observation that more lipophilic drug candidates often have more side effects resulting from a greater number of non-specific interactions. LLE is defined by the equation: $LLE = pIC_{50}$ (or pK_i) - $cLogP$ (or $LogD$). For a nanomolar potent compound, the ideal LLE value would be in the range of 5-7.

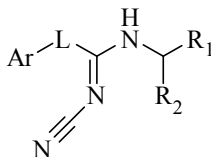
As shown in Figure 12, over the course of the Pfizer program, the LE value increased only modestly from 0.30 in the Sanofi compound to 0.36 for the final drug candidate. In

contrast, the value of LLE increased from 2.7 to 5.2 which may indicate a cleaner side effect profile for the final drug candidate.

“Synthesis and SAR of Novel Cyanoamidines as P2X7 Receptor Antagonists”

S. Peddi, A. S. Florjancic, A. Perez-Medrano, Y. Wang, D. L. Donnelly-Robert, C. R. Faltynek, G. Grayson, M. F. Jarvis, M. T. Namovic and William A. Carroll, Neurological Diseases Research, Global Pharmaceutical Research and Development, Abbott Laboratories

The P2X7 receptor is a member of the P2X family of ATP-activated ion channels and is a very attractive therapeutic target for treatment of inflammatory and neuropathic pain conditions. A series of aryl and heteroaryl cyanoamidines with varying amino substitution was designed and synthesized. Compounds were evaluated for activity to inhibit calcium flux in both human and rat recombinant P2X7 cell lines. The synthesis, SAR and in vitro data of these compounds will be discussed.



P2X receptors are a family of cation-permeable ligand gated ion channels that open in response to the binding of extracellular adenosine 5'-triphosphate (ATP). They belong to a larger family of receptors known as the purinergic receptors. P2X receptors are present in a diverse array of organisms including humans, mouse, rat, rabbit, chicken, zebra fish, bullfrog, fluke and amoeba. Each functional P2X receptor is a trimer, with the three protein subunits arranged around an ion-permeable channel pore. To date, seven separate genes coding for P2X subunits have been identified, and referred to as P2X1 through P2X7. The subunits all share a common topology, possessing two plasma membrane spanning domains, a large extracellular loop and intracellular carboxyl and amino termini. ATP binds to the extracellular loop of the P2X receptor, whereupon it evokes a conformational change in the structure of the ion channel that results in the opening of the ion-permeable pore. This allows cations such as Na⁺ and Ca²⁺ to enter the cell, leading to depolarization of the cell membrane and the activation of various Ca²⁺ - sensitive intracellular processes. The channel opening time is dependent upon the subunit makeup of the receptor. Three ATP molecules are thought to be required to activate a P2X receptor, suggesting that ATP needs to bind to each of the three subunits in order to open the channel pore, though recent evidence suggests that ATP binds at the three subunit interfaces.

The P2X7 receptor, a member of the P2X family, has received considerable attention as a therapeutic target due to its role in inflammatory and neuropathic pain mechanisms. It is expressed in both the central nervous system (microglia and astrocytes) and the periphery (macrophages, human epidermal langerhans's cells and a number of tumor cell lines). On glial cells, the P2X7 receptor regulates the release of glutamate which plays a role in

neurotransmission of pain signals. In the immune system, P2X7 activation by extracellular ATP causes ion flux and release of pro-inflammatory cytokines (IL-1beta).

Previously, the researchers in Abbott Laboratories have discovered several cyanoguanidine-type compounds as potent P2X7 antagonists. Here, they introduced a new structural variant where the cyanoguanidine has been replaced with cyanoamidine. Several of these cyanoamidines are found to be very potent P2X7 antagonists. The syntheses of cyanoamidines (aminal type and non-aminal type) were shown in Scheme 9.

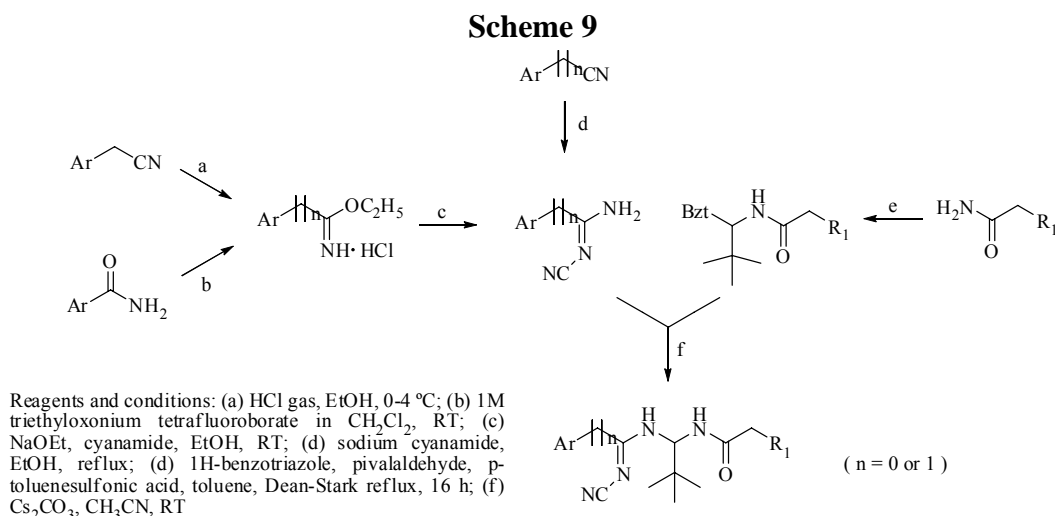
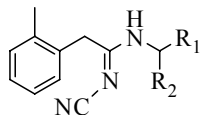


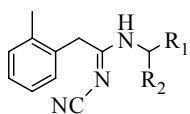
Table 4. Initial SAR of Cyanoamidines Focused on Substituted Phenyls.

Compd #	Ar	n	R _{1a}	^h P ₂ X ₇ IC ₅₀ (nM)	ⁱ P ₂ X ₇ IC ₅₀ (nM)	Compd #	Ar	n	R _{1a}	^h P ₂ X ₇ IC ₅₀ (nM)	ⁱ P ₂ X ₇ IC ₅₀ (nM)
1		0		44	18	11		1		30	19
2		1		12	7	12		1		34	20
3		1		14	5	13		1		105	45
4		1		77	3	14		1		15	5
5		1		19	8	15		1		27	10
6		1		22	4	16		1		27	10
7		1		39	19	17		1		98	29
8		1		50	21	18		0		71	155
9		1		14	5	19		0		715	1396
10		1		38	7						

Table 5. SAR of Cyanoamidines Focused on Substituted Phenyls at R₁.

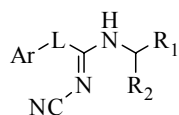
- (1) Only fluoro substituted phenyls are tolerated at R₁.
- (2) Any other larger substitutions on the phenyl ring caused a loss of potency.
- (3) Compounds are generally less potent than aminal-type cyanoamidines.

Compd #	R ₂	R ₁	hP ₂ X ₇ IC ₅₀ (nM)	rP ₂ X ₇ IC ₅₀ (nM)	Compd #	R ₂	R ₁	hP ₂ X ₇ IC ₅₀ (nM)	rP ₂ X ₇ IC ₅₀ (nM)
20	CH ₃		32	165	25	CH ₃		150	374
21	CH ₃		336	57	26	CH ₃		592	314
22	CH ₃		238	40	27	CH ₃		1930	1114
23	CH ₃		245	184	28	CH ₃		935	2209
24	CH ₃		351	111	29	CH ₃		1063	3946

Table 6. SAR of Cyanoamidines Focused Primarily on R₂.

- (1) At R₂ position, R-isomers are more potent than S-isomers.
- (2) Homologated R₂ improved the potency (compound 32) and the potency was retained when the R₂ linked to phenyl moiety (R₁) to form indane and naphthalene ring (compounds 34-36).
- (3) Any other bulkier substitutions at R₂ diminished the potency.

Compd #	R ₂	R ₁	hP ₂ X ₇ IC ₅₀ (nM)	rP ₂ X ₇ IC ₅₀ (nM)	Compd #	R ₂	R ₁	hP ₂ X ₇ IC ₅₀ (nM)	rP ₂ X ₇ IC ₅₀ (nM)
30	CH ₃ (R)		287	103	35			31	98
31	CH ₃ (S)		413	413	36			102	227
32			42	103	37			138	96
33			242	335	38			276	315
34			58	175	39			1760	1509

Table 7. SAR of Cyanoamidines Focusing on Aryl Moiety and Carbon Link Chains.

- (1) Pyridyl and quinolyl replacements for phenyl at aryl position (Ar) retained potency.
- (2) Various other substitutions around the phenyl group (Ar) resulted in loss of potency. 2-methyl phenyl was the optimal.
- (3) Extended chain linkers at R₁ was good only when they were tied with substitution at R₂
- (4) Substituted or extended carbon linkers at L diminished the potency.

Compd #	Ar	L	R ₁	R ₂	hP ₂ X ₇ IC ₅₀ ^a (nM)	rP ₂ X ₇ IC ₅₀ ^b (nM)	Compd #	Ar	L	R ₁	R ₂	hP ₂ X ₇ IC ₅₀ ^a (nM)	rP ₂ X ₇ IC ₅₀ ^b (nM)
40		CH ₂		CH ₃ (R)	297	185	46		CH ₂		CH ₃ (R)	326	457
41		CH ₂		CH ₃	99	18	47		CH ₂			513	449
42		CH ₂		CH ₃ (R)	265	231	48		CH ₂			56	220
43		CH ₂		CH ₃ (R)	1038	1236	49				CH ₃	2429	923
44		CH ₂		CH ₃ (R)	174	168	50				CH ₃	1484	985
45		CH ₂		CH ₃ (R)	2808	828							

“The Discovery of CCR2 Antagonist MK-812”

Lihu Yang, Merck Research Laboratories

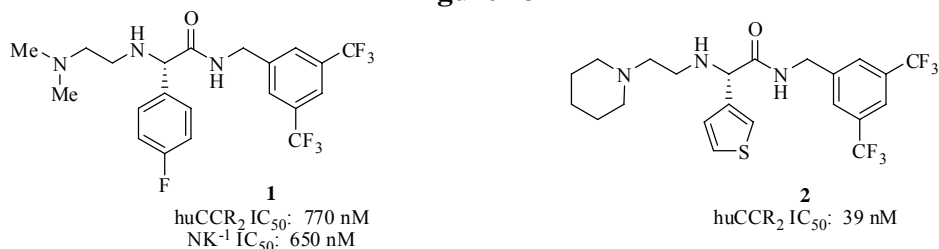
This talk gave an overview of the efforts at Merck Research Laboratories that led to the identification and development of a novel CCR2 antagonist, MK-812. Chemokines are a large family of small structurally related proteins (70–150 amino acids) that are involved in the migration and activation of leukocytes or white blood cells. They are known to activate specific cell-surface seven-transmembrane G-protein coupled receptors (GPCRs). At present, the chemokine/chemokine receptor superfamily contains 20+ receptors and 40+ ligands. Monocyte chemoattractant protein 1 (MCP-1/CCL2) is a member of the chemokine family that binds to the CC chemokine receptor 2 (CCR2), which is most abundantly expressed on monocytes. Prior research has suggested that interruption of the MCP-1/CCR2 interaction may provide potential therapies for a variety of diseases including rheumatoid arthritis, multiple sclerosis and atherosclerosis.

The objective of the program was to obtain a potent, selective, orally bioavailable CCR2 antagonist. This talk outlined the discovery of the potent CCR2 antagonist, MK-812. The Merck team was able to overcome a number of hurdles, including PK, ion channel selectivity, potency through a series of structural restrictions, modulation of amine basicity and water solubility.

An initial screening of the entire Merck sample collection identified a single hit **1** (Figure 13). This compound had been previously prepared for a neurokinin antagonist program. The initial efforts to develop this lead focused on identification of an optimal central amino acid, modification/replacement of the diamine and the effect of amide replacement. The Merck team found that the bis-trifluoromethyl benzyl group was very sensitive to replacement. Likewise, the secondary amide was also critical. Additionally, replacement of the glycine NH resulted in less active compounds. The stereochemistry

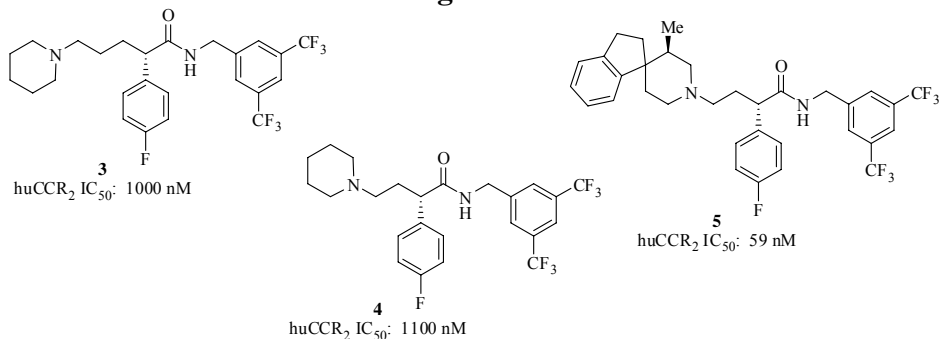
and nature of the amino acid were optimized along with the tertiary amine functionality to provide **2** (Figure 13). Compound **2** was found to be a CCR2-specific antagonist; showing good selectivity against other chemokine and neurokinin receptors. Upon further evaluation in rats, compound **2** was found to have low oral bioavailability ($F = 4.5\%$), high clearance and a modest half life (<2 h). Although optimization of **1** had resulted in good potency enhancement, pharmacokinetic studies in rats revealed rapid clearance as a major issue for this series of compounds.

Figure 13



Subsequent SAR efforts attempted to address this concern by modifying the backbone of **1** by replacing the α -amine with a carbon atom (compound **3**; Figure 14), removing it entirely resulting in a shortening of the amide to amine chain length (compound **4**) and by incorporation of several optimized 4-substituted piperidines from a structurally similar CCR5 antagonist program. None of the modifications led to a compound more potent than **2**; however, the spiroindenylpiperidine analogue **5** approached **2** in terms of potency. In addition to being active in the CCR2 binding assay, it was also active in a functional assay measuring inhibition of MCP-1 induced monocyte chemotaxis ($IC_{50} = 41$ nM). The des-methyl analogue of **5** showed good oral bioavailability in rats; however, these compounds showed binding affinity for the NK1 receptor. These compounds were found to be selective against all other chemokine receptors assayed, except the CCR5 receptor for which it had similar potency. The 3,5-bis(trifluoromethyl) benzyl group was found to be critical for activity.

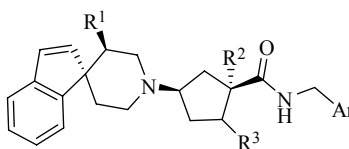
Figure 14



Unfortunately, all of these acyclic leads also lacked activity against the mouse CCR2 receptor. The Merck team felt it would be highly desirable to have murine CCR2 activity for proof of concept studies in various rodent models that would greatly facilitate target validation. Their approach was to introduce structural rigidity into the flexible backbone. For compounds such as **5**, it had been determined that the 4-fluorophenyl group could be

deleted with some retention of activity. Using a systematic and rational approach, a number of constrained scaffolds were synthesized. The 3-amino-1-cyclopentane-carboxamide was identified as the optimal scaffold for CCR2 antagonism. Additionally, this series provided structural novelty and increased potency against human CCR2. As shown in Table 8, the *cis* stereochemistry was optimal and the introduction of a methyl group at R₁ (**6b**) also led to some weak activity at the mouse CCR2 receptor. Introduction of a methyl group at R₂ (**6c**) resulted in modest mCCR2 activity (130 nM) with retention of the single-digit nanomolar hCCR2 activity. Furthermore, introduction of a methyl group at R₃ (**6d**) totally eliminated CCR2 activity. Compound **6c** also showed good selectivity versus the NK1 and NK2 receptors as well as ~500-fold selectivity against the CCR5 receptor. This was a marked improvement over the linear analogues. No other significant chemokine receptor activity was observed. The compound had modest bioavailability (*F* = 15%), high clearance, a large volume of distribution and a long half-life (*t*_{1/2} = 8.1 h).

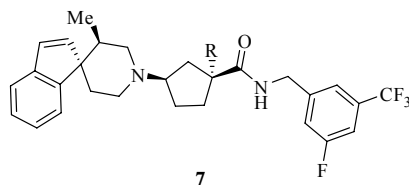
Table 8



6

Cmpd	R ₁	R ₂	R ₃	hCCR2 IC ₅₀ (nM)	mCCR2 IC ₅₀ (nM)
a	H	H	H	7.5	inactive
b	Me	H	H	1.4	27% @ 1 μM
c	Me	Me	H	1.3	130
d	Me	H	Me	>1000	inactive
e	H	Me	H	47	27% @ 1 μM

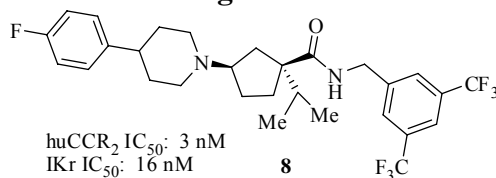
Given the promising results obtained where R₂ = Me (Table 8); additional SAR studies were undertaken to determine the optimal substitution at this position. At the same time, the nature of the benzyl amide was also studied. It was determined that for the spiroindanyl series, the 3-fluoro-5-trifluoromethyl benzyl amide yielded the most active compounds; both in terms of the human and murine CCR2 receptors. Using this optimized amide, a number of different R₂ groups were introduced; with some of the results shown in Table 9. It was found that the most active compounds in the binding assays were also the most active in the functional assays (chemotaxis and calcium flux). The pharmacokinetic properties of several of these compounds were evaluated in rats. Compound **7d** (R = *i*-Pr) exhibited excellent drug levels after both intravenous and oral administration. This compound had a moderate clearance, low volume of distribution and good oral bioavailability. These properties allowed for additional target validation studies requiring a rat model.

Table 9

Cmpd	R	hCCR2 IC ₅₀ (nM)	mCCR2 IC ₅₀ (nM)
a	Me	4	57
b	Et ^a	3.3	3.5
c	<i>n</i> -Pr ^a	5	17
d	<i>i</i> -Pr	3.1	5
e	<i>c</i> -Pr	1.9	8
f	<i>i</i> -Bu	3.9	7.5
g	<i>c</i> -PrCH ₂ ^a	5.5	15
h	MeSCH ₂	3.5	17
i	MeS	67	193

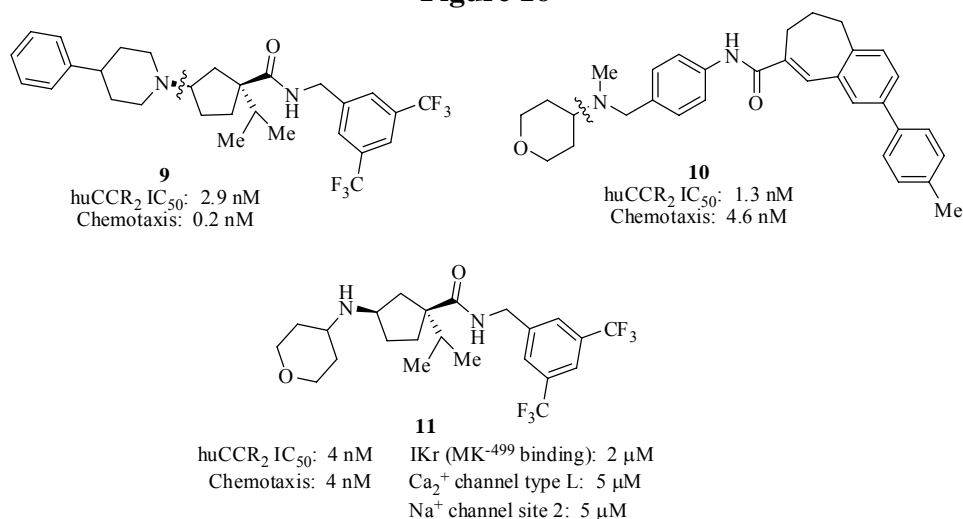
^a Mixture of two 1,3-*cis*-cyclopropane isomers.

Another related series of compounds was also pursued by the Merck team. These contained a series of 4-arylsubstituted piperidines for the left-hand side (LHS) groups. Several of these analogues were very potent; however, counter-screening of a number of compounds within this family revealed a high affinity at the outward delayed rectifier potassium channel (I_{Kr}, human ether-a-go-go-related gene, hERG). This gene has been linked to cardiac arrhythmias for a broad range of drugs. A typical example of a compound from this series is shown in Figure 15. Through the introduction of more polar aryl and heteroaryl groups in the piperidine 4-position, a number of compounds were identified for which there was significant I_{Kr} inhibition. The 5-(pyrimidyl)-piperidine analogue retained most of the potency for **8** (huCCR2 IC₅₀ = 12 nM) and had greater than 100-fold selectivity against the I_{Kr} channel.

Figure 15

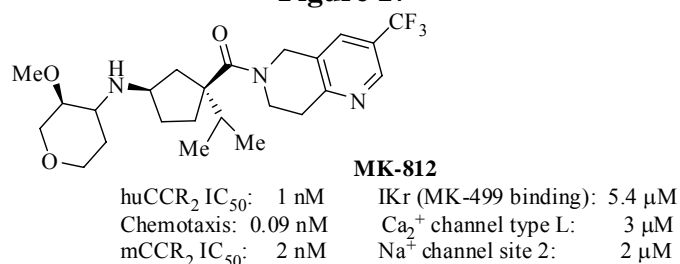
A breakthrough for the Merck team with regard to hERG issues occurred when one of their lead 4-arylpiperidine compounds **9** was hybridized with a Takeda lead compound **10** as shown in Figure 16. Compound **11** was still very potent and exhibited very low off target ion activity.

Figure 16



Further SAR studies led to the discovery of **MK-812** (Figure 17); the most potent CCR2 antagonist in the series. This compound had good mouse PK and was found to inhibit thioglycolate-induced monocyte recruitment in the mouse. Additional studies found that **MK-812** also protected mice from developing experimental allergic encephalomyelitis (EAE), a model of multiple sclerosis. Additional PK studies found this compound to have good to excellent oral bioavailability in the rat, dog and rhesus monkey. Phase I human trials indicated dose proportionate increases in C_{max} and AUC_{0-inf} for doses of 0.2 to 200 mg. The t_{1/2} for this compound is ~11 hours; much better than had been predicted from earlier work and it showed a balanced clearance mechanism. The compound was also found to be generally safe and well-tolerated in Phase II trials. The compound is currently being evaluated in Phase II for multiple sclerosis.

Figure 17



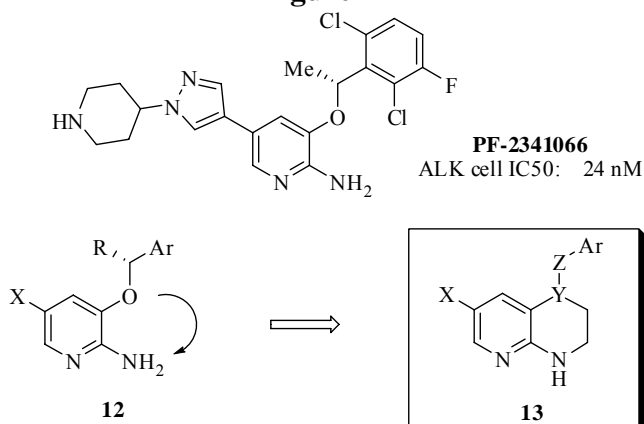
In summary, the Merck team started from micromolar lead and developed a low nanomolar potency, orally bioavailable CCR2 antagonist. They were able to resolve a number of issues, including off-target ion channel activity and demonstrated efficacy in a number of mouse models. Ultimately, this program led to the discovery of a very potent and selective clinical candidate, **MK-812**.

“Pyrido[2,3-*b*]pyrazine and Related Analogs as Constrained Aminopyridine ALK Inhibitors”

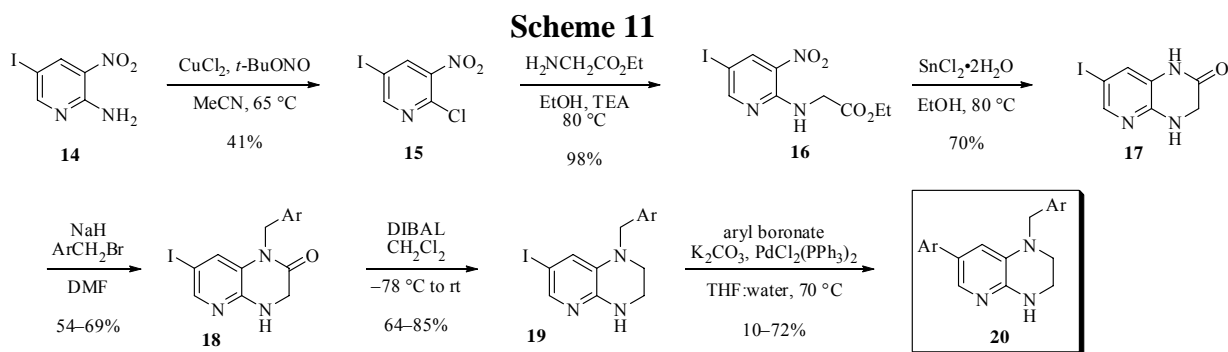
K. L. Milkiewicz; M. S. Albom; T. S. Angeles; R. C. Roemmele; J. P. Theroff; T. L. Underiner; L. R. Weinberg; C. A. Zifcsak; B. D. Dorsey, Cephalon, Inc., Malvern, PA

This presentation outlined the synthesis and activity of a number of constrained aminopyridine ALK (anaplastic lymphoma kinase) inhibitors. ALK is a cell membrane receptor tyrosine kinase (RTK) that has been postulated to be involved in many cancers. It was originally identified due to its involvement in anaplastic large cell lymphoma and expression of full-length ALK has been found in many solid tumors. The ALK gene can be affected by translocations which lead to the expression of oncogenic fusion kinases. The most common of these is NPM-ALK, a fusion of the nucleophosmin (NPM) gene and the intracellular domain of ALK. Small molecule inhibitors of NPM-ALK have exhibited promising *in vivo* biological activity. One novel ALK inhibitor, **PF-2341066** (Figure 17) was recently reported to show cytoreductive antitumor activity.

Figure 17

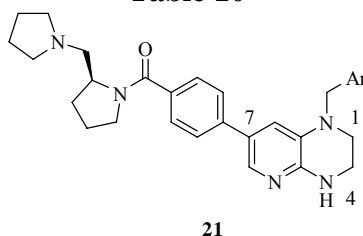


The Cephalon team designed and synthesized a series of constrained aminopyridine analogues (**13**) of **PF-2341066** and outlined their resulting SAR. A number of different ring systems were evaluated including pyridopyrazines (Y = N; Z = CH₂), phenoxynaphthyridines (Y = CH; Z = O) and benzylnaphthyridines (Y = CH; Z = CH₂). The pyridopyrazine analogues were found to be typically 50 to 10,000-fold more active than the corresponding naphthyridine analogues. The synthesis of the pyridopyrazine analogues was accomplished using the synthetic route shown in Scheme 11.



The nature of the benzyl group in **20** proved critical for potency with halogen substitution being required (Table 10). The 2-chloro-3,6-difluorobenzyl benzyl group (**21e**) provided the most potent ALK inhibition for the series. A variety of groups were tolerated at C7 and a number of single digit nanomolar compounds were identified. Methylation of N4 resulted in a dramatic loss of activity. Computational modeling had predicted that the pyrazine N4-H and N5 were necessary for hinge binding as an H-bond donor/acceptor.

Table 10



Cmpd	Ar	ALK IC ₅₀ (nM)
a	Ph	256
b	2,5-di-Cl-Ph	19
c	2,6-di-Cl-Ph	27
d	2,5-di-F-Ph	15
e	2,5-di-F-6-Cl-Ph	3

In summary, the constrained aminopyridine analogues prepared had similar ALK inhibitory activity as compared with the “unconstrained” aminopyridines previously reported (e.g. **PF-2341066**). All compounds prepared maintained selectivity over the related insulin receptor kinase with IC₅₀ values >3000 nM.