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(54) Title: ALPHA-HELIX MIMETIC USING A 2,5-OLIGOPYRIMIDINE SCAFFOLD

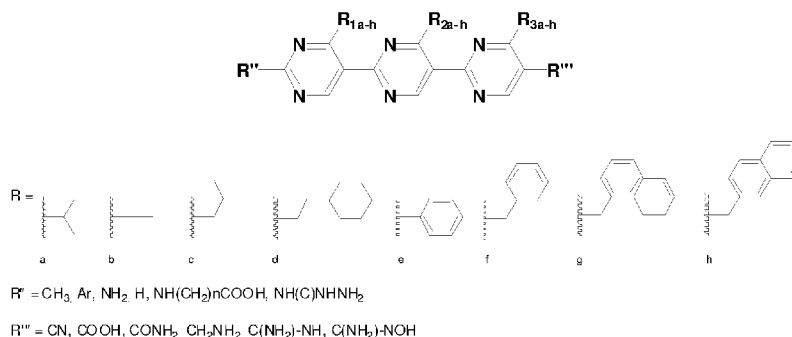


FIG. 1

(57) Abstract: Alpha-helix mimetics and associated methods of making are provided. These compounds are constructed using a 2,5-oligopyrimidine scaffold. The semi-rigid scaffold holds individual side chain-like residues in orientations that mimic the orientations of side chain residues of an α -helical protein domain. The new scaffold is easier to make than previous scaffolds and has much more favorable physical properties than previous alpha-helix mimics. The amphiphilic alpha-helix mimetics have application for making libraries and for treating diseases or conditions effected by the inhibition or disruption of interactions with the alpha helix of a protein.

5 **ALPHA-HELIX MIMETIC USING A 2,5-
OLIGOPYRIMIDINE SCAFFOLD**

CROSS REFERENCE TO RELATED APPLICATIONS

This application claims priority to currently pending U.S. Provisional Patent Application 61/145,357, entitled, "Synthesis of 2,5-Oligopyrimidines with Variable Groups at 4-Position as
10 Alpha-Helix Proteomics", filed January 16, 2009, the contents of which are herein incorporated by reference.

STATEMENT OF GOVERNMENT INTEREST

This invention was made with Government support under Grant No. CA118210 awarded by the National Institutes of Health/NCI. The Government has certain rights in the invention.

15 **FIELD OF INVENTION**

This invention relates to peptide mimetics. More specifically, this invention relates to 2,5-oligopyrimidine scaffolds employable as α -helix mimetics and to compounds, intermediates and methods for the preparation and utilization of the same.

BACKGROUND OF THE INVENTION

20 Protein-protein interactions (PPIs) are involved in several cellular processes and the specific modulation of these interactions will boost our understanding of these processes [Thorsten, B, *Curr. Opin. Drug Discovery Dev.* (2008) 11: 666-674; Saraogi, I, et al., *Biochem. Soc. Trans.* (2008) 36: 1414-1417]. Furthermore, PPI inhibitors can be the basis of important therapeutic interventions [Ockey, DA, et al., *Expert Opin. Ther. Pat.* (2002) 12: 393-400; Arkin, MR, et al.,
25 *Nat. Rev. Drug Discovery* (2004) 3: 301-317; Pagliaro, L, et al., *Curr. Opin. Chem. Biol.* (2004) 8: 442-449; Fletcher, S, et al., *Curr. Top. Med. Chem. (Sharjah, United Arab Emirates)* (2007) 7: 922-927; Wells, JA, et al., *Nature (London)* (2007) 450: 1001-1009; Yin, H, et al., *Chem. Biol.* (2007) 1: 250-269]. PPIs are typically shallow surface interactions over relatively large surface areas to accrue sufficient contact between the protein surfaces to stabilize their
30 specific interaction with each other [Parthasarathi, L, et al., *J. Chem. Inf. Model.* (2008) 48: 1943-1948; Neugebauer, A.; Hartmann, R. W.; Klein, C. D. *J. Med. Chem.* (2007) 50: 4665-4668; Nieddu, E, et al., *Curr. Top. Med. Chem. (Sharjah, United Arab Emirates)* (2007) 7: 21-32; Fletcher, S, et al., *J. R. Soc. Interface* (2006), 3: 215-233]. This makes the development specific PPI inhibitors challenging compared with inhibiting enzymes which typically have
35 relatively small and deep binding pockets.

Several different secondary and tertiary structures can be involved in the PPI. One of the most important is the α -helix. α -Helix mimics that act as sub-micromolar inhibitors of the Bcl-

- 5 xL interaction with the Bak peptide and the MDM2 interaction with the p53 peptide derived from the p53 N-terminus have been reported, as produced by the Hamilton group [Yin, H, et al., *Angew. Chem., Int. Ed.* (2005) 44: 2704-2707; Chen, L, et al., *J. Mol. Cancer Ther.* (2005) 4: 1019-1025]. Additionally, several more polar versions of the 1,4-terphenylene scaffold used to create the α -helix mimics have been reported, but none of them have been more active than the best reported 1,4-terphenylene scaffolds against the same targets in the same in vitro assays [Ernst, JT, et al., *Angew. Chem., Int. Ed.* (2003) 42: 535-539; Yin, H, et al., *J. Am. Chem. Soc.* (2005) 127: 5463-5468; Davis, JM, et al., *Org. Lett.* (2005) 7: 5405-5408; Rodriguez, JM, et al., *Tetrahedron Lett.* (2006) 47: 7443-7446; Saraogi, I, et al., *Angew. Chem., Int. Ed.* (2008) 47: 9691-9694]
- 10
- 15 Hamilton's α -helix mimic approach has been pursued by others, leading to several published examples of more polar versions of the original 1,4-terphenylene scaffold approach. Rebek's synthetic approach is more convergent than the original terphenylene scaffolds, resulting in trimeric heterocyclic scaffolds that are more drug-like than earlier 1,4-terphenylene scaffolds [Moisan, L, et al., *Eur. J. Org. Chem.* (2008) 10: 1673-1676; Moisan, L, et al., *Heterocycles* (2007) 73: 661-671; Volonterio, A, et al., *Bioorg. Med. Chem. Lett.* (2007) 17: 4641-4645]. A tetrameric heterocyclic α -helix mimic scaffold has also been reported. It is designed to position four side chains in the *ith*, *ith*+4, *ith*+7, and *ith*+11 positions of an α -helix [Restorp, P, et al., *Bioorg. Med. Chem. Lett.* (2008) 18: 5909-5911]. Hamilton's group has also recently published a repetitive heterocyclic scaffold approach. [Cummings, CG, et al., *Org. Lett.* (2009) 11: 25-28].
- 20
- 25

Despite the promise of these molecules, some important shortcomings exist in their synthesis and application. First, the prior art scaffolds have proven to be too difficult to prepare in sufficient quantities to make them readily amenable to hit to lead focused library design. Second, many of the prior art molecules do not show promising bioactivity and exhibit poor water solubility. Lastly, many of the prior art molecules exhibit poor polarity. The present invention solves these and other important shortcomings in the prior art as will become apparent in the following Summary and Detailed Description.

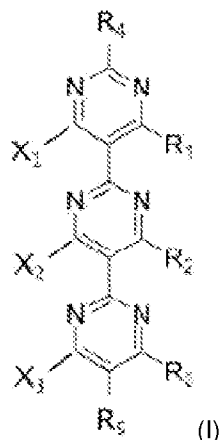
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SUMMARY OF INVENTION

A facile iterative synthesis of a 2,5-terpyrimidinylene library structurally analogous to α -helix mimics prepared using the less drug-like 1,4-terphenylene scaffold is provided. Condensation of amidines with readily prepared α,β -unsaturated α -cyanoketones gives 5-cyano substituted pyrimidines. Iterative transformation of the 5-cyano group into an amidine allows build up of 2,5-terpyrimidinylene libraries with variable groups at the 2-, 4-, 4', 4'', and 5''-positions. These compounds are designed to mimic the *ith*, *ith*+4, and *ith*+7 sites of an α -helix.

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- 5 In a first aspect, the present invention provides an alpha-helix mimetic represented by formula (I):

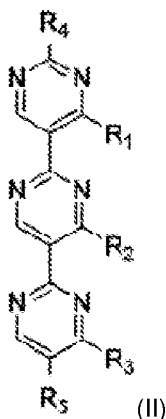


In Formula (I) X_1 , X_2 , and X_3 are independently selected from the group H and NH_2 .

- 10 R_1 , R_2 and R_3 in Formula (I) are independently selected from the group of moieties consisting of $-H$, $-C_1-C_6$ alkyl, $-C_6-C_{12}$ aryl; $-C_7-C_{18}$ alkylaryl, $-C_4-C_{18}$ alkylheterocycle, and $-C_7-C_{18}$ alkylheteroaryl. One $-CH_2-$ of the alkyl may be replaced by $-S-$ and the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from the group consisting of C_1-C_3 alkyl, OH, SH, NH_2 , and COOH.

- 15 R_4 and R_5 are independently selected from the group of moieties consisting of $-H$, $-C_1-C_6$ alkyl, $-C(O)O(C_1-C_6$ alkyl), $-C(O)O(C_6-C_{12}$ aryl), $-C(O)O(C_7-C_{18}$ alkylaryl), $-C(O)O(C_7-C_{18}$ alkylheteroaryl), $-SO_2(C_1-C_6$ alkyl), $-SO_2(C_6-C_{12}$ aryl), $-SO_2(C_7-C_{18}$ alkylaryl), $-SO_2(C_7-C_{18}$ alkylheteroaryl), $-C(O)NH(C_1-C_6$ alkyl), $-C(O)NH(C_6-C_{12}$ aryl); $-C(O)NH(C_7-C_{18}$ alkylaryl), and $(C_7-C_{18}$ alkylheteroaryl). One of the $-CH_2-$ of the alkyl may be replaced by $-S-$, and wherein the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from the group consisting of C_1-C_3 alkyl, OH, SH, NH_2 , and COOH.
- 20

In a second aspect, the present invention provides an alpha-helix mimetic represented by formula (II):



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In Formula (II) R_1 , R_2 and R_3 are independently selected from the group of moieties consisting of $-H$, $-C_1-C_6$ alkyl, $-C_6-C_{12}$ aryl; $-C_7-C_{18}$ alkylaryl, $-C_4-C_{18}$ alkylheterocycle, and $-C_7-C_{18}$ alkylheteroaryl. One $-CH_2-$ of the alkyl may be replaced by $-S-$ and the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from the group consisting of C_1-C_3 alkyl, OH, SH, NH_2 , and COOH.

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R_4 and R_5 are independently selected from the group of moieties consisting of $-H$, $-C_1-C_6$ alkyl, $-C(O)O(C_1-C_6$ alkyl), $-C(O)O(C_6-C_{12}$ aryl), $-C(O)O(C_7-C_{18}$ alkylaryl), $-C(O)O(C_7-C_{18}$ alkylheteroaryl), $-SO_2(C_1-C_6$ alkyl), $-SO_2(C_6-C_{12}$ aryl), $-SO_2(C_7-C_{18}$ alkylaryl), $-SO_2(C_7-C_{18}$ alkylheteroaryl), $-C(O)NH(C_1-C_6$ alkyl), $-C(O)NH(C_6-C_{12}$ aryl); $-C(O)NH(C_7-C_{18}$ alkylaryl), and $(C_7-C_{18}$ alkylheteroaryl). One of the $-CH_2-$ of the alkyl may be replaced by $-S-$, and wherein the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from the group consisting of C_1-C_3 alkyl, OH, SH, NH_2 , and COOH.

15

A plurality of formula (II) subunits can be linked together to form a peptidomimetic or proteinmimetic by covalently coupling an R_4 moiety to an R_5 moiety of an adjacent subunit. In certain embodiments of the second aspect at least one of R^1 , R^2 and R^3 are not H. In further embodiments each of R^1 , R^2 and R^3 independently form an amino acid residue selected from the group consisting of alanine, arginine, asparagine, aspartic acid, cysteine, glutamine, glutamic acid, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine and valine. In still further embodiments each of R^1 , R^2 and R^3 is a moiety individually selected from the group consisting of H, CH_3 , NH_2 , COOH, Ph, *i*-Bu, *t*-Bu, CN, Bn, methyl(1-naphthyl), methyl(2-naphthyl), and *i*-Pr.

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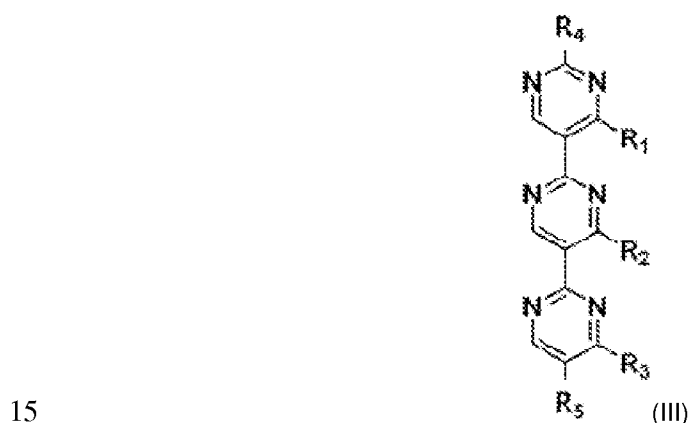
According to the second aspect there is further provided a method of treating a condition or disease state in a patient where the condition or disease state being modulated through the interaction of an α -helical first protein with a binding site of a second protein. The method includes the step of administering to a patient in need of therapy an effective amount of one

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5 or more compounds according to formula (II), optionally in a pharmaceutically acceptable carrier, additive or excipient.

Also according to the second aspect there is provided a method of inhibiting or disrupting the interactions between an alpha helix of a first protein and an alpha helix binding pocket of a second protein. The method includes the step of contacting the first protein and the second
 10 protein with a compound according to formula (II) under conditions wherein the interactions between the alpha helix of the first protein and the alpha helix binding pocket of the second protein are inhibited or disrupted.

In a third aspect the present invention provides an alpha-helix mimetic represented by formula (III):



In Formula (III) R₁, R₂ and R₃ are independently selected from the group of moieties consisting of -H, -C₁-C₆ alkyl, -C₆-C₁₂ aryl; -C₇-C₁₈ alkylaryl, -C₄-C₁₈ alkylheterocycle, and -C₇-C₁₈ alkylheteroaryl. One -CH₂- of the alkyl may be replaced by -S- and the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from the group consisting of
 20 C₁-C₃ alkyl, OH, SH, NH₂, and COOH.

R₄ and R₅ are independently selected from the group of moieties consisting of -H, -C₁-C₆ alkyl, -C(O)O(C₁-C₆ alkyl), -C(O)O(C₆-C₁₂ aryl), -C(O)O(C₇-C₁₈ alkylaryl), -C(O)O(C₇-C₁₈ alkylheteroaryl), -SO₂(C₁-C₆ alkyl), -SO₂(C₆-C₁₂ aryl), -SO₂(C₇-C₁₈ alkylaryl), -SO₂(C₇-C₁₈ alkylheteroaryl), -C(O)NH(C₁-C₆ alkyl), -C(O)NH(C₆-C₁₂ aryl); -C(O)NH(C₇-C₁₈ alkylaryl), and
 25 (C₇-C₁₈ alkylheteroaryl). One of the -CH₂- of the alkyl may be replaced by -S-, and wherein the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from the group consisting of C₁-C₃ alkyl, OH, SH, NH₂, and COOH.

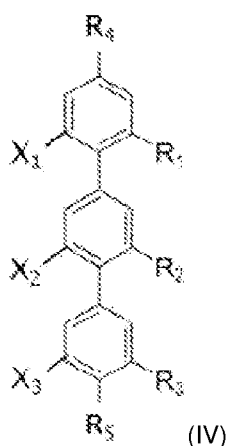
A plurality of formula (III) subunits can be linked together to form a peptidomimetic or proteomimetic by covalently coupling an R₄ moiety to an R₅ moiety of an adjacent subunit. In
 30 certain embodiments of the second aspect at least one of R¹, R² and R³ are not H. In further

- 5 embodiments each of R^1 , R^2 and R^3 independently form an amino acid residue selected from the group consisting of alanine, arginine, asparagine, aspartic acid, cysteine, glutamine, glutamic acid, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine and valine. In still further embodiments each of R^1 , R^2 and R^3 is a moiety individually selected from the group consisting of H, CH_3 , NH_2 ,
 10 $COOH$, Ph, *i*-Bu, *t*-Bu, CN, Bn, methyl(1-naphthyl), methyl(2-naphthyl), and *i*-Pr.

According to the third aspect, there is further provided a method of treating a condition or disease state in a patient where the condition or disease state being modulated through the interaction of an α -helical first protein with a binding site of a second protein. The method includes the step of administering to a patient in need of therapy an effective amount of one
 15 or more compounds according to formula (III), optionally in a pharmaceutically acceptable carrier, additive or excipient.

Also according to the third aspect, there is provided a method of inhibiting or disrupting the interactions between an alpha helix of a first protein and an alpha helix binding pocket of a second protein. The method includes the step of contacting the first protein and the second
 20 protein with a compound according to formula (III) under conditions wherein the interactions between the alpha helix of the first protein and the alpha helix binding pocket of the second protein are inhibited or disrupted.

In a fourth aspect, the present invention provides an alpha-helix mimetic represented by formula (IV):



25

In Formula (IV) X_1 , X_2 , and X_3 are independently selected from the group H and NH_2 .

R_1 , R_2 and R_3 in Formula (IV) are independently selected from the group of moieties consisting of -H, $-C_1-C_6$ alkyl, $-C_6-C_{12}$ aryl; $-C_7-C_{18}$ alkylaryl, $-C_4-C_{18}$ alkylheterocycle, and $-C_7-C_{18}$ alkylheteroaryl. One $-CH_2-$ of the alkyl may be replaced by $-S-$ and the alkyl, aryl, or

- 5 heteroaryl may be substituted with one or two moieties selected from the group consisting of C₁-C₃ alkyl, OH, SH, NH₂, and COOH.

R₄ and R₅ are independently selected from the group of moieties consisting of -H, -C₁-C₆ alkyl, -C(O)O(C₁-C₆ alkyl), -C(O)O(C₆-C₁₂ aryl), -C(O)O(C₇-C₁₈ alkylaryl), -C(O)O(C₇-C₁₈ alkylheteroaryl), -SO₂(C₁-C₆ alkyl), -SO₂(C₆-C₁₂ aryl), -SO₂(C₇-C₁₈ alkylaryl), -SO₂(C₇-C₁₈ alkylheteroaryl), -C(O)NH(C₁-C₆ alkyl), -C(O)NH(C₆-C₁₂ aryl); -C(O)NH(C₇-C₁₈ alkylaryl), and (C₇-C₁₈ alkylheteroaryl). One of the -CH₂- of the alkyl may be replaced by -S-, and wherein the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from the group consisting of C₁-C₃ alkyl, OH, SH, NH₂, and COOH.

BRIEF DESCRIPTION OF THE DRAWINGS

- 15 For a fuller understanding of the invention, reference should be made to the following detailed description, taken in connection with the accompanying drawings, in which:

FIG. 1 is an illustration of a target α -helical mimic

FIG. 2 is an illustration of the general experimental procedure for the synthesis of 2a-h.

FIG. 3 is an illustration of the general experimental procedure for the synthesis of 3a-h.

- 20 FIG. 4 is an illustration of the general experimental procedure for the synthesis of 5a-h.

FIG. 5 is an illustration of the general experimental procedure for the synthesis of amidoxime 6a-h.

FIG. 6 is an illustration of the general experimental procedure for the synthesis of amidine 7a-h.

- 25 FIG. 7 is an illustration of the general experimental procedure for the synthesis of dimer 8(R_{1a-h}, R_{2a-h}).

FIG. 8 is an illustration of the general experimental procedure for the synthesis of trimer 9(R_{1a-h}, R_{2a-h}, R_{3a-h}).

- 30 FIG. 9 is an illustration of a trimeric 2,5-pyrimidine scaffold with structurally diverse 4-R, 4'-R', and 4''-R'' groups that is designed to structurally mimic the *ith*, *ith*+3 or *ith*+4, and *ith*+7 positions of an α -helix. Example 8, below, will prepare the scaffold with the 6, 6', and 6'' substituents equal to H. Example 9, below, will prepare the scaffold with the 6, 6', and 6'' substituents equal to NH₂.

- 35 FIG. 10 is an illustration of a comparison of an Example 8 and Example 9 library member with similar molecular weights, but with different numbers of rotatable bonds. Example 8 and Example 9 library members with the same side chains will have the same number of freely rotatable bonds, because the NH₂ group is not freely rotatable.

5 FIG. 11 is an illustration of an ORTEP diagram of the 2-(4-benzyl-2-phenyl-pyrimidinyl)-5-cyano-4-methyl(1-naphthyl)pyrimidine crystal structure. The conformer shown in darker shading had 70% occupancy and the other minor conformer is shown in outline form. The dynamic nature of this crystal structure documents the very low barrier to rotation about the single bonds between 2,5-pyrimidine rings. The simple NMR spectra that we have
10 consistently seen for many different examples of dimeric and trimeric 2,5-pyrimidines reported above also indicates that no hindered rotation about pyrimidine-pyrimidine single bonds is occurring at least at room temperature in deuteriochloroform or deuterated DMSO, otherwise we would expect to see more complex NMR spectra from atropoisomerism effects.

FIG. 12 is an illustration of the chemical structure of 4-(2-methylpropyl)-2-(2'-methyl(3-indolyl)-4'-(2-methylphenyl)-4''-(4'''-phenylcarboxylate)-3'',5''-pyrimidinyl)-3',5'-pyrimidinyl)pyrimidine-5-carboxylate, as is shown in part A. Parts B and C are the poses of molecule A and the N-terminal domain of p53 docked to MDM2, respectively.

FIG. 13 is an illustration of the chemical structure of 4-(methyl(hydroxyl)-2-(2'-(3-methylpropyl)-4'-(2-methyl(4-hydroxyphenyl)-4''-(4'''-phenyl)-3'',5''-pyrimidinyl)-3',5'-pyrimidinyl)pyrimidine-5-carboxylate, as is shown in part A. Parts B and C are the poses of molecule D and molecule D and ABT-737 superimposed and docked to Bcl-xL, respectively. ABT-737 is a sub-nanomolar antagonist of Bcl-2 family proteins with pro-apoptotic proteins (Oltersdorf, 2005).

FIG. 14 is an illustration of representative trimeric 2,5-pyrimidines. QikProp v3.0 (rel213)
25 calculations were done on the representative trimeric 2,5-pyrimidines shown above and compared with the structurally analogous Hamilton terphenylene scaffold. The log $P_{\text{octanol/water}}$ values differed significantly when the terphenylene portion of the Hamilton scaffold was replaced with the trimeric 2,5-pyrimidine scaffolds with all of the terminating groups and side chains the same. The Hamilton scaffold with a log $P_{\text{octanol/water}} = 10.2$ is significantly outside
30 the range of the training data set of drugs, log $P_{\text{octanol/water}} = (-2.0/6.5)$. The Example 8-type scaffold is barely outside the range at log $P_{\text{octanol/water}} = 6.8$ and the log $P_{\text{octanol/water}} = 5.2$ for the Example 9-type scaffold is well within the range of the drugs in the training set.

FIG. 15 is an illustration providing a tube representation of an idealized α -helix of octa-alanine (with transparent ribbon) with the *ith*, *ith*+4, and *ith*+7 methyl groups highlighted as spheres
35 and an overlay with a 4,4',4''-trimethyl-2,5-terpyrimidinylene with its methyl groups highlighted as spheres. The RMSD for the highlighted methyl groups is 0.68 Å. For clarity, only polar hydrogens are shown. The details of these calculations can be found in the supporting detailed discussion.

- 5 FIG. 16 is an illustration of 1,4-terphenylene and 2,5-terpyrimidinylene scaffolds. QikProp calculations for the most active terphenyl-based Bcl-xL-Bak inhibitor and the analogous terpyrimidine-based analog shows improved drug-like physical characteristics.

FIG. 17 is an ORTEP diagram of compound 6bac.3.

DETAILED DESCRIPTION OF THE PREFERRED EMBODIMENT

- 10 The α -helix is the most common secondary structure found in proteins. Many protein-protein interactions involve recognition of three or more side chains along a single face of an α -helix at the protein-protein interaction surface [Wells, JA, et al., *Nature (London)* (2007) 450: 1001-1009]. The present invention provides for the design and synthesis of semi-rigid scaffolds that hold individual side chain-like residues in orientations that mimic the orientations of i th, i th + 4, i th + 7, and i th + 11, i th + 14 and so on side chain residues of an α -helical protein domain. The semi-rigid scaffolds are based on 2,5-oligo-pyrimidine scaffolds (FIG. 1) and are designed to have drug-like physical properties that enhance the potential therapeutic applications of the resulting protein-protein interaction inhibitor.

- While there are several possible protein-protein interactions that can be inhibited using this new α -helical mimetic scaffold approach, the current studies focus on disrupting the protein-protein interactions between the MDM family proteins with the N-terminal helical domain of the tumor suppressor p53 and the Bcl family of proteins with the helical BH3 only pro-apoptotic proteins. Numerous specific targets are provided herein as specific representative examples of molecules for targeting, but it is contemplated that many other protein-protein interactions can be inhibited using this new technology.

- Programmed cell death, or apoptosis, is a regulated process that contributes to the elimination of defective or unnecessary cells. Apoptosis occurs when a cell has fulfilled its biological function. Many diseases, such as autoimmunity, cancer, inflammatory, and neurodegenerative disorders are the result of dysregulation of this process [Afford, S., et al., *J. Clin. Pathol: Mol. Pathol.*, (2000) 53(2), 55-63]. Proteins in the B-cell lymphoma-2 (Bcl-2) and the murine double minute (MDM2) families play a significant role in the regulation of the apoptotic process. Within the Bcl-2 family, Bcl-2 and Bcl-x_L are some of the proteins that can inhibit apoptosis, whereas Bax and Bak can promote it. NMR studies have revealed that the BH3 domain of the pro-apoptotic protein Bak is required for activity; this region takes an amphipathic α -helical conformation when it binds to Bcl-x_L. The Bak peptide interacts via hydrophobic side chains projecting into the hydrophobic cleft of the Bcl-x_L protein. The overall balance of pro- and anti-apoptotic protein interactions controls the susceptibility of a cell towards programmed cell death. Bax and Bak proteins may form aggregates within the mitochondrial outer membrane, through homooligomerization; this process forms pores in the mitochondrial membrane and activates the apoptotic pathway by releasing, among other

5 proteins, cytochrome *c*. Bcl-2 and Bcl-x_L form heterodimers with the death-promoting region, the BH3 domain, of Bak and Bax neutralizing their activity; apoptosis is then prevented since the release of cytochrome *c* is inhibited [Wendt, M. D., et al., *J. Med. Chem.*, (2006) 49(3), 1165-1181].

The MDM2 protein family is a major regulator of the tumor suppressor protein 53 (p53). The amino terminal of p53 can bind to the hydrophobic cleft of the MDM2 inhibiting the degradation of p53 leaving the wild type p53 free to be phosphorylated and activated for apoptosis induction. The crystal structure of MDM2 in complex with the amino terminal peptide of p53 suggests that the binding is a pocket-ligand type interaction in which the p53 peptide forms an amphipathic α -helix and interacts with a hydrophobic groove on the globular MDM2 domain [Kussie, P., et al., *Science* (1996) 274, 948-53; Popowicz, G., et al., *Cell Cycle*, (2008) 7(15), 2441-2443].

MDMX is a p53-binding protein with strong sequence homology to MDM2. Similar to MDM2, MDMX can bind to the p53 transactivation domain and suppress activation of p53 target genes. Unlike MDM2, MDMX is not transcriptionally induced by p53, does not have intrinsic E3 ligase activity, and does not promote p53 degradation [Shvarts, A., et al., *Embo J* (1996) 15, 5349-57; Stad, R., et al., *EMBO Rep* 2, (2001) 1029-34]. However, MDMX forms a heterodimer with MDM2 through its C terminal RING domain interaction, and stimulates the ability of MDM2 to ubiquitinate and degrade p53 [Sharp, D., et al., *J Biol Chem*, (1999) 274, 38189-96].

Many target proteins show potential for therapeutic approaches. Bcl-2, Bcl-x_L, MDM2, and MDMX are examples of such proteins. Previous studies have demonstrated that overexpression of Bcl-2, Bcl-x_L, MDM2, and MDMX proteins is associated with tumor progression and drug resistance [Strasser, A., et al., *Biochim. Biophys. Acta*, (1997) 1333 (2), F171-F178; Rayburn, E., et al., *Current Cancer Drug Targets*, (2005) 5, 27-41] Therefore, synthesis of compounds designed to mimic the α -helical binding domains of these proteins have great potential for targeted therapeutics. However, the development of such molecules that specifically target the disruption of pro- and anti-apoptotic protein interactions has been a challenging task. During the past few years, some groups have designed compounds that can mimic the structure and recognition properties of α -helices. For example, small peptides have been developed that function as α -helical mimics by means of protein grafting [Chin, J., et al., *Angew. Chem. Int. Ed.*, (2001) 40(20), 3806-3809]. Walensky *et al.* reported the development of hydrocarbon-stapled peptides that mimic the BH3 domain of BID, another pro-apoptotic protein of the Bcl-2 family [Walensky, L., et al., *Science*, (2004) 305, 1466-1470]. Also reported was a novel approach using a semi-rigid non-peptidic scaffold based on substituted *p*-terphenyls [Yin, H., et al., *J. Am. Chem. Soc.*, (2005) 127(29), 5463-5468] and a

5 more hydrophilic terephthalamide scaffold [Yin, H., et al., *Bioorg. Med. Chem. Lett.*, (2004) 14(6), 1375-1379]. Some of these derivatives have exhibited binding affinities in the micromolar range, *in vitro*. For example, the terphenyl- based scaffold has a $K_i = 0.114 \mu\text{M}$ and the terephthalamide scaffold has a $K_i = 0.78 \mu\text{M}$.

Protein-protein interaction inhibitors: Protein-protein interactions are involved in most of the known signal transduction pathways [Berg, T, et al., *Curr. Opin. Drug Discovery Dev.*, (2008) 11(5), 666-674; Saraogi, I, et al., *Biochem. Soc. Trans.*, (2008) 36(6), 1414-1417]. Protein-protein interactions are very important, potentially druggable targets that can in some cases be modulated effectively by typical small molecule inhibitors [Ockey, DA, et al., *Expert Opinion on Therapeutic Patents*, (2002) 12(3), 393-400; Arkin, M. R, et al., *Nat. Rev. Drug Discovery* (2004) 3: 301-317; Pagliaro, L, et al., *Curr. Opin. Chem. Biol.* (2004) 8: 442-449; Fletcher, S.; Hamilton, A. D. *Curr. Top. Med. Chem. (Sharjah, United Arab Emirates)* (2007) 7: 922-927; Wells, JA, et al., *Nature*, (2007) 450(7172), 1001-1009; Yin, H, et al., *Chem. Biol.* (2007) 1: 250-269]. Protein-protein interaction surfaces are typically shallow surface interactions that occur over relatively large surface areas to accrue sufficient interactions between the protein surfaces to stabilize their specific interaction with each other [Parthasarathi, L, et al., *J. Chem. Inf. Model.* (2008) 48: 1943-1948; Neugebauer, A, et al., *J. Med. Chem.* (2007) 50: 4665-4668; Nieddu, E.; Pasa, S. *Curr. Top. Med. Chem. (Sharjah, United Arab Emirates)* (2007) 7: 21-32; Fletcher, S.; Hamilton, A. D. *J. R. Soc. Interface* (2006), 3: 215-233]. Hot spots in protein-protein interacting surfaces are found that if inhibited by relatively small molecules will inhibit the protein-protein interaction [Fletcher, S.; Hamilton, A. D. *Curr. Top. Med. Chem. (Sharjah, United Arab Emirates)* (2007) 7: 922-927; Fry, DC, et al., *Biopolymers*, (2006) 84(6), 535-552], although there are probably some examples where only molecules with physical dimensions that can only be found in higher molecular weight compounds are needed to inhibit protein-protein interactions [Nieddu, E.; Pasa, S. *Curr. Top. Med. Chem. (Sharjah, United Arab Emirates)* (2007) 7: 21-32]. Molecules that follow the rule of five in terms of their molecular weight can often modulate the activity of an enzyme because nature has designed a deep binding cleft to orient and isolate its substrates, but there are enzyme-binding pockets that form very extensive interactions. Perhaps the best-studied example is HIV protease (ritonavir, MW=720.946 Da; indinavir, MW=613.79 Da); this protease is successfully modulated only by drugs with MW>500 [Tsantirizos, YS, et al., *Acc Chem Res.*, (2008) 41(10), 1252-1263; Rana, KZ, et al., *Pharmacotherapy*, (1999) 19(1), 35-59; Ren, S, et al., *Prog Drug Res.*, (1998) 51, 1-31; Vieth, M, et al., *J Med Chem.*, (2004) 47(1), 224-232]. In addition, there are more and more examples of drug candidates that are being developed that completely change the rules of the game of drug design beyond those developed as HIV protease inhibitors. For instance, therapeutic antibodies are among the fastest growing class of drugs in terms of new molecular entities and have enormous

5 molecular weights, for example, adalimumab, HumiraTM, MW=144,190.3 Da, bioavailability 64% [Reichert, JM, et al., *Current Pharmaceutical Biotechnology*, (2008) 9(6), 423-430; Stiehm, E, et al., *Biologicals*, (2008) 36(6), 363-374]. Therapeutic peptides such as enfuvirtide (FuzeonTM, MW=4492.1 Da, bioavailability 84.3% SC) [Este, JA, et al., *Lancet*, (2007) 370(9581), 81-88; Kitchen, CM, et al., *Therapeutics and Clinical Risk Management*, (2008) 4(2), 433-439], cyclosporine (MW=1202.61 Da, bioavailability variable, oral, IV, topical, etc.) [Hess, AD, et al., *Crit Rev Immunol.*, (1986) 6(2), 123-49] and the macrolide antibiotics and semi-synthetic analogues (azithromycin, MW = 748.984 Da, bioavailability 38% for 250 mg capsules) [Kaneko, T, et al., (2006) Macrolide antibiotics., *Comprehensive Medicinal Chemistry II*, Editor(s): Taylor, John B.] are just a few of many other drug classes that violate the rule of five molecular weight guidelines [Ganesan, A, *Curr. Opin. Chem. Biol.*, (2008) 12(3), 306-317; Vistoli, G, et al., *Drug Discovery Today*, (2008) 13(7/8), 285-294; Zhang, M, et al., *Curr. Opin. Biotechnol.*, (2007) 18(6), 478-488; Vieth, M, et al., *J. Med. Chem.*, (2006) 49(12), 3451-3453; Lipinski, CA, *Drug Discovery Today: Technologies*, (2004) 1(4), 337-341]. There are developing paradigms to predict the drug-like character of molecules with much higher than 500 Daltons molecular weight [Veber, DF, et al., *J. Med. Chem.*, (2002) 45(12), 2615-23; Vieth, M, et al., *J. Med. Chem.*, (2006) 49(12), 3451-3453; Vistoli, 2008; Zhang, M, et al., *Curr. Opin. Biotechnol.*, (2007) 18(6), 478-488]. For instance, the number of freely rotatable bonds has a better correlation with drug-like character than molecular weight alone [Veber, DF, et al., *J. Med. Chem.*, (2002) 45(12), 2615-23; Parthasarathi, L, et al., *J. Chem. Inf. Model.* (2008) 48: 1943-1948; Gill, AL, et al., *Topics in Medicinal Chemistry* (Sharjah, United Arab Emirates), (2007) 7(14), 1408-1422]. The average molecular weights of marketed drugs have been consistently increasing over time [Leeson, PD, et al., *J. Med. Chem.*, (2004) 47(25), 6338-6348] and consistently higher molecular weights are found in non-oral vs. oral administration drugs [Vieth, M, et al., *J Med Chem.*, (2004) 47(1), 224-232]. Often molecular weight correlates with the number of freely rotatable bonds, but it does not for semi-rigid systems like the oligo-2,5-pyrimidines proposed herein, because, while the molecular weight grows substantially as additional 2,5-pyrimidine units are added, there is just one additional freely rotatable bond between the 2,5-pyrimidines that is necessarily added for each 2,5-pyrimidine unit added. The side chains on the oligo-2,5-pyrimidine scaffold also add freely rotatable bonds, but that depends on the nature of the side chain. For instance, the isobutyl group of a Leu-like side chain adds less than half the weight as the methyl(3-indolyl) group of a Trp-like side chain, but the same number of freely rotatable bonds (methyl groups freely rotate, but are not considered rotatable because their rotation has a minimal steric effect). The rotatable bonds of two 4, 4', 4"-substituted trimeric 2,5-pyrimidines proposed herein are shown in Figure 10; although they have similar molecular weights, the first has 11 freely rotatable bonds and the second has 9 freely rotatable bonds.

5 α -Helix peptido- and proteomimetics: Terphenylene α -helix mimics [Ernst, JT, et al., *Angew. Chem., Int. Ed.* (2002) 41: 278-281; Kutzki, O, et al., *J. Am. Chem. Soc.* (2002) 124: 11838-11839] provide a useful reference point when considering the trimeric 2,5-pyrimidines proposed herein. Sub-micromolar inhibitors of protein-protein interaction inhibitors including the Bcl-xL interaction with the BH3 only Bak peptide and the MDM2 interaction with the p53
10 peptide derived from the p53 N-terminus have been reported [Yin, H, et al., *J. Am. Chem. Soc.* (2005) 127: 10191-10196; Chen, L, et al., *Mol. Cancer Ther.* (2005) 4: 1019-1025].

We have prepared several monomer units for the repetitive 2,6-piperazinedione and a repetitive 2,5-piperazinedione, some dimers and trimer with these scaffolds. They have greatly improved water solubility compared with Hamilton's terphenylene scaffolds and one
15 showed promising bioactivity, but these 2,6- and 2,5-piperazinedione scaffolds have in general proven to be too difficult to prepare in sufficient quantities to make them readily amenable to hit to lead focused library design, so the synthesis of the trimeric 2,5-pyrimidine scaffold, as taught herein, is provided as an alternative. The 2,5-pyrimidine scaffold can be compared to terphenylene scaffolds with pyrimidine rings instead of phenyl rings, but these
20 changes make the synthesis much easier because the pyrimidine synthetic chemistry is wonderfully convergent. Also, as predicted by QikProp calculations summarized in the results presented below, the trimeric 2,5-pyrimidine scaffold is more polar than the terphenylene scaffold. A key facilitating conversion of this approach is the *ortho*-substituted 5-cyanopyrimidine to 5-carboxamide conversion step. After working out this step, a substantial
25 number of monomeric, dimeric and trimeric 2,5-pyrimidines resulted that are described in detail in the results section below.

Rebek has recognized the importance of Hamilton's α -helix mimetic approach and has published several examples of more polar versions of the original Hamilton terphenylene scaffold approach. Rebek's synthetic approach is a little more convergent than Hamilton's
30 original terphenylene scaffold and his trimeric heterocyclic scaffolds are more drug-like than Hamilton's terphenylene scaffold [Moisan, L, et al., *Eur. J. Org. Chem.* (2008) 10: 1673-1676; Moisan, L, et al. *Heterocycles* (2007) 73: 661-671; Volonterio, A, et al., *Org. Lett.* (2007) 9: 3733-3736; Biro, SM, et al., *Bioorg. Med. Chem. Lett.* (2007) 17: 4641-4645]. The development of the trimeric 2,5-pyrimidine scaffold, documented in the sections below, very
35 strongly supports a synthetic approach that is much easier and more adaptable to hit to lead development than prior synthetic approaches. The compounds taught herein are squarely focused on the synthesis of new structural entities with a strong likelihood of bioactivity based on studies on repetitive heterocyclic scaffolds from prior groups. The molecular modeling studies reported below also strongly support this assertion.

40 Definitions

5 As used throughout the entire application, the terms “a” and “an” are used in the sense that they mean “at least one”, “at least a first”, “one or more” or “a plurality” of the referenced components or steps, unless the context clearly dictates otherwise. For example, the term “a cell” includes a plurality of cells, including mixtures thereof.

The term “and/or” wherever used herein includes the meaning of “and”, “or” and “all or any other combination of the elements connected by said term”.

The term “about” or “approximately” as used herein means within 20%, preferably within 10%, and more preferably within 5% of a given value or range.

Other than in the operating examples, or unless otherwise expressly specified, all of the numerical ranges, amounts, values and percentages such as those for amounts of materials, 15 times and temperatures of reaction, ratios of amounts, values for molecular weight (whether number average molecular weight (“ M_n ”) or weight average molecular weight (“ M_w ”), and others in the following portion of the specification may be read as if prefaced by the word “about” even though the term “about” may not expressly appear with the value, amount or range. Accordingly, unless indicated to the contrary, the numerical parameters set forth in the 20 following specification and attached claims are approximations that may vary depending upon the desired properties sought to be obtained by the present disclosure. At the very least, and not as an attempt to limit the application of the doctrine of equivalents to the scope of the claims, each numerical parameter should at least be construed in light of the number of reported significant digits and by applying ordinary rounding techniques.

25 Notwithstanding that the numerical ranges and parameters setting forth the broad scope of the disclosure are approximations, the numerical values set forth in the specific examples are reported as precisely as possible. Any numerical value, however, inherently contain certain errors necessarily resulting from the standard deviation found in their respective testing measurements. Furthermore, when numerical ranges of varying scope are set forth herein, it 30 is contemplated that any combination of these values inclusive of the recited values may be used.

As used herein, the term “comprising” is intended to mean that the products, compositions and methods include the referenced components or steps, but not excluding others. “Consisting essentially of” when used to define products, compositions and methods, shall 35 mean excluding other components or steps of any essential significance. Thus, a composition consisting essentially of the recited components would not exclude trace contaminants and pharmaceutically acceptable carriers. “Consisting of” shall mean excluding more than trace elements of other components or steps.

5 Alkyl means a straight-chain or branched-chain aliphatic hydrocarbon moiety having 1 to 12 or more preferably 1 to 6 carbon atoms. Suitable alkyl groups include, but are not limited to, the linear alkyl moieties methyl, ethyl, propyl, isopropyl, butyl, sec-butyl, tert-butyl, pentyl, and hexyl. Other examples of suitable alkyl groups include, but are not limited to, the substituted
10 linear alkyl moieties 1-, 2- or 3-methylbutyl, 1,1-, 1,2- or 2,2-dimethylpropyl, 1-ethylpropyl, 1-, 2-, 3- or 4-methylpentyl, 1,1-, 1,2-, 1,3-, 2,2-, 2,3- or 3,3-dimethylbutyl, 1- or 2-ethylbutyl, ethylmethylpropyl, trimethylpropyl, and the like. In one embodiment, the alkyl may be a cyclic alkyl.

Aryl refers to an aromatic carbocyclic moiety containing 6 to 14 carbon atoms, preferably 6 to 12 carbon atoms, especially 6 to 10 carbon atoms. Exemplary aryls are phenyl and naphthyl.

15 Heteroaryl refers to an aromatic carbocyclic moiety containing 3 to 12 carbon atoms, preferably 3 to 8 carbon atoms, and at least one heteroatom selected from O, N, and S. The heteroaryl may be monocyclic or bicyclic. Exemplary heteroaryls moieties include pyridyl, furyl, benzofuranyl, thiophenyl, benzothiophenyl, quinoliny, pyrrolyl, indolyl, oxazolyl, benzoxazolyl, imidazolyl, benzimidazolyl, thiazolyl, benzothiazolyl, isoxazolyl, pyrazolyl,
20 isothiazolyl, pyridazinyl, pyrimidinyl, pyrazinyl, triazinyl, cinnoliny, phthalazinyl, quinazolinyl, and the like. In one embodiment, the heteroaryl is an indolyl, pyrrolyl, or imidazolyl.

Heterocycle refers to a non-aromatic, cyclic moiety containing 3 to 12 carbon atoms, preferably 3 to 8 carbon atoms, and at least one heteroatom selected from O, N, and S. The heterocycle may be monocyclic or bicyclic. Exemplary heterocycles moieties include
25 pyrrolidinyl.

In some embodiments the alkyl, aryl, or heteroaryl may be substituted. Substituted moieties preferably have 1 to 3 substituents, or they may have 1 to 2 substituents which may be the same or different. In one embodiment, a single substituent is present. Preferred substitutions include C₁-C₃ alkyl, —OH, —SH, NH₂, —CO₂, and —C(O)NH.

30 Alkylaryl means an alkyl having at least one alkyl hydrogen atom replaced with an aryl moiety, such as —CH₂-phenyl, —CH₂CH₂CH₂-phenyl, and —CH₂-naphthyl.

Alkylheterocycle means an alkyl having at least one alkyl hydrogen atom replaced with a heterocycle moiety, such as —CH₂-pyrrolidinyl and —CH₂CH₂-pyrrolidinyl.

Alkylheteroaryl means an alkyl having at least one alkyl hydrogen atom replaced with a
35 heteroaryl moiety, such as —CH₂-indolyl, —CH₂CH₂CH₂-indolyl, and —CH₂-pyrimidinyl.

As used herein, disrupting an interaction between a first protein and a second protein refers to the process of perturbing one or more covalent or non-covalent bonding interactions between the first and the second protein. Covalent bonding interactions between proteins include, for example, disulfide bonds, ester bonds, amide bonds and the like. Non-covalent bonding

5 interactions between proteins include, for example, hydrophobic interactions, van der Waals interactions, ionic interactions, hydrogen bonding interactions and the like.

As used herein, inhibiting an interaction between a first protein and a second protein refers to the process of lowering the overall ability of the two proteins to bind or associate.

10 The term "administration" and variants thereof (e.g., "administering" a compound) in reference to a compound of the invention means introducing the compound or a prodrug of the compound into the system of the animal in need of treatment. When a compound of the invention or prodrug thereof is provided in combination with one or more other active agents (e.g., a cytotoxic agent, etc.), "administration" and its variants are each understood to include concurrent and sequential introduction of the compound or prodrug thereof and other agents.

15 As used herein, the term "composition" is intended to encompass a product comprising the specified ingredients in the specified amounts, as well as any product which results, directly or indirectly, from combination of the specified ingredients in the specified amounts.

The term "therapeutically effective amount" as used herein means that amount of active compound or pharmaceutical agent that elicits the biological or medicinal response in a
20 tissue, system, animal or human that is being sought by a researcher, veterinarian, medical doctor or other clinician. In reference to cancers or other unwanted cell proliferation, an effective amount comprises an amount sufficient to cause a tumor to shrink and/or to decrease the growth rate of the tumor (such as to suppress tumor growth) or to prevent or delay other unwanted cell proliferation. In some embodiments, an effective amount is an
25 amount sufficient to delay development. In some embodiments, an effective amount is an amount sufficient to prevent or delay occurrence and/or recurrence. An effective amount can be administered in one or more doses. In the case of cancer, the effective amount of the drug or composition may: (i) reduce the number of cancer cells; (ii) reduce tumor size; (iii) inhibit, retard, slow to some extent and preferably stop cancer cell infiltration into peripheral organs;
30 (iv) inhibit (i.e., slow to some extent and preferably stop) tumor metastasis; (v) inhibit tumor growth; (vi) prevent or delay occurrence and/or recurrence of tumor; and/or (vii) relieve to some extent one or more of the symptoms associated with the cancer.

As used herein, "treatment" refers to obtaining beneficial or desired clinical results. Beneficial or desired clinical results include, but are not limited to, any one or more of: alleviation of one
35 or more symptoms (such as tumor growth or metastasis), diminishment of extent of cancer, stabilized (i.e., not worsening) state of cancer, preventing or delaying spread (e.g., metastasis) of the cancer, preventing or delaying occurrence or recurrence of cancer, delay or slowing of cancer progression, amelioration of the cancer state, and remission (whether partial or total). The methods of the invention contemplate any one or more of these aspects
40 of treatment.

5 A "subject in need of treatment" is a mammal with a condition that is life-threatening or that impairs health or shortens the lifespan of the mammal.

A "pharmaceutically acceptable" component is one that is suitable for use with humans and/or animals without undue adverse side effects (such as toxicity, irritation, and allergic response) commensurate with a reasonable benefit/risk ratio.

10 A "safe and effective amount" refers to the quantity of a component that is sufficient to yield a desired therapeutic response without undue adverse side effects (such as toxicity, irritation, or allergic response) commensurate with a reasonable benefit/risk ratio when used in the manner of this invention.

A "pharmaceutically acceptable carrier" is a carrier, such as a solvent, suspending agent or vehicle, for delivering the compound or compounds in question to the animal or human. The carrier may be liquid or solid and is selected with the planned manner of administration in mind. Liposomes are also a pharmaceutical carrier. As used herein, "carrier" includes any and all solvents, dispersion media, vehicles, coatings, diluents, antibacterial and antifungal agents, isotonic and absorption delaying agents, buffers, carrier solutions, suspensions, 15 colloids, and the like. The use of such media and agents for pharmaceutical active substances is well known in the art. Except insofar as any conventional media or agent is incompatible with the active ingredient, its use in the therapeutic compositions is contemplated.

As used herein, the term "patient" preferably refers to a human in need of treatment with an anti-cancer agent or treatment for any purpose, and more preferably a human in need of such a treatment to treat cancer, or a precancerous condition or lesion. However, the term "patient" 20 can also refer to non-human animals, preferably mammals such as dogs, cats, horses, cows, pigs, sheep and non-human primates, among others, that are in need of treatment with an anti-cancer agent or treatment.

30 Compounds according to the present invention may be used as active agents in pharmaceutical compositions as agonists or inhibitors of α -helical proteins in their interactions with proteins (such as receptors, enzymes, other proteins) or other binding sites, said compositions comprising an effective amount of one or more of the compounds disclosed above, formulated as a pharmaceutical dosage form, optionally in combination with a pharmaceutically acceptable carrier, additive or excipient. Pharmaceutical compositions 35 according to the present invention may be used in the treatment of cancer (as, for example, a suppressor of Mdm2/p53 tumor, to inhibit Bcl protein family/Bak protein family or AP-1 transcription factor/DNA complex), proliferative diseases including, for example, psoriasis, genital warts and hyperproliferative keratinocyte diseases including hyperkeratosis, 40 ichthyosis, keratoderma or lichen planus, neuropeptide Y receptor interactions, including the

- 5 resulting hypertension and neuronal/neurological effects (to facilitate neuromodulation through, for example, inhibition of calmodulin binding on calmodulin dependent phosphodiesterase including PDE1A, PDE1B and PDE1C, among others), neurodegenerative diseases including Alzheimer's disease and Parkinson's disease, Herpes simplex virus infections (HSV, through inhibition of the HSV VP 16/human TAF1131 HSV infection complex), HIV infections (through inhibition of HIVp7 nuclear capsid protein/RNA interaction or alternatively, through inhibition of the REV protein RNA complex), asthma, hypertension, cancer and autoimmune diseases (through immunomodulation, for example, by inhibition or modulation of interleukin/receptor interaction), numerous viral infections other than HIV or HSV through inhibition of ribonucleotide reductase dimerization, or to modulate nuclear receptor/coactivator protein complex interaction (eg. estrogen receptor for anticancer therapy) and to disrupt G protein coupled receptor (GPCR) function (through displacement of one of the helices and disruption of the helix packing interactions or alternatively, by blocking the interaction of the ligand with GPCR, e.g. where the ligand contains a key helix binding domain (e.g. GCSF, calcitonin, interleukins, parathyroid hormones, among others).
- 10
- 15
- 20 Compositions according to the present invention may be coadministered with another active compound such as antimicrobial agents, antineoplastic agents, anti-cancer agents or preservatives. When co-administered with compounds according to the present invention for the treatment of tumors, including cancer, other agents such as antimetabolites, etoposide, doxorubicin, taxol, hydroxyurea, vincristine, cytoxan (cyclophosphamide) or mitomycin C, among numerous others, including topoisomerase I and topoisomerase II inhibitors, such as adriamycin, topotecan, camptothecin and irinotecan, other agent such as gemcitabine and agents based upon camptothecin and cis-platin may be included. These compounds may also be included in pharmaceutical formulations or coadministered with compounds according to the present invention to produce additive or synergistic anti-cancer activity.
- 25
- 30 The individual components of such combinations as described above may be administered either sequentially or simultaneously in separate or combined pharmaceutical formulations. When one or more of the compounds according to the present invention is used in combination with a second therapeutic agent active the dose of each compound may be either the same as or differ from that when the compound is used alone. Appropriate doses will be readily appreciated by those skilled in the art.
- 35

In other aspects of the present invention, certain compounds according to the present invention may be used as agonists or antagonists in binding assays, as analytical agents, as agents to be used to isolate or purify proteins, and as intermediates in the synthesis of further peptidomimetic agents, among other uses.

5 Another aspect of the present invention is directed to compounds according to the present invention which may be used to mimic α -helical proteins in an agonistic or antagonistic manner. In this aspect of the present invention, one or more of the compounds according to the present invention may be used to mimic or inhibit the binding of an α -helical protein for its binding site, whether that binding site is another protein, a receptor (such as a cell surface
10 receptor or a G-protein coupled receptor), signaling proteins, proteins involved in apoptotic pathways (especially neuronal apoptosis), active sites and regulatory domains of enzymes, growth factors, DNA, RNA (including oligonucleotides), viral fusion proteins and viral coat proteins, among numerous others.

The present invention also relates to methods of treating patients in need thereof for
15 conditions or disease states which are modulated through interactions between alpha helical proteins and other proteins or binding sites are other aspects of the invention. Thus, in the method aspect of the present invention, pharmaceutical compositions comprising α -helical protein agonists or antagonists may be used to treat any condition or disease state in which α -helical proteins modulate their activity through a receptor or other binding site. In particular,
20 the method aspect of the present invention relates to the inhibition of protein binding to binding sites within the patient in order to effect a biological/pharmacological result. Compounds according to the present invention may be used as proteomimetics to inhibit the interaction between a native α helical protein (i.e., a natural α helical protein normally found in a patient) and its binding site. Preferred compounds according to the present invention may
25 be used to disrupt or compete with the binding of a number of proteins including, for example, calmodulin (CaM) with binding sites on smooth muscle light chain kinase (smMLCK) or phosphodiesterase (PDE1A, PDE1B, PDE1C) with resulting neuromuscular and neuronal (among other) effects in the treating of disease states or conditions, gp41 (HIV) and other viruses such as HSV or HBV, for the viral invasive binding sites in CD4 and/or other
30 hematopoietic cells, genital/mucosal cells, among others (HSV) and hepatocytes (HBV), among numerous others and pro-apoptotic Bak- and/or Bad-proteins, for their binding interaction with Bcl-x_L protein in a preferred treatment for cancer.

Thus, the present application is directed to the treatment of disease states or conditions which are modulated through interactions between α -helical proteins and other proteins or binding
35 sites of the α -helical proteins preferably selected from the group consisting of viral infections (including Hepatitis B virus (HBV) infections, human immunodeficiency virus (HIV) infections or conditions associated with such infections (AIDS), Herpes Simplex virus infections (HSV) infections, tumors and/or cancer, proliferative diseases including psoriasis, genital warts and hyperproliferative keratinocyte diseases including hyperkeratosis, ichthyosis, keratoderma,
40 lichen planus, hypertension, neuronal disorders by promoting neuromodulation including, for example, attention deficit disorder, memory loss, language and learning disorders, asthma,

5 autoimmune diseases including lupus (lupus erythematosus), multiple sclerosis, arthritis, including rheumatoid arthritis, rheumatic diseases, fibromyalgia, Sjögren's disease and Grave's disease and neurodegenerative diseases including Alzheimer's disease and Parkinson's disease, said method comprising administering to a patient in need thereof an effective amount of a pharmaceutical composition comprising any one or more of the
10 compounds previously described above.

Dosage: A person of ordinary skill in the art can easily determine an appropriate dose of one of the instant compositions to administer to a subject without undue experimentation. Typically, a physician will determine the actual dosage which will be most suitable for an individual patient and it will depend on a variety of factors including the activity of the specific
15 compound employed, the metabolic stability and length of action of that compound, the age, body weight, general health, sex, diet, mode and time of administration, rate of excretion, drug combination, the severity of the particular condition, and the individual undergoing therapy. The dosages disclosed herein are exemplary of the average case. There can of course be individual instances where higher or lower dosage ranges are merited, and such
20 are within the scope of this invention.

In another aspect, the present invention is directed to the use of one or more compounds according to the present invention in a pharmaceutically acceptable carrier, additive or excipient at a suitable dose ranging from about 0.05 to about 100 mg/kg of body weight per day, preferably within the range of about 0.1 to 50 mg/kg/day, most preferably in the range of
25 1 to 20 mg/kg/day. The desired dose may conveniently be presented in a single dose or as divided doses administered at appropriate intervals, for example as two, three, four or more sub-doses per day.

Ideally, the active ingredient should be administered to achieve effective peak plasma concentrations of the active compound within the range of from about 0.05 to about 5 μ M.
30 This may be achieved, for example, by the intravenous injection of about a 0.05 to 10% solution of the active ingredient, optionally in saline, or orally administered as a bolus containing about 1 mg to about 5 g, preferably about 5 mg to about 500 mg of the active ingredient, depending upon the active compound and its intended target. Desirable blood levels may be maintained by a continuous infusion to preferably provide about 0.01 to about
35 2.0 mg/kg/hour or by intermittent infusions containing about 0.05 to about 15 mg/kg of the active ingredient. Oral dosages, where applicable, will depend on the bioavailability of the compounds from the GI tract, as well as the pharmacokinetics of the compounds to be administered. While it is possible that, for use in therapy, a compound of the invention may be administered as the raw chemical, it is preferable to present the active ingredient as a

- 5 pharmaceutical formulation, presented in combination with a pharmaceutically acceptable carrier, excipient or additive.

Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art (e.g., in cell culture, molecular genetics, nucleic acid chemistry, hybridisation techniques and biochemistry).

10 Standard techniques are used for molecular, genetic and biochemical methods. See, generally, Sambrook et al., *Molecular Cloning: A Laboratory Manual*, 2d ed. (1989) Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y. and Ausubel et al., *Short Protocols in Molecular Biology* (1999) 4th Ed, John Wiley & Sons, Inc.; as well as Guthrie et al., *Guide to Yeast Genetics and Molecular Biology*, *Methods in Enzymology*, Vol. 194, Academic Press,

15 Inc., (1991), *PCR Protocols: A Guide to Methods and Applications* (Innis, et al. 1990. Academic Press, San Diego, Calif.), McPherson et al., *PCR Volume 1*, Oxford University Press, (1991), *Culture of Animal Cells: A Manual of Basic Technique*, 2nd Ed. (R. I. Freshney. 1987. Liss, Inc. New York, N.Y.), and *Gene Transfer and Expression Protocols*, pp. 109-128, ed. E. J. Murray, The Humana Press Inc., Clifton, N.J.).

20 EXAMPLES

EXAMPLE – 1 – GENERAL EXPERIMENTAL PROCEDURE FOR THE SYNTHESIS OF 2a-h:

An illustration of the procedure for the synthesis of 2a-h is presented in FIG. 2. A mixture of potassium *t*-pentylate (~1.7 M in toluene, 106 mmol, 62.3 mL) and anhydrous THF (20 mL)

25 was cooled at 0 °C in an ice-bath and under an argon atmosphere. Then anhydrous acetonitrile (106 mmol, 6 mL) and the methyl ester 1f (71 mmol, 10 mL) were added simultaneously to the cold solution. The reaction mixture was allowed to warm to room temperature and stirred under an argon atmosphere for 22 hours. A precipitate was formed within a few minutes (3-5 min.) of reaction and it remained cloudy until completion. The

30 reaction was monitored by TLC and it was stopped when the TLC indicated the consumption of the ester (1f). The mixture was filtered and the filtered cake was washed thoroughly with hexanes (80 mL). The filtered residue was then transferred to a separatory funnel and saturated aqueous KHSO₄ solution (400 mL) was added to acidify to pH= ~2-3. An equal volume amount of DCM (400 mL) was used to extract the desired compound (2a-h). The

35 organic layer was washed with brine (100 mL), dried over Na₂SO₄, and concentrated under reduced pressure to yield compound 2f as a yellow oil in 65% yield. Compound 2f was used without further purification in the next step [Ji, Y., et al., *Org. Lett.*, (2006) 8(6),1161-1163]. FIG. 2 provides a further illustration of the synthesis of 2a-h.

EXAMPLE – 2 – GENERAL EXPERIMENTAL PROCEDURE FOR THE SYNTHESIS OF 3a-h

5 An illustration of the procedure for the synthesis of 3a-h is presented in FIG. 3. Nitrile derivative 2f, as produced in Example 1, above, (0.75 mmol, 120 mg) was dissolved in dry THF (3 mL) under an argon atmosphere. Then *N,N*-dimethylformamide dimethyl acetal (0.98 mmol, 0.16 mL) was added and the mixture was stirred at room temperature. Progress of the reaction was monitored by TLC until complete consumption of starting material 2f was
10 observed (~2 hr). The solvent was evaporated under reduced pressure and the desired compound was obtained as an E/Z isomeric mixture in nearly quantitative yield (> 90%). Compound 3f was isolated as a yellow solid [Reuman, M., et al., *J. Org. Chem.*, (2008) 73(3), 1121-1123]. To the extent that, or if, a precipitate was formed during the reaction, the mixture was cooled to 0 °C and then it was filtered and rinsed with cold THF to isolate the desired
15 compound (3a-h).

EXAMPLE – 3 – GENERAL EXPERIMENTAL PROCEDURE FOR THE SYNTHESIS OF 5a-h

An illustration of the procedure for the synthesis of 5a-h is presented in FIG. 4. To a mixture of compound 3f, as produced in Example 2, above, (1.42 g, 6.61 mmol) and guanidine hydrochloride (2.53g, 26.5 mmol) was added ethanol (absolute, 200 proof, 5mL), and
20 triethylamine (3.7 mL, 26.5 mmol). The mixture was refluxed for 23 hours, then cooled to room temperature, and kept in the freezer for 12 hours. The precipitate was filtered by vacuum filtration, and rinsed with ice cold ethanol (3 mL) three times, to yield compound 5f as yellow crystals in 91 % yield. (Černuchová, P. et al., *Tetrahedron*, (2005) 61(22), 5379-5387]. The reaction also was found to proceed in the absence of triethylamine with similar yields.

25 Microwave-assisted reactions have been done in the presence or absence of base (e.g. Et₃N or NaOEt) to obtain the desired compounds 5a-h. For example, to a mixture of nitrile 3c (0.933 mmol, 200 mg) and guanidine hydrochloride (1.87 mmol, 178.3 mg) in ethanol (5 mL) was added sodium ethoxide (0.933 mmol, 63.49 mg) suspended in ethanol (1 mL). The reaction mixture was placed on a microwave reactor for 20 min. x 2 at 120 °C. A white crystal
30 precipitate formed in the solution upon cooling to room temperature. The precipitate was filtered and rinsed with cold ethanol (3 mL x 2) to isolate compound 5c as white crystals in 80% yield.

EXAMPLE – 4 – GENERAL EXPERIMENTAL PROCEDURE FOR THE SYNTHESIS OF AMIDOXIME 6a-h

35 An illustration of the procedure for the synthesis of amidoxime 6a-h is presented in FIG. 5. Two methods are provided for the synthesis of amidoxime 6a-h; Method A and Method B.

Method A: To a mixture of compound 5c (R" = phenyl, 4.21 mmol, 1 g) and hydroxylamine hydrochloride (10.53 mmol, 0.72 g) in methanol (12 mL) was added triethylamine (10.53 mmol, 1.46 mL). The reaction mixture was stirred under refluxing conditions until the TLC

- 5 indicated the consumption of starting material 5c. The solvent was removed under reduced pressure to obtain a white crude solid. The crude was dissolved in DCM (50 mL), washed with water (40 mL), dried over Na₂SO₄, and concentrated under vacuum to obtain compound **6c** as an isomeric mixture. Compound 6c was isolated as an off-white, fluffy solid in 95% (Nadrah and Dolenc, 2007).
- 10 Method B: To a mixture of compound 5g (R" = methyl, 2.03g, 7.81 mmol), hydroxylamine hydrochloride (3.26g, 46.9 mmol), and potassium carbonate (3.24g, 23.4 mmol) was added ethanol (absolute, 32 mL) and water (20 mL). The mixture was refluxed for 12 hours, and the solution was concentrated under reduced pressure. The residue was extracted between DCM (100 mL) and water (100 mL). The organic layer was then extracted by brine (50 mL), dried
- 15 using anhydrous Na₂SO₄. The solvent was removed under reduced pressure to yield compound 6g as a yellow solid in 99% yield [Judkins, B. D., et al., *Synth. Comm.*, (1996) 26(23), 4351-4367].

EXAMPLE – 5 – GENERAL EXPERIMENTAL PROCEDURE FOR THE SYNTHESIS OF AMIDINE 7a-h

- 20 An illustration of the procedure for the synthesis of amidine 7a-h is presented in FIG. 6. Two methods are provided for the synthesis of amidine 7a-h; Method A and Method B.

Method A: Compound 6c, as produced in Example 5 (Method A), above, (R" = phenyl, 0.92 mmol, 0.25 g) was dissolved in glacial acetic acid (1 mL) and acetic anhydride (1.01 mmol, 95 μ L). After 5 min. of stirring, potassium formate prepared *in situ* from K₂CO₃ (5 mmol, 0.69 g),

25 formic acid (10 mmol, 0.37 mL) in methanol (2.5 mL) was added to the mixture followed by the addition of 10% Pd/C (10 mmol %, 98 mg). The reaction mixture was stirred at room temperature until the TLC indicated the consumption of starting material 6c. The crude was filtered through Celite™ (1.5 g) and rinsed with methanol (3 mL x 3). The filtrate was concentrated under reduced pressure to obtain a yellow crude residue. To this residue, DCM

30 (6 mL) was added and formed white precipitated was removed by vacuum filtration. The filtrate was concentrated under vacuum to obtain the crude acetate salt (7c) as a yellow solid, which was used without further purification in the next step [Nadrah, K., et. al., *Synlett*, (2007) 8, 1257-1258.).

Method B: To a solution of compound 6g, as produced in Example 5 (Method B), above,

35 (0.1656g, 0.57 mmol) in acetic acid (glacial, 10 mL) was added acetic anhydride (0.08 mL, 0.85 mmol). After stirring for 20 min, Pd/C (10 wt%, 0.0606g, 0.057 mmol) was added and the mixture was hydrogenated under H₂ atmosphere (55 psi) at room temperature for 4 hours. The crude was then filtered through Celite™ (1 g) and rinsed with acetic acid (5 mL x 3). The solvent was removed under reduced pressure to obtain the crude acetate salt (7g) as a yellow

- 5 solid, which was used without further purification in the next step [Judkins, B. D., et al., *Synth. Comm.*, (1996) 26(23), 4351-4367].

EXAMPLE – 6 – GENERAL EXPERIMENTAL PROCEDURE FOR THE SYNTHESIS OF DIMER 8(R_{1a-h}, R_{2a-h})

An illustration of the procedure for the synthesis of dimer 8(R_{1a-h}, R_{2a-h}) is presented in FIG. 7.

- 10 To obtain compound 8(R_{1a-h}, R_{2a-h}), steps to form 5a-h to 7a-h, as above, were followed. Dimers were obtained as white, off-white, and yellow solid compounds.

For example, to the crude acetate salt (7c, R"= phenyl) dissolved in ethanol (0.8 mL) was added compound 3f (0.69 mmol, 147.8 mg) and triethylamine (1.38 mmol, 0.19 mL). The reaction mixture was stirred under refluxing conditions for 2 hours and then it was stirred at
15 room temperature for 18 hours. The formed precipitate was filtered and rinsed with cold ethanol (5 mL) to yield dimer 8(R_{1c}, R_f) as a yellow solid in 28% yield after two steps.

EXAMPLE – 7 – GENERAL EXPERIMENTAL PROCEDURE FOR THE SYNTHESIS OF TRIMER 9(R_{1a-h}, R_{2a-h}, R_{3a-h})

- 20 An illustration of the procedure for the synthesis of trimer 9(R_{1a-h}, R_{2a-h}, R_{3a-h}) is presented in FIG. 8.

To obtain compound 9(R_{1a-h}, R_{2a-h}, R_{3a-h}), steps to form 8(R_{1a-h}, R_{2a-h}), presented in Example 6, above, were followed. Trimers were obtained as off-white to yellow solid compounds. Tetramers and longer scaffolds can be obtained following the same procedures.

- It is to be noted that representative R groups (a-h) and R" can be varied broadly to optimize
25 the biological activity and specificity of the α -helix mimics. The nitrile group can be modified to optimize the physical properties of the scaffolds (the cyano group can be reduced to the primary amine, hydrolyzed to a carboxylate, etc.) using conditions leaving the remainder of the scaffold unchanged.

- 30 EXAMPLE 8 – SEMI-RIGID SCAFFOLD DERIVED FROM A REPETITIVE TRIMERIC 2,5-PYRIMIDINE SCAFFOLD.

A 216-membered library using a semi-rigid scaffold derived from a repetitive trimeric 2,5-pyrimidine scaffold that holds 6 structurally diverse side chain residues, (methyl, hydroxymethyl, isobutyl, benzyl, methyl(4-hydroxyphenyl), and methyl(3-indolyl) groups) in the 4, 4', 4" positions of a repetitive trimeric 2,5-pyrimidine scaffold can be synthesized
35 employing the six side chain residues, characterized and subjected to high-throughput screening (HTS) evaluation. The methyl, hydroxymethyl, isobutyl, benzyl, methyl(4-hydroxyphenyl), and methyl(3-indolyl) groups are selected to mimic natural, but structurally diverse amino acid side chains found in alanine, serine, leucine, phenylalanine, tyrosine, and

5 tryptophan, respectively. The 4, 4', 4'' positions of the repetitive trimeric 2,5-pyrimidine scaffold are designed to mimic the *ith*, *ith*+3 or *ith*+4, and *ith*+7 positions of an α -helix. A 6 x 6 x 6 matrix with 3 independent positions (the 4, 4', and 4'' positions, the R, R' and R'' groups shown in Figure 9 with the 6, 6' and 6'' positions equal to H) generates 216 different combinations. The N-terminus-like and C-terminus-like groups can be varied, but
10 carboxylates have been chosen to generate good water-solubility for the resulting library members as has been done by others with the less polar terphenylene scaffolds [Ernst, J.T., et al., *Angewandte Chemie, International Edition*, (2002) 41(2), 278-281; Kutzki, O., et al., *J Am Chem Soc.*, (2002) 124(40), 11838-11839]. The associated chemistry generates six intermediate 4-substituted monomeric 2,5-pyrimidine dicarboxylates, as well as the 6 x 6
15 matrix of 36 possible intermediate 4, 4'-substituted dimeric 2,5-pyrimidine dicarboxylates. These intermediate monomeric and dimeric 2,5-pyrimidines can act as SAR probes for trimeric 2,5-pyrimidines that are confirmed HTS hits and can have bioactivity independent of the α -helix mimicry designed for the trimeric 2,5-pyrimidine library. The monomeric and dimeric 2,5-pyrimidines are structural mimics of the biphenyl moiety; the biphenyl moiety is a
20 well recognized privileged structure in drug discovery [Horton, DA, et al., *Chem Rev.* (Washington, DC, United States), (2003) 103(3), 893-930] and pyrimidines are the basic unit in a number of active site tyrosine kinase inhibitors [Deininger, M., et al., *Pharmacol. Rev.*, (2003) 55, 401-423; Lagoja, IM, et al., *Chem Biodivers.*, (2005) 2(1), 1-50]. The proposed intermediate monomeric, dimeric, and trimeric 2,5-pyrimidine dicarboxylates gives 258
25 compounds for HTS testing.

EXAMPLE 9 – SEMI-RIGID SCAFFOLD DERIVED FROM A REPETITIVE TRIMERIC 2,5-PYRIMIDINE SCAFFOLD EXHIBITING MORE POLAR PROFILE DUE TO THE NH₂ 6, 6', AND 6'' SUBSTITUENTS.

A structurally similar (to the Example 8 semi-rigid scaffold), but more polar 216-membered
30 library, where the 6, 6', and 6'' substituents equal to NH₂ as shown in Figure 9, can also be synthesized, characterized and subjected to high-throughput screening (HTS) evaluation. The associated chemistry generates six intermediate 6-amino-4-substituted monomeric 2,5-pyrimidine dicarboxylates and a 6 x 6 matrix of 36 possible intermediate 6, 6'-diamino-4, 4'-substituted dimeric 2,5-pyrimidine dicarboxylates for this library as well. The intermediate
35 monomeric, dimeric, and trimeric 2,5-pyrimidine dicarboxylates gives 258 compounds that can be submitted for HTS testing. The di-*ortho* substitution of this library (the 4, 4' and 4'' and 6, 6', and 6'' substituents) will reduce the rotational freedom between the single bonds connecting the 2,5-pyrimidine subunits for the Example 9 library relative to the Example 8 library, this additional rigidity can reduce or increase the bioactivity of the resulting library, so
40 SAR comparisons between the Example 8 and Example 9 libraries are of interest. The NH₂ functional groups can also be used as sites for further structural diversification, for instance,

5 acylation or reductive amination of the NH₂ functional groups before each iterative 2,5-pyrimidine extension can give further diversified substituents, whereas all of the NH₂ functional groups can be derivatized after the trimeric 2,5-pyrimidine synthesis is complete to give the same new substituent at all of those positions. The Michael acceptors, discussed in the immediately following paragraphs, makes hybrids, based upon the compounds of
10 Example 8 and Example 9 libraries, an additional possibility.

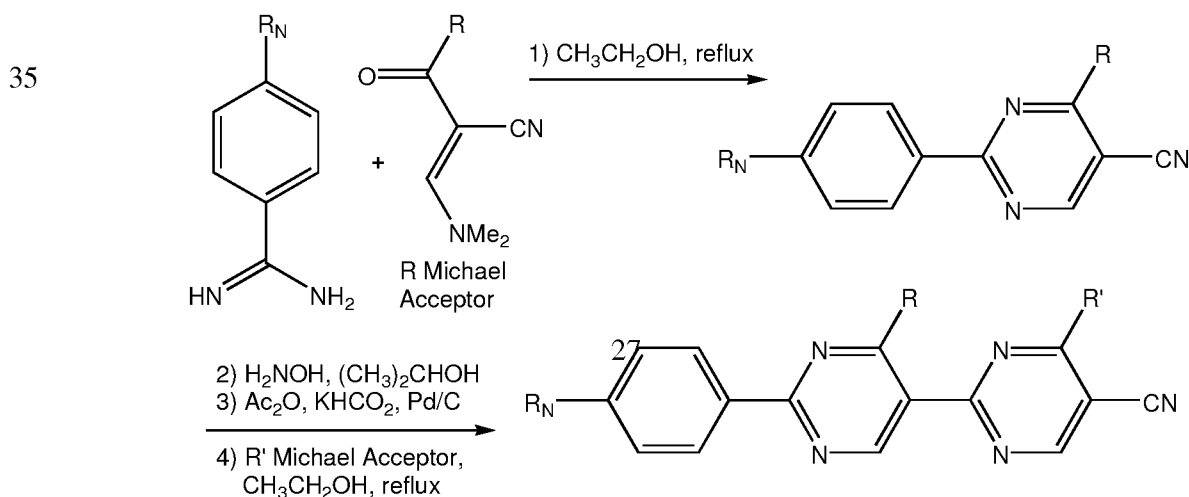
Experimental Design For the Production of – Examples 8 and 9.

D1. Overall approach and Example 8-type library synthesis:

Hundreds of pyrimidines are naturally occurring [Lagoja, IM, et al., *Chem Biodivers.*, (2005) 2(1), 1-50] and >400,000 different monomeric pyrimidines have been reported according to a
15 SciFinder Scholar search for pyrimidine preparations with biological data. [See also von Angerer, S., *Science of Synthesis*, (2004) 16, 379-572]. As a result of the ability of pyrimidines to mimic the basic unit of active site kinase inhibitors, many of the 400,000 pyrimidines have been reported in the patent literature. [Deininger, M., et al., *Pharmacol. Rev.*, (2003) 55, 401-423]. Examples of such reported pyrimidines include imatinib or
20 GleevecTM, dasatinib, and nilotinib. Consequently, the chemistry required to synthesize pyrimidines is well developed. The most common method of making pyrimidines is by condensation of an carboxamide or a guanidine with a Michael acceptor, such as the approach illustrated in Scheme D1 or D3 [Hill, MD, et al., *Chemistry*, (2008) 14(23), 6836-6844; von Angerer, S., *Science of Synthesis*, (2004) 16, 379-572]. Pyrimidines preparation is
25 straight-forward with these techniques. With the exception of a report on the synthesis of unsubstituted oligo-2,5-pyrimidines [Gompper, R, et al., *Synthesis*, (1997) 6:696-71], trimeric-2,5-pyrimidines or longer oligo-2,5-pyrimidines were unknown prior to the present exploration of a facile iterative synthesis of specifically 4,4',4"-substituted trimeric 2,5-pyrimidines. Gompper, et al examined the optical and electronic properties of the oligo-2,5-pyrimidines and
30 used a Stille-coupling-based synthesis. A Stille-coupling-like approach could be utilized to make the basic oligo-2,5-pyrimidines structures, but it would require more steps due to the starting materials, making that synthetic approach less practical for the synthesis of 4,4',4"-substituted trimeric 2,5-pyrimidines. The adaptation of a key enabling synthetic transformation as taught herein allows efficient 5-cyano and 5'-cyano group conversion to the
35 5- or 5'-carboxamide group using excess hydroxylamine followed by reduction of the intermediate N-hydroxylcarboxamide to the carboxamide as shown in Steps 2 and 3 of Scheme D1. The N-hydroxylamine mediated transformation of *ortho*-substituted and di-*ortho*-substituted arylcyano groups to carboxamides [Basso, A., et al., *Eur. J. Org. Chem.*, (2000) 23:3887-3891; Jozko, C, et al., *Tetrahedron Letters*, (2004) 45(40): 7445-7449; Judkins, BD, et al., *Synth. Comm.* (1996) 26(23):4351-4367] had not been used previously to prepare
40

- 5 oligo-2,5-pyrimidines as is proposed and taught herein. Without purification, the carboxamidine is reacted with the desired Michael acceptor to complete the iterative 2,5-pyrimidine synthesis. Repetition of these same steps gives the third pyrimidine unit and simple hydrolysis of the cyano group terminates the trimeric 2,5-pyrimidine library with a water-solubilizing substituent.
- 10 The greater than century-old Pinner reaction for conversion of cyano groups to carboxamidine groups fails when the arylcyano group has an *ortho*-substituent, which may provide one explanation for the absence of prior reports and the novelty of the compounds taught herein. The adaptation of the hydroxylamine mediated conversion of the 4- or 4'-substituted 5- or 5'-cyano group intermediate to the 4- or 4'-substituted 5- or 5'-carboxamidine group is one
- 15 critical, enabling step that makes it possible to prepare a non-peptide based α -helical mimetic library with just four easy isolation steps. Although Steps 2-4 in Scheme D1 are optimized in different solvents, it is submitted that solvent changes can be reduced or eliminated. However, even without common solvents for Steps 2-4, as in the examples, this is a remarkably efficient synthesis of libraries that can act as protein-protein interaction inhibitors
- 20 or as biological probes where the targets are not well-defined. The starting material for the library with $R_N = CO_2H$ is commercially available, but is expensive, therefore adaptation of the published procedures for the synthesis of 4-carboxamidinebenzoic acid or one its esters as a starting material for the library is one possibility [Basso, A., et al., *Eur. J. Org. Chem.*, (2000) 23:3887-3891; Jozko, C, et al., *Tetrahedron Letters*, (2004) 45(40): 7445-7449;
- 25 Judkins, BD, et al., *Synth. Comm.* (1996) 26(23):4351-4367]. An alternative is use of the ester, instead, if isolation of the intermediate carboxylate when $R_N = CO_2H$ is unexpectedly troublesome. Another potential alternative is the use a solid-phase synthetic approach, wherein the starting carboxamide is covalently attached to a solid-phase resin that, when cleaved from the solid-phase, unmask the R_N group that is desired. This could enable easy
- 30 split, pool and mix strategies to make very diverse libraries very quickly. However, the chemical steps needed to the pyrimidine ring formation and the cyano to carboxamidine conversion steps are relatively vigorous.

SCHEME D1



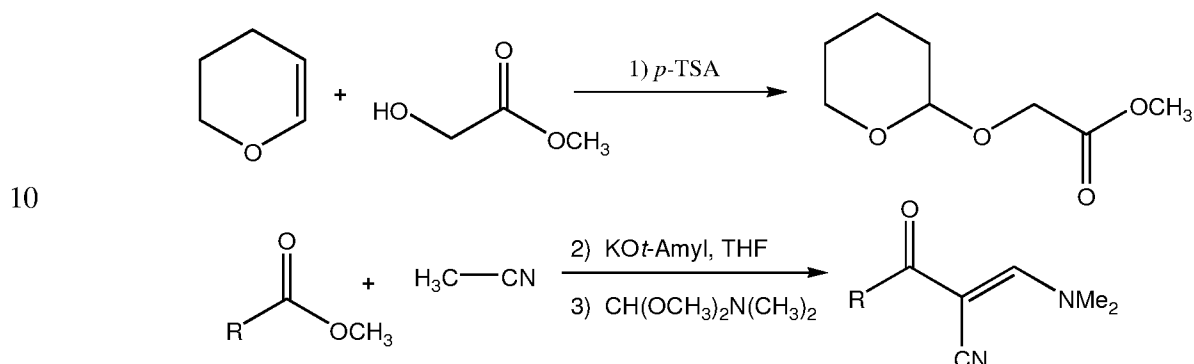
- 5 Scheme D1. An iterative synthesis of specifically 4, 4',4''-substituted trimeric 2,5-pyrimidines is summarized above. Steps 2-4 can be run without purification of the intermediates, following complete conversion of the cyano group as evidenced by TLC (4:1 hexanes/ethyl acetate), the solvent (typically isopropanol, dioxane for very hydrophobic intermediates) is removed in vacuo and the residue is taken up in anhydrous THF, acetic anhydride, potassium
- 10 formate, and 5 mole percent of 10% Pd/C is added. The reduction is continued until the least polar spot visualized by TLC (1:1 hexanes/ethyl acetate) is consumed and then Celite™ is added and the solids are removed by filtration on a sintered glass funnel. The filtrate is concentrated in vacuo and treated with the Michael acceptor in refluxing anhydrous ethanol. This step has also been run using microwave irradiation and the reaction is at least equally
- 15 efficient and much faster. Step 5, the cyano group hydrolysis, has been run exclusively in the microwave in a sealed reactor vessel with a yield greater than 90% and the product is greater than 90% pure. For the libraries containing the R, R', and/or R'' = CH₂-OTHP, an additional mild aqueous acid treatment will yield the fully deprotected product with R, R', and/or R'' = CH₂-OH.
- 20 D2. Michael acceptor selection and synthesis and Example 8-type Michael acceptors: A wide variety of R, R', and R'' substituents can be selected that are unreactive, or can be protected from reaction, during the trimeric 2,5-pyrimidine synthesis and cyano to carboxylate conversion steps. However, to explore the use of trimeric 2,5-pyrimidines R, R' and R''
- 25 substituents were limited to those that mimic a structurally diverse subset of the DNA-encoded amino acid side chains. This decision was based on the premise that these trimeric 2,5-pyrimidines are designed to be α -helical mimics. Furthermore, the subset was chosen to minimize chances that the vigorous reaction conditions during pyrimidine condensation, carboxamide synthesis, or cyano to carboxylate conversion will cause any undesirable side reactions. For instance, primary amides in Asn-like or Gln-like side chains would hydrolyze
- 30 or, worse, incompletely hydrolyze, during the cyano to carboxylate conversion and the Asp-like and Glu-like side chains could partially react with the hydroxylamine used in Step 2 of Scheme D1 and then incompletely hydrolyze during the cyano to carboxylate conversion, Step 5 of Scheme D1.
- Ile-like and Thr-like side chains were not chosen to avoid chiral starting materials due to their
- 35 costs. However, Ile-like and Thr-like side chains otherwise could have been excellent choices with the caveat that the Thr-like hydroxyl group would need protection/deprotection similar to the Ser-like side chain. Sulfur containing side chains were also not selected due to possible oxidation during synthesis or storage.
- The possibility of photoactivated side chain oxidation with the Tyr-like and Trp-like side chains
- 40 during long term storage is also a concern, but the structural diversity that these side chains

5 add to the library, makes them worth this slight risk, especially since these side chains are likely to be much less prone to oxidation than sulfur-containing side chains. Arg-like side chains were not selected because it would be incompatible with the pyrimidine condensation steps unless it was very well protected. Lys-like and His-like side chains could also have been selected, but protection/deprotection strategies would have been necessary, which
10 would add to the cost and time required to synthesize the library. Furthermore, variable amounts of titrable groups for the repetitive 2,5-pyrimidine scaffold side chains would have made the isolation steps substantially more complicated. For instance, if Asp- or Glu-like side chains were included as possibilities, the resulting overall trimeric 2,5-pyrimidine library would have had 2 to 5 possible negative charges at physiological pH, whereas the overall charge of
15 our proposed 216 member libraries are all the same within that library. With those factors in mind, Ala-like, Ser-like, Leu-like, Phe-like, Tyr-like and Trp-like side chains were selected for the libraries.

Val-like, Leu-like, Phe-like, and more hydrophobic unnatural amino acid-like side chains, as discussed below in the Results, are provided for ongoing studies to prepare focused libraries
20 as potential MDM family with p53 and Bcl family with Bak family protein-protein interaction inhibitors. For the Michael acceptors proposed, the synthesis is detailed in Schemes D2 and D4. Additional protection and eventual deprotection of the hydroxymethyl group for the Ser-like side chain will be necessary to eliminate unwanted cyclization reactions between the reactive hydroxyl group and the intermediate cyano derivative and/or the intermediate Michael
25 acceptor. THP protection and deprotection with primary alcohols, like the methyl glyoxylate example, is well-characterized (see e.g. has over 4000 examples in the SciFinder Scholar reaction database search engine; a recent example is cited [Minatti, A, et al., Org. Lett., (2008) 10(13), 2721-2724]. Tyr-like and Trp-like side chain functional groups need not be protected unless they unexpectedly cause side reactions. If needed, the OH and NH groups
30 on the Tyr-like and Trp-like side chains can be protected with acid-labile protecting groups to make the eventual deprotection easier since the THP de-protection can be done simultaneously. For instance, *t*-butyl ether and the *t*-Boc derivatives of the Tyr-like and Trp-like side chain starting materials are readily available, respectively.

5

Scheme D2



Where R = isobutyl and benzyl the overall yield for both Michael acceptors is > 60%

Where R = methyl the product is commercially available but expensive and very inexpensive to prepare from ethyl acetate

Where R = CH₂(4-hydroxyphenyl) and CH₂(3-indolyl) the esters are commercially available

Where R = CH₂-OTHP, commercially available methyl glyoxalate will be THP protected in Step 1

15

Scheme D2. The synthesis of the required Michael acceptors can utilize the methods used to prepare the isobutyl and benzyl substituted Michael acceptors [Ji, Y, et al., *Org. Lett.*, (2006) 8(6), 1161-1163; Reuman, M, et al., *J. Org. Chem.*, (2008) 73(3), 1121-1123]. The only difference is that the methyl glyoxylate should be protected as the THP ether [Minatti, A, et al., *Org. Lett.*, (2008) 10(13), 2721-2724] to prevent the hydroxyl group of that nitrile or that Michael acceptor intermediate to cyclize intramolecularly to form a five-membered ring product.

20

The β-ketonitriles from Step 2 of Scheme D2 can be made in excellent yields and purity by adapting the procedure of Ji et al. [Ji, Y, et al., *Org. Lett.*, (2006) 8(6), 1161-1163]. Step 3, the chemistry to prepare the Michael acceptors in excellent yields and purity can proceed according to Reuman et al. [Reuman, M, et al., *J. Org. Chem.*, (2008) 73(3), 1121-1123]. The basic or neutral conditions of Steps 2 and 3 in Scheme D2 will leave THP protecting group unchanged.

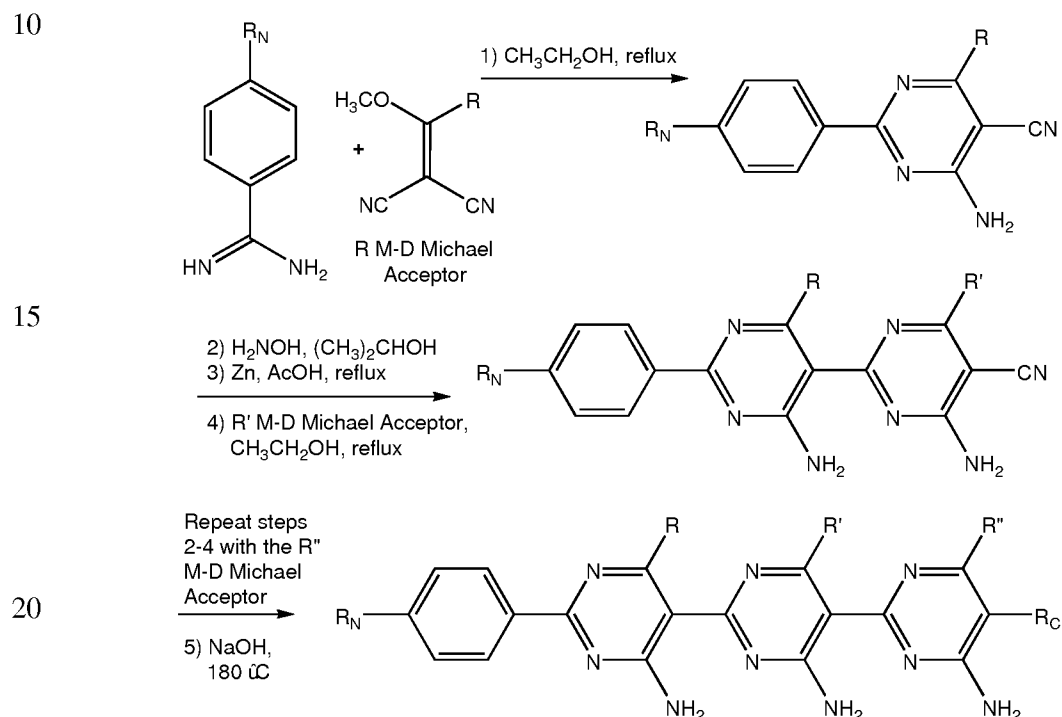
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D3. Example 9-type library synthesis: The chemistry to prepare the more polar 6, 6', 6''-triamino-4-R-, 4'-R'-, 4''-R''-substituted trimeric 2,5-pyrimidine scaffold is briefly outlined below in Scheme D3. The pyrimidine condensation where one of the cyano groups of a malonitrile-derived Michael acceptor (M-D Michael acceptor) acts as an acceptor to generate the 6-amino pyrimidine unit is more obscure than the pyrimidine condensation used in Scheme D1, but there exists corollaries for the process [Svetlik, J, et al., *J. of the Chem. Soc., Perkin Transactions* (2002) 1, (10), 1260-1265; Ochiai, M, et al., *Org. Lett.*, (2001) 3(17), 2753-2756; Masquelin, T, et al., *Helvetica Chimica Acta*, (1998) 81(4), 646-660; Baxter, RL, et al., *J. of the Chem. Soc., Perkin Transactions 1: Organic and Bio-Organic Chemistry* (1972-1999),

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- 5 (1990) (11), 2963-6; Tominaga, Y, et al., *Heterocycles*, (1987) 26(3), 613-16; Tominaga, Y, et al., *J. Heterocycl. Chem.*, (1990) 27(3), 647-60]. The design and rationale for selection of the side chains is the same as above and only minor differences in the actual synthetic schemes gives a chemically distinct, more polar trimeric 2,5-pyrimidine scaffold.

Scheme D3

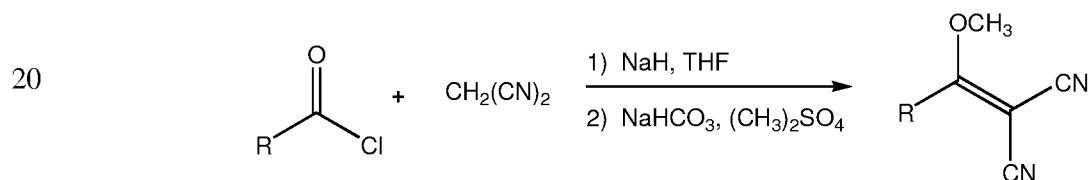


- Scheme D3. A more polar 4-R-, 4'-R', 4''-R''-substituted trimeric 2,5-pyrimidine scaffold using a malonitrile-derived Michael acceptor (M-D Michael acceptor), but having the same R, R', and R'' groups as in Scheme D1, is provided. Malonitrile derivatives used for this alternative trimeric 2,5-pyrimidine scaffold have been reported in the art. A synthesis is shown Scheme D4. The NH₂ group(s) would be acetylated if the acetic anhydride reagent, used in the hydrogenolysis step of Scheme D1 to reduce the intermediate N-hydroxylcarboxamidine intermediates, is utilized in the present reaction, so Step 3 has been changed to a dissolving metal reduction instead. A dissolving metal reduction approach is also an alternative for Step 3 of Scheme D1. That approach would be necessary to enable a solid-phase synthetic approach. Otherwise, Scheme D3 as outlined is similar to the earlier Scheme D1. When the R, R', or R'' side chains are benzyl protected, an additional hydrogenolysis step will be necessary. Benzyl side chain protection is proposed for all of the functionalized side chains in this scheme, but it may be unnecessary except for the Ser-like side chain. Here, as in

- 5 Scheme D1, probably none of the side chains need functional group protection except during the initial preparation of the M-D Michael acceptors in Scheme D4.

The M-D Michael acceptor where R = methyl is commercially available as the ethyl ether instead of the methyl ether. The other M-D Michael acceptors can be prepared by adapting the procedure described by Shokat and co-workers [Kraybill, BC, et al., *J. Am. Chem. Soc.*, (2002) 124(41), 12118-12128]. The R = benzyl M-D Michael acceptor is specifically reported with 90 % yields for Steps 1 & 2 of Scheme D4. The R = isobutyl M-D Michael acceptor is likely to give similar yields as the benzyl analog. The benzyloxyacetic acid and 4-(benzyloxy)phenylacetic acid are commercially available. Synthesis of N-benzyl-3-indolylacetic acid and its derivatives is as reported [Chapman, RF, et al., *Tetrahedron*, (1985) 41(22), 5229-34; Ihara, M, et al., *Tetrahedron*, (1985) 41(11), 2109-14]. All of these acids can be transformed into the acid chlorides and should prove efficient at converting to the methyl ether derivatives as shown in Steps 1 & 2 of Scheme D4.

Scheme D4



Where R = methyl the ethyl ether M-D Michael acceptor is commercially available

Where R = benzyl the yield for **Steps 1 & 2** are 90%

Where R = isobutyl the acid chloride is commercially available and inexpensive

Where R = CH₂OH, CH₂(4-hydroxyphenyl) and CH₂(3-indolyl) the OH and NH groups will be O-benzyl, O-benzyl, and N-benzyl protected, respectively

25

Scheme D4. The synthesis of the M-D Michael acceptors needed to prepare the 6, 6', 6''-triamino-4-R, 4'-R', 4''-R''-substituted trimeric 2,5-pyrimidines is shown above.

D4. Purification and synthesis equipment for the monomeric, dimeric, and trimeric 2,5-pyrimidines: Example 8-type monomeric pyrimidines prepared thus far, examples of which are presented in the Results, below, precipitate or crystallize as the pure products from cooled ethanol solutions. Purity has been pursued at the expense of yield, because none of these intermediates are expensive to prepare and this same strategy is adopted for the libraries. It is easy to separate unreacted carboxamidines from the much less polar pyrimidine products. On occasions where unreacted Michael acceptors were present, those compounds will coprecipitate with the product pyrimidines. Monomeric Example 8-type pyrimidines where R_N = CO₂H should precipitate or crystallize similarly. Their purification maybe aided due to the titratable carboxyl group of the intermediate monomeric and dimeric 2,5-pyrimidines. Several of the dimeric and trimeric Example 8-type 2,5-pyrimidines have

5 also precipitated or crystallized upon cooling and recrystallized. For example, a dimer crystal structure has been determined; the ORTEP diagram of the crystal structure of this dimeric 2,5-pyrimidine scaffold is shown in the Results section in Figure 11. Also, some of the dimeric and trimeric Example 8-type 2,5-pyrimidines have required flash column chromatographic purification. This has been done using manual columns or using Biotage
10 Flashmaster Parallel purification. CEM Microwave Synthesizers have also been used.

D5. Characterization of the Michael acceptors, monomeric, dimeric, and trimeric 2,5-pyrimidines: Compounds will be analyzed by ^1H , ^{13}C NMR, and IR spectra for all new compounds. MestReNova (v. 5.0.2) software can be used to process the generated data for the intermediates and final products of the large focused libraries. ESI- or APCI-MS data can
15 also be obtained for all compounds. An HPLC system can be used to verify greater than 90% purity using two different conditions such as different solid supports, eluting solvents, and/or temperature for all library members.

D6. Preparation of MLPCN library member samples for submission: A robotic weighing station can be used for testing in the Moffitt High-Throughput Screening Core
20 Facility. Alternatively, samples can be prepared manually for submission to the MLPCN. Where a robotic weighing station is used, manual preparation of starting materials and to prepare samples to obtain NMR spectra, MS, IR, and HPLC data can be performed, as well as determining the solubility of the library members in (90:10) chloroform/methanol or the DMSO solubility when the library members are not soluble in those solvents.

25 D7. Alternative water-solubilizing R_N or R_C groups: The cyano group can be converted to the carboxylate group (the R_C group in Figure 9). This approach is one of the primary approaches that has been used in the chemistry to prepare the water-solubilized trimeric Example 8-type 2,5-pyrimidines reported in the Results section C1 where the R_N group equals to H. Those library members have some water solubility, but to improve the water
30 solubility, R_N and R_C groups can be equal to carboxylate groups. The R_C group can be converted to a primary amino group or the cyano group can be converted to an aldehyde group and then reductively aminated with almost any primary or secondary amine to prepare groups that would be positively charged at physiological pH. The cyano group or R_C group of a few Example 8-type pyrimidines have been converted to a $\text{CH}_2\text{-NH}_2$ or the CHO group by
35 reduction and partial reduction and hydrolysis as described in the Results section C1. The conditions used for those reductions, lithium aluminum hydride and Dibal-H reactions followed by mild aqueous acid treatment, respectively, are not compatible with the R_N group when it equals a carboxylate group because it would also be reduced. Switching to a positively charged group at the R_C group could be accomplished by starting the pyrimidine synthesis
40 with a *p*-substituted tertiary amine already appended to the benzamidine. That *p*-substituted

- 5 tertiary amine benzamidine would be used to initiate the trimeric 2,5-pyrimidine synthesis. Where benzamidine or a substituted benzamidine is not required, monomeric Example 8-type pyrimidines have been prepared using formamidine, acetamidine, and guanidine, but the benzamidine derived monomeric Example 8-type pyrimidines have exhibited better crystallization. The cyano group, the R_C group of the pyrimidines, has been converted to a
10 tetrazole ring by microwave-assisted reaction of sodium azide and triethylammonium acetate in DMF at 180 °C. The tetrazole ring is often considered a bioisostere for a carboxylate group that makes the resulting drug candidate less susceptible to *in vivo* modification, such as a carboxylate to ester conversion, but the cyano to carboxylate conversion can be completed in much less time and better purity of the final product.
- 15 D8. Alternative trimeric 2,5-pyrimidine side chains: Since the goal is to prepare a chemically diverse trimeric 2,5-pyrimidine library, the actual side chains used can be varied. Almost any side chain structurally distinct from those already chosen and that behave well in the library synthesis can be exchanged for a side chain that does not behave well. For instance, a trityl-protected His-like side chain is likely to be tolerated well and is structurally
20 very diverse compared to the other selected side chains. Also, numerous side chains that are not at all structurally related to the DNA-encoded amino acid-like side chains could be chosen. Molecular diversity analysis can guide the selection of alternative side chains if needed. Structural diversity from the other selected side chains and chemical compatibility with the synthetic methods used to prepare alternative trimeric 2,5-pyrimidine libraries can be
25 employed as the selection criteria. If needed, the OH and NH groups on the Tyr-like and Trp-like side chains can also be protected with acid-labile protecting groups to make the eventual deprotection easier since the THP deprotection can be done simultaneously. For instance, *t*-butyl ether and the *t*-Boc derivatives of the Tyr-like and Trp-like side chains are readily available, respectively. Tyr-like and Trp-like side chain functional groups need not be
30 protected unless they unexpectedly cause side reactions. The acidic conditions used in the dissolving metal reduction conditions can remove the acid labile protecting groups, but that should not matter since the OH and NH protecting groups are only likely to be a potential issue during the Michael acceptor synthesis steps.
- 35 CombiGLIDE Diverse Side-chain Collection, version 1.2, available from Schrödinger, L. L. C., can be used to guide selection. This library consists of 817 side chains that are suitable for combinatorial library design using the CombiGLIDE computer program (see section D10). These substituents have been selected not only for maximum structural diversity but also to provide representative groups commonly found in pharmacologically active compounds and they possess linkers of various lengths.

5 In order to characterize the chemical diversity of the oligopyrimidine library, software such as ChemoSoft (available from ChemDiv, Inc.) or Canvas (currently in beta release from Schrödinger, LLC) can be employed. Thus, clustering of the similarity (or distance, i.e. dis-
10 2D chemical fingerprints. This would allow for an assessment of the chemical diversity of the library relative to other collections.

D9. Development of verified hits, hit to lead (HTL) optimization: HTL optimization could proceed along several parallel or independent pathways and can be guided by molecular modeling studies described in detail in section D10. Making Michael acceptors with side
15 chains designed to fine tune binding to identified targets can be done since the Michael acceptors are prepared from readily available starting materials in two experimentally uncomplicated and high yielding steps (Steps 2 and 3 of Scheme D2 or Steps 1 and 2 of Scheme D4). Fine tuning the binding to the R" position while leaving the R and R' positions unchanged would be the recommended course since only a single dimeric 2,5-pyrimidine
20 would give trimeric 2,5-pyrimidines with the variable R" position. Once the R" position is fine-tuned, the same process can be used to fine tune the R' and then R positions. To speed the analoging efforts, a 20 g stockpile of each of the Michael and M-D Michael acceptors can be prepared, along with preparation of 36 g stockpiles of each of the monomeric Example 8- and Example 9-type 4-R-substituted-5-cyanopyrimidines, which is the 12 possible monomeric
25 pyrimidines, and 6 g stockpiles of each of the 36 dimeric Example 8- and Example 9-type 4-R-, 4'-R'-substituted-5-cyano-2,5-pyrimidines, which is 72 possible dimeric 5"-cyano-2,5-pyrimidines, and 1 g stockpiles of each of the 216 different trimeric Example 8- and Example 9-type 4-R-, 4'-R'-, 4"-R"-substituted-5"-cyano-2,5-pyrimidines, which is 432 possible trimeric
30 intermediate nitriles. For instance, if one or more trimeric library members is active, a tetrameric library with the same previously prepared trimeric 5"-cyano-2,5-pyrimidines can be made within days by just doing Steps 2-5 of Scheme D1 just once to give a tetrameric 4-R-, 4'-R'-, 4"-R"-variable 4""-R"" substituted tetrameric 2,5-pyrimidine library. Similarly, the charge character of the R_N and R_C groups can then be varied and/or the starting benzamidine
35 can be changed to many other fragments as shown in the Results section C1. It is also possible to prepare pentameric or longer 2,5-pyrimidines by simply repeating Steps 2-4 the appropriate number of times and then proceed to Step 5. The trimeric 2,5-pyrimidines are designed to mimic the side chain interactions of two turns of an α -helix (the *ith*, *ith*+3 or *ith*+4, and *ith*+7 residues), but there is no reason that a well-designed α -helix mimic cannot mimic
40 three or more turns of an α -helix. For instance, a tetrameric heterocyclic scaffold designed to mimic the *ith*, *ith*+4, *ith*+7, *ith*+11 residues of an α -helix has been reported [Restorp, P., et al.,

5 Bioorganic & Medicinal Chemistry Letters, (2008) 18(22), 5909-5911]. A hexameric 2,5-pyrimidine could theoretically mimic five turns of an α -helix (the *ith*, *ith*+4, *ith*+7, *ith*+11, *ith*+14, and *ith*+18 residues). Two turns of an α -helix places the *ith* and *ith*+7 residues approximately 10 Å apart, five turns of an α -helix places the *ith* and *ith*+18 residues approximately 27 Å apart, so a hexameric 2,5-pyrimidine is reasonably referred to as
10 proteomimetic instead of a peptidomimetic.

D10. Structure-based molecular modeling of verified hits as a component of hit to lead (HTL) optimization: Molecular modeling studies involving structure-based computational docking can be employed to aid in the HTL optimization described in section D9. The GLIDE program (Grid Based Ligand Docking from Energetics – available from Schrödinger, LLC) can
15 serve as the foundation for docking studies performed using X-ray or NMR structures of protein targets of interest, like MDM2, MDMX, Bcl-xL and Mcl-2. GLIDE is well suited for such investigations since studies comparing various docking methods rank GLIDE among the most accurate [Kellenberger, E, et al., *Proteins*, (2004) 57, 225-242; Perola, E, et al., *Proteins*, (2004) 56, 235-49]. CombiGLIDE (Schrödinger, LLC), a variant of GLIDE, can be used to aid
20 in the design of the focused chemical libraries. CombiGLIDE allows the user to utilize a docked core structure (for example, our oligo-pyrimidine core) as a template upon which substituents are added combinatorially to user-defined attachment sites to generate a library of structures that are then docked to the protein binding site using GLIDE. Other docking software can also be utilized (for consensus docking and scoring). AutoDock (The Scripps
25 Research Institute) and GOLD (The Cambridge Crystallographic Data Center) are examples of software that can be used for this purpose. The MM/GBSA method available within the PRIME program (Schrödinger, Inc.) will be used to compute more accurate binding free energies for our compounds than the ones calculated using the approximate scoring functions utilized by GLIDE, AutoDock and GOLD. Protein flexibility that accompanies binding will be
30 modeled using Schrödinger's Induced Fit Module that combines PRIME and GLIDE and/or the Low Mode method [Kolossvary, I., et al., *J. Comput. Chem.*, (1999) 20, 1671-84].

In order to generate oligo-2,5-pyrimidines that have optimal ADME characteristics, the QikProp program (developed by Prof. Bill Jorgensen, Yale University and available from Schrödinger, LLC) can be employed. QikProp is based upon linear correlations that were
35 previously established between a number of ADME properties and 2D and 3D descriptors calculated for a "training set" of known drugs with experimentally determined ADME properties (ca. 700 compounds). Use of 2D and 3D descriptors calculated for the compound of interest, provides a prediction of the experimental ADME properties for the molecule. Thus, a number of important ADME properties including Caco-2 cell permeability, aqueous
40 solubility, log $P_{\text{octanol/water}}$, and human serum albumin binding can be predicted using QikProp. QikProp automatically flags those properties that fall outside of the 95% range of the known

- 5 drugs in its training set. The raw data comparing a Hamilton-like terphenylene scaffold, and an Example 8-type and Example 9-type library with the same side chains are compared in the Results section in Figure 14.

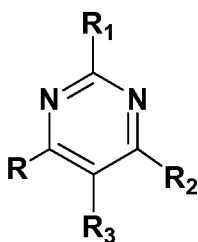
These experiments in section D10 specifically refer to our ongoing studies with structure-based molecular modeling of verified hits as a component of HTL optimization using MDM2,
10 MDMX, Bcl-xL and Mcl-2 as examples, but other targets found by MLPCN screening with at least 2.5 Å resolution structural data available will also be pursued.

RESULTS – Examples 8 and 9:

The tables presented below document the collection of data for the monomeric, dimeric, and trimeric 2,5-pyrimidines synthesized and tabulated to date.

- 15 C1. Tabulated synthesis results to date on monomeric, dimeric, and trimeric 2,5-pyrimidines: Starting materials, organic and inorganic reagents (ACS grade), and solvents were obtained from commercial sources and used as received unless otherwise indicated. Moisture- and air-sensitive reactions were carried out under an atmosphere of argon. Thin layer chromatography (TLC) was performed on glass plates pre-coated with 0.25 mm
20 thickness of silica gel (60 F-254) with fluorescent indicator (EMD or Whatman). Column chromatographic purification was performed using silica gel 60 Å, #70-230 mesh (Selecto Scientific). Automated flash chromatography was performed in a Flashmaster II system (Argonaut-Biotage) using Biotage silica cartridges. ¹H NMR and ¹³C NMR spectra were obtained using a 400 MHz Varian Mercury plus instrument at 25 °C in chloroform-*d* (CDCl₃)
25 unless otherwise noted. Chemical shifts (δ) are reported in parts per million (ppm) relative to internal tetramethylsilane (TMS). Multiplicity is expressed as (s = singlet, br s = broad singlet, d = doublet, t = triplet, q = quartet, dd = doublet of doublets, dt = doublet of triplets, or m = multiplet) and the values of coupling constants (J) are given in hertz (Hz) as determined by the MestRec (v. 4.9.9.9) or MestReNova (v. 5.0.2) software. High Resolution Mass
30 Spectrometry (HRMS) spectra were carried out on an Agilent 1100 Series in the ESI-TOF mode. Microwave reactions were performed in a Biotage Initiator I microwave reactor. Melting points (uncorrected) were determined using a Mel-Temp II®, Laboratory Devices, MA, USA.

VARIATION OF 2- POSITION



5

Entry No.: 1

$R_1 = \text{Ph}$; % Yield for $R = i\text{Pr}$ = 87%

Characterization: ^1H NMR (CDCl_3 , 400 MHz) δ 8.90 (s, 1H), 8.53(d, 2H, $J=3.6$ Hz), 7.53(m, 3H), 3.48(m, 1H), 1.43(d, 6H, $J=3.4$ Hz) ^{13}C NMR (400 MHz, CDCl_3): δ 178.2, 165.9, 160.6, 136.5, 132.4, 129.4, 128.9, 115.6, 105.2, 35.2, 21.4. HRMS (ESI+) $\text{C}_{14}\text{H}_{13}\text{N}_3$ (M+H), 223.1187, found, 224.1178.

10

Entry No.: 2

$R_1 = \text{Me}$; % Yield for $R = i\text{Pr}$ = 90%

Characterization: ^1H NMR (CDCl_3 , 400 MHz) δ 8.74 (s, 1H), 3.38(m, 1H), 2.75 (s, 3H), 1.30(d, 6H, $J=5.8$ Hz).

15

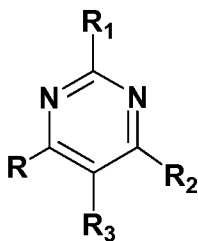
Entry No.: 3

$R_1 = \text{NH}_2$; % Yield for $R = i\text{Pr}$ = 80%

Characterization: ^1H NMR (CD_3COCD_3 , 400 MHz) δ 8.41 (s, 1H), 6.95 (br, 2H, NH), 3.11 (m, 1H), 1.13(d, 6H, $J=6$ Hz). ^{13}C NMR (400 MHz, CD_3COCD_3): δ 178.9, 164.2, 162.4, 116.6, 95.2, 34.3, 20.4. HRMS (ESI+) $\text{C}_8\text{H}_{10}\text{N}_4$ (M+H), 163.0983, found, 163.0981.

20

VARIATION OF THE R-GROUP

Entry No.: 1

$R_1 = \text{Ph}$; $R = i\text{Pr}$; % Yield = 87%

Characterization: ^1H NMR (CDCl_3 , 400 MHz) δ 8.90 (s, 1H), 8.53(d, 2H, $J=3.6$ Hz), 7.53(m, 3H), 3.48(m, 1H), 1.43(d, 6H, $J=3.4$ Hz) ^{13}C NMR (400 MHz, CDCl_3): δ 178.2, 165.9, 160.6,

25

- 5 136.5, 132.4, 129.4, 128.9, 115.6, 105.2, 35.2, 21.4. HRMS (ESI+) $C_{14}H_{13}N_3$ (M+H), 223.1187, found, 224.1178.

Entry No.: 2

R_1 = Ph; R = 2-(cyclohexyl)ethyl; % Yield = 90%

- 10 Characterization: ^1H NMR ($CDCl_3$, 400 MHz) δ 8.83 (s, 1H), 8.44 (d, 2H, J = 3.2 Hz), 7.46 (m, 3H), 2.98 (m, 2H), 1.78-0.82 (m, 13H). HRMS (ESI+) $C_{19}H_{21}N_3$ (M+H), 292.1813, found, 292.1830.

Entry No.: 3

R_1 = Ph; R = *i*-Bu; % Yield = 70%

- 15 Characterization: ^1H NMR ($CDCl_3$, 400 MHz) δ 8.93 (s, 1H), 8.52 (dd, 2H, J = 1.6, 8.2), 7.54 (m, 3H), 2.93 (d, 2H, J = 7.2), 2.39 (d p, 1H, J = 6.8, 13.6), 1.05 (s, 3H), 1.04 (s, 3H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 22.57, 28.88, 45.71, 106.71, 115.80, 129.00, 129.38, 132.44, 136.36, 160.37, 165.72, 173.29. HR-MS 238.1337 $[M+H]^+$. m.p. 68-69 °C.

Entry No.: 4

R_1 = Ph; R = methyl(1-naphthyl); % Yield = 49%

- 20 Characterization: ^1H NMR ($CDCl_3$, 400 MHz) δ 8.94 (s, 1H), 8.45-8.38 (m, 3H), 7.87 (d, J = 8.12 Hz, 1H), 7.82 (d, J = 8.22 Hz, 1H), 7.63-7.44 (m, 7H), 4.83 (s, 2H).

Entry No.: 5

R_1 = Ph; R = Bn; % Yield = 86%

- 25 Characterization: ^1H NMR ($CDCl_3$, 400 MHz) δ 8.92 (s, 1H), 8.53-8.49 (m, 2H), 7.59-7.44 (m, 5H), 7.37-7.24 (m, 3H), 4.36 (s, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 173.49, 163.08, 162.49, 136.20, 129.45, 128.97, 127.45, 116.62, 97.75, 42.97.

Entry No.: 6

R_1 = NH_2 ; R = Bn; % Yield = 91%

- 30 Characterization: ^1H NMR ($CDCl_3$, 400 MHz) δ 8.44 (s, 1H), 7.38-7.23 (m, 5H), 5.56 (s, 2H), 4.09 (s, 2H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 173.49, 163.08, 162.49, 136.20, 129.45, 128.97, 127.45, 116.62, 97.75, 42.97.

Entry No.: 7

R_1 = NH_2 ; R = *i*-Pr; % Yield = 80%

- 5 Characterization: ^1H NMR (CD_3COCD_3 , 400 MHz) δ 8.41 (s, 1H), 6.95 (br, 2H, NH), 3.11 (m, 1H), 1.13(d, 6H, $J=6$ Hz). ^{13}C NMR (400 MHz, CD_3COCD_3) δ 178.9, 164.2, 162.4, 116.6, 95.2, 34.3, 20.4. HRMS (ESI+) $\text{C}_8\text{H}_{10}\text{N}_4$ (M+H), 163.0983, found, 163.0981.

Entry No.: 8

$\text{R}_1 = \text{NH}_2$; R= *i*-Bu; % Yield = 75%

- 10 Characterization: ^1H NMR (CD_3SOCD_3 , 400 MHz) δ 8.54 (s, 1H), 7.66 (s, 2H), 2.50 (d, 3H, $J = 7.4$), 2.07 (m, 1H), 0.90 (s, 3H), 0.89 (s, 3H). ^{13}C NMR (100 MHz, CDCl_3) δ 22.57, 28.88, 45.71, 106.71, 115.80, 129.00, 129.38, 132.44, 136.36, 160.37, 165.72, 173.29. HR-MS 238.1337 $[\text{M}+\text{H}]^+$. M.P. 68-69 $^\circ\text{C}$.

Entry No.: 9

- 15 $\text{R}_1 = \text{Me}$; R= *i*-Pr; % Yield = 90%

Characterization: ^1H NMR (CDCl_3 , 400 MHz) δ 8.74 (s, 1H), 3.38(m, 1H), 2.75 (s, 3H), 1.30(d, 6H, $J=5.8$ Hz).

Entry No.: 10

$\text{R}_1 = \text{Me}$; R= 2-(cyclohexyl)ethyl; % Yield = 64%

- 20 Characterization: ^1H NMR (CDCl_3 , 400 MHz) δ 8.73 (s, 1H), 2.91 (m, 2H), 2.73 (s, 3H), 1.76-0.85(m, 13H); ^{13}C NMR (400 MHz, CDCl_3) δ 174.4, 170.8, 159.9, 115.2, 105.3, 37.8, 36.4, 34.7, 33.2, 26.8, 26.7, 26.4. HRMS (ESI+) $\text{C}_{14}\text{H}_{19}\text{N}_3$ (M+H), 230.1657, found, 230.1646.

Entry No.: 11

$\text{R}_1 = \text{Me}$; R= methyl(1-naphthyl); % Yield = 60%

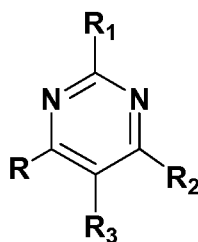
- 25 Characterization: ^1H NMR (CDCl_3 , 400 MHz) δ 8.80 (s, 1H), 8.27 (d, $J = 8.41$ Hz, 1H), 7.87 (d, $J = 8.15$ Hz, 1H), 7.81 (d, $J = 7.99$ Hz, 1H), 7.57-7.42 (m, 4H), 4.73 (s, 2H), 2.75 (s, 3H); ^{13}C NMR (100 MHz, CDCl_3) δ 171.51, 171.07, 160.64, 134.18, 132.18, 132.13, 129.02, 128.60, 128.56, 126.59, 126.10, 125.70, 124.44, 115.43, 106.24, 40.66, 26.87.

Entry No.: 12

- 30 $\text{R}_1 = \text{H}$; R= Bn; % Yield = 10%

Characterization: ^1H NMR (400 MHz, CDCl_3) δ 8.44 (s, 1H), 7.38-7.23 (m, 5H), 5.56 (s, 2H), 4.09 (s, 2H).

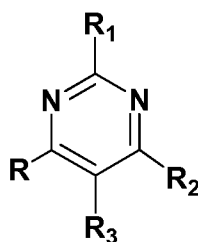
VARIATION IN R_2



5

Entry No.: 1R = NH₂; R₁ = Ph; R₂ = Ph; R₃ = CN; % Yield = 84%Characterization: ¹H NMR (400 MHz, CDCl₃) δ 8.51 (m, 2H), 8.13 (m, 2H), 7.53 (m, 6H), 5.77 (s, 2H). ¹³C NMR (100 MHz, CDCl₃) δ 85.41, 116.67, 128.74, 128.93, 129.03, 129.25,

10 131.67, 132.10, 136.47, 136.58, 164.82, 165.38, 168.37.

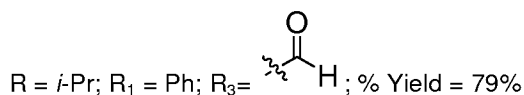
Entry No.: 2R = NH₂; R₁ = Ph; R₂ = H; R₃ = CN; % Yield = 74%Characterization: ¹H NMR (400 MHz, CDCl₃) δ 8.59 (s, 1H), 8.33 (d, 2H, J = 6.8), 7.44 (m, 3H), 5.58 (s, 2H).15 VARIATION IN R₃: FUNCTIONALIZATION OF TERMINAL GROUPEntry No.: 1R = 2-(cyclohexyl)ethyl; R₁ = Me; R₃ = ;

% Yield = 83%

20 Characterization: ¹H NMR (CDCl₃, 400 MHz) δ 9.20 (s, 1H), 3.19 (m, 2H), 2.81 (s, 3H), 1.80-0.92 (m, 13H). HRMS (ESI+) C₁₄H₂₀N₂O₂ (M+H), 249.1614, found, 249.1617.Entry No.: 2R = i-Bu; R₁ = Ph; R₃ = ; % Yield = 75%

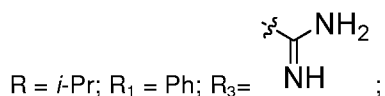
- 5 Characterization: ^1H NMR (400 MHz, CD_3OD) δ 9.19 (s, 1H), 8.48 (d, 2H, $J = 8.1$), 7.51 (m, 3H, $J = 7.5$), 3.17 (d, 2H, $J = 7.1$), 2.28 (d p, 1H, $J = 6.8, 13.6$), 1.00 (s, 3H), 0.99 (s, 3H). ^{13}C NMR (100 MHz, CD_3OD) δ 165.2, 161.9, 159.4, 137.0, 131.4, 128.6, 128.5, 44.5, 28.7, 21.7.

Entry No.: 3



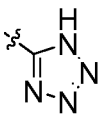
- 10 Characterization: ^1H NMR (400 MHz, CDCl_3) δ 10.28(s, 1H), 9.01(s, 1H), 8.52(d, 2H, $J=7.2$ Hz), 7.47-7.54(m, 3H), 3.90(quintet, 1H), (1.34 (d, $J=8$ Hz, 6 H); ^{13}C NMR (100 MHz, CDCl_3) δ 188.5, 175.8, 165.4, 159.8, 135.8, 130.9, 128.2, 127.9, 30.6, 28.9, 20.4.

Entry No.: 4

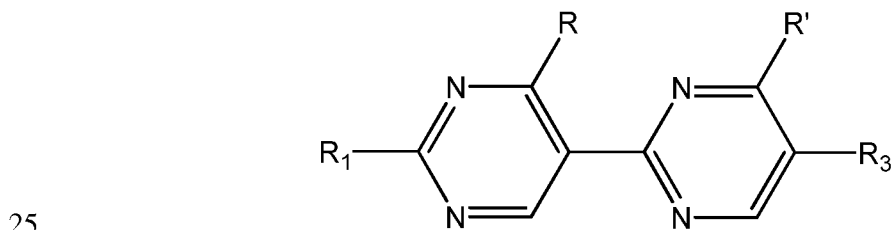


- 15 Characterization: ^1H NMR (CDCl_3 , 400 MHz) δ 8.72 (s, 1H), 8.45 (m, 2H), 7.45(m, 3H), 5.80 (br, 2H), 3.57 (m, 1H), 1.31(d, 6H, $J=3.4$ Hz). ^{13}C NMR (400 MHz, CDCl_3 +2 drops MeOH): δ 173.0, 167.3, 164.0, 154.2, 136.1, 130.2, 127.6, 124.2, 120.9, 31.6, 20.9.

Entry No.: 5

- 20 R = *i*-Pr; R_1 = Ph; R_3 = ; % Yield = 89%
- Characterization: ^1H NMR ($\text{DMSO}-d_6$, 400 MHz) δ 9.15 (s, 1H), 8.48 (m, 2H), 7.56 (m, 3H), 5.77(br, 1H), 1.30(d, 6H, $J=3.2$ Hz). ^{13}C NMR (400 MHz, $\text{DMSO}-d_6$): δ 173.5, 163.9, 158.1, 155.0, 137.5, 131.9, 129.5, 128.6, 118.2, 32.4, 22.1.

OLIGOMERS—PYRIMIDINE DIMERS



Entry No.: 1

R_1 = Ph; R = *i*-Bu; R' = Bn; R_3 = CN; % Yield = 28%

- 5 Characterization: ^1H NMR (400 MHz, CDCl_3) δ 9.38 (s, 1H), 9.02 (s, 1H), 8.56 (dd, 2H, J = 4.1, 5.7), 7.53 (m, 3H), 7.45 (d, 2H, J = 7.1), 7.34 (m, 3H), 4.40 (s, 2H), 3.19 (d, 2H, J = 7.1), 2.30 (dp, 1H, J = 6.8, 13.4), 0.93 (s, 3H), 0.91 (s, 3H). HR-MS 406.2049 $[\text{M}+\text{H}]^+$

Entry No.: 2

R_1 = Ph; R = *i*-Bu; R' = methyl(2-naphthyl); R_3 = CN; % Yield = 50%

- 10 Characterization: ^1H NMR (400 MHz, CDCl_3) δ 9.39 (s, 1H), 9.03 (s, 1H), 8.55 (dd, 2H, J = 2.8, 7.0), 7.90 (s, 1H), 7.83 (d, 3H, J = 8.6), 7.51 (m, 6H), 4.56 (s, 2H), 3.17 (d, 2H, J = 7.0), 2.28 (dt, 1H, J = 6.8, 13.5), 0.88 (s, 3H), 0.86 (s, 3H)

Entry No.: 3

R_1 = NH_2 ; R = *i*-Bu; R' = Bn; R_3 = CN; % Yield = 46%

- 15 Characterization: ^1H NMR (400 MHz, CDCl_3) δ 9.06 (s, 1H), 8.90 (s, 1H), 7.34 (m, 5H), 5.71 (s, 2H), 4.32 (s, 2H), 3.05 (d, 2H, J = 7.1), 2.02 (m, 1H), 0.85 (s, 3H), 0.84 (s, 3H). ^{13}C NMR (100 MHz, $(\text{CD}_3)_2\text{SO}$) δ 22.83, 24.30, 31.37, 119.41, 129.56, 131.49, 173.54, 183.67, 207.16.

Entry No.: 4

R_1 = Ph; R = methyl(1-naphthyl); R' = *t*-Bu; R_3 = CN; % Yield = 31%

- 20 Characterization: ^1H NMR (400 MHz, CDCl_3) δ 9.32 (s, 1H), 8.80 (s, 1H), 8.02 (d, J = 8.96 Hz, 1H), 7.80 (d, J = 9.33 Hz, 1H), 7.64 (d, J = 8.27 Hz, 1H), 7.51-7.43 (m, 2H), 7.23-7.17 (m, 1H), 6.80 (d, J = 7.09 Hz, 1H), 5.11 (s, 2H), 2.77 (s, 3H), 1.35 (s, 9H). ^{13}C NMR (100 MHz, CDCl_3) δ 179.5, 171.3, 169.6, 168.61, 164.2, 162.2, 159.7, 134.7, 133.9, 132.3, 128.9, 127.8, 127.3, 126.2, 126.2, 125.9, 125.4, 124.0, 116.4, 104.6, 60.4, 40.1, 39.7, 28.6, 26.4, 21.3, 14.4.
- 25

Entry No.: 5

R_1 = Ph; R = *i*-Bu; R' = Bn; R_3 = COOH; % Yield = 93%

- Characterization: ^1H NMR (400 MHz, $(\text{CD}_3)_2\text{SO}$) δ 9.30 (s, 1H), 9.26 (s, 1H), 8.47 (m, 2H), 7.57 (m, 3H, J = 5.1), 7.31 (d, 4H, J = 4.4), 7.23 (m, 1H, J = 4.3, 8.6), 4.60 (s, 2H), 3.06 (d, 2H, J = 7.1), 2.12 (dt, 1H, J = 6.8, 13.4), 0.79 (s, 3H), 0.78 (s, 3H). ^{13}C NMR (100 MHz, $(\text{CD}_3)_2\text{SO}$) δ 22.9, 28.4, 41.7, 44.2, 122.8, 127.1, 128.8, 129.0, 129.1, 129.5, 129.9, 132.0, 137.4, 138.7, 159.7, 160.2, 163.6, 164.8, 166.7, 169.5, 170.0. HR-MS 425.1989 $[\text{M}+\text{H}]^+$.
- 30

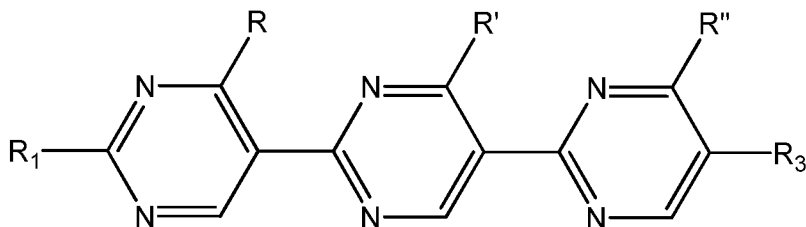
Entry No.: 6

R_1 = Ph; R = *i*-Bu; R' = methyl(2-naphthyl); R_3 = COOH; % Yield = 68%

- 35 Characterization: ^1H NMR (400 MHz, CDCl_3) δ 9.43 (s, 1H), 9.41 (s, 1H), 8.53 (s, 2H), 7.77 (d, 4H, J = 8.2), 7.52 (m, 4H), 7.43 (m, 2H), 4.85 (s, 2H), 3.16 (d, 2H, J = 7.0), 2.25 (dt, 1H, J

- 5 = 6.7, 13.5), 1.25 (s, 1H), 0.84 (s, 3H), 0.83 (s, 3H). ¹³C NMR (100 MHz, CD₃OD) δ 21.5, 28.3, 41.5, 44.1, 104.9, 105.0, 125.4, 125.9, 127.4, 127.5, 127.7, 127.8, 127.8, 128.4, 128.4, 131.0, 131.3, 132.6, 133.9, 135.9, 137.2, 159.0, 159.8, 169.9, 170.5.

PYRIMIDINE TRIMERS



10 Entry No.:

R₁ = Ph; R = *i*-Bu; R' = Bn; R'' = *i*-Pr; R₃ = CN; % Yield = 41%

Characterization: ¹H NMR (400 MHz, CDCl₃) δ 9.56 (s, 1H), 9.37 (s, 1H), 9.03 (s, 1H), 8.55 (dd, 2H, J = 2.9, 6.5), 7.52 (m, 3H), 7.23 (m, 5H), 4.81 (s, 2H), 3.54 (dt, 1H, J = 6.8, 13.5), 3.18 (d, 2H, J = 7.1), 2.29 (dt, 1H, J = 6.7, 13.5), 1.40 (d, 6H, J = 6.8), 0.90 (s, 3H), 0.88 (s, 3H). HR-MS 526.2707 [M+H]⁺

15 Entry No.:

Entry No.: 2

R₁ = Ph; R = methyl(1-naphthyl); R' = Bn; R'' = *i*-Bu; R₃ = CN; % Yield = Not Determined

- Characterization: ¹H NMR (400 MHz, CDCl₃) δ ppm 9.60 (s, 1H), 9.38 (s, 1H), 8.94 (s, 1H), 8.50-8.45 (m, 2H), 8.05-7.99 (m, 1H), 7.86-7.80 (m, 1H), 7.69 (d, J = 8.24 Hz, 1H), 7.50-7.44 (m, 5H), 7.27 (dd, J = 8.11, 7.11 Hz, 1H), 7.08 (ddd, J = 9.57, 5.70, 1.94 Hz, 5H), 6.99 (d, J = 7.02 Hz, 1H), 5.21 (s, 2H), 4.47 (s, 2H), 2.75 (d, J = 7.25 Hz, 2H), 0.85 (d, J = 6.66 Hz, 6H). ¹³C NMR (100 MHz, CDCl₃) ppm 173.77, 169.16, 168.72, 164.42, 164.23, 160.24, 160.04, 138.83, 137.64, 134.66, 134.04, 132.28, 131.26, 129.49, 129.10, 128.88, 128.76, 128.25, 127.81, 127.71, 127.59, 126.86, 126.39, 126.22, 125.95, 125.56, 123.93, 115.00, 107.56, 45.63, 41.91, 40.09, 29.93, 29.00, 22.42.

25 Entry No.:

Entry No.: 3

R₁ = Ph; R = Bn; R' = methyl(1-naphthyl); R'' = *i*-Bu; R₃ = CN; % Yield = Not Determined

- Characterization: ¹H NMR (250 MHz, CDCl₃) δ ppm 9.44 (d, J = 0.52 Hz, 1H), 9.38 (d, J = 0.52 Hz, 1H), 8.92 (d, J = 0.52 Hz, 1H), 8.28-8.23 (m, 2H), 8.06 (dd, J = 5.93, 3.44 Hz, 1H), 7.75 (dd, J = 6.18, 3.33 Hz, 1H), 7.62 (d, J = 8.20 Hz, 1H), 7.40-7.29 (m, 6H), 7.21 (t, J = 7.61 Hz, 1H), 7.08-6.94 (m, 5H), 5.10 (s, 2H), 4.55 (s, 2H), 2.85 (d, J = 7.20 Hz, 2H), 0.93 (d, J = 6.57 Hz, 6H). ¹³C NMR (100 MHz, CDCl₃) ppm 173.74, 169.13, 168.60, 164.50, 164.47, 164.15, 160.23, 160.12, 159.93, 138.12, 137.38, 135.50, 133.97, 132.66, 131.22, 129.30,

30

5 128.86, 128.74, 128.70, 128.59, 128.23, 127.51, 127.16, 127.02, 126.72, 125.98, 125.62, 125.56, 124.76, 115.03, 107.61, 45.77, 42.23, 39.69, 29.93, 29.16, 22.58

Entry No.: 4

R₁ = Ph; R = *i*-Bu; R' = methyl(2-naphthyl); R'' = *i*-Bu; R₃ = CN; % Yield = 40%

Characterization: ¹H NMR (400 MHz, CDCl₃) δ 9.55 (s, 1H), 9.40 (s, 1H), 9.03 (s, 1H), 8.58 – 8.51 (2H), 7.79 – 7.65 (3H), 7.58 (s, 1H), 7.54 – 7.49 (m, 3H), 7.45 – 7.36 (m, 3H), 4.93 (s, 2H), 3.17 (d, *J* = 7.1, 2H), 2.93 (d, *J* = 7.2, 2H), 2.26 (dp, *J* = 13.5, 6.9, 2H), 0.97 (d, *J* = 6.7, 6H), 0.83 (d, *J* = 6.7, 6H). ¹³C NMR (100 MHz, CDCl₃) δ 22.5, 22.6, 28.5, 29.1, 42.5, 44.9, 45.7, 107.7, 114.9, 125.9, 127.6, 127.7, 129.0, 132.0, 132.4, 133.6, 135.6, 160.2, 160.2, 164.3, 169.1, 173.7.

15 Entry No.: 5

R₁ = Ph; R = *i*-Bu; R' = methyl(1-naphthyl); R'' = *i*-Bu; R₃ = CN; % Yield = Not Determined

Characterization: ¹H NMR (250 MHz, CDCl₃) δ ppm 9.53 (s, 1H), 9.21 (s, 1H), 8.86 (s, 1H), 8.48-8.41 (m, 2H), 7.98 (dd, *J* = 6.08, 3.44 Hz, 1H), 7.79 (dd, *J* = 6.16, 3.29 Hz, 1H), 7.65 (d, *J* = 8.22 Hz, 1H), 7.47-7.39 (m, 5H), 7.25-7.19 (m, 1H), 6.95 (d, *J* = 7.00 Hz, 1H), 5.14 (s, 2H), 2.95 (d, *J* = 7.04 Hz, 2H), 2.66 (d, *J* = 7.25 Hz, 2H), 2.18-2.05 (m, 1H), 2.01-1.89 (m, 1H), 0.76 (d, *J* = 6.64 Hz, 6H), 0.71 (d, *J* = 6.65 Hz, 6H). ¹³C NMR (100 MHz, CDCl₃) ppm 173.78, 169.99, 169.08, 164.85, 164.50, 164.14, 160.23, 160.04, 159.52, 137.76, 134.74, 134.05, 132.26, 131.11, 129.07, 128.83, 128.76, 128.54, 127.68, 127.54, 126.70, 126.36, 125.96, 125.54, 123.95, 114.99, 107.54, 45.61, 44.50, 40.06, 29.93, 29.00, 28.43, 22.69, 22.40.

The isolated yields reported in the tables above are for isolation conditions that favor the purity of the product over getting all of the product isolated, because the starting materials for these products are inexpensive or easily prepared. The majority of these compounds were single spots on TLC after precipitation from the reaction solvents.

30 Several of the dimeric 2,5-pyrimidines reported above have crystallized and one of them has been submitted for X-ray structure determination. The ORTEP diagram is shown in Figure 11.

C2. Molecular modeling studies comparing various trimeric-2,5-pyrimidines as α-helix mimics and as potential inhibitors of Mdm2-p53 and Bcl-xL-Bad interaction inhibitors: The GLIDE program (Grid Based Ligand Docking from Energetics, available from Schrödinger, LLC) was employed to dock trimeric-2,5-pyrimidines and nutlin-3 (a known MDM2 inhibitor with an IC₅₀ of 0.09 μM to MDM2 (Vassilev, 2004; Tovar, 2006; Coll-Mulet, 2006; Xia, 2008). The MDM2 structure employed was obtained from the Protein Databank (www.rcsb.org, PDB ID: 1RV1,

5 MDM2 co-crystallized with nutlin-2; 2.3 Å resolution). The XP (extra precision) version of GLIDE was used and the side chains for the trimeric 2,5-pyrimidine shown below, molecule A, Figure 12, were selected based on the p53 α -helical domain that interacts with MDM2. Although the average unsigned error in GLIDE XP docking scores is approximately 1.8 kcal/mol (Friesner, 2006), it is noteworthy that the docking score obtained for nutlin-3 and the
10 selected trimeric-2,5-pyrimidine were comparable (-8.98 kcal/mol for nutlin-3 vs. -9.29 kcal/mol for molecule A shown below). Note that the docked structure of nutlin-3 to MDM2 was entirely consistent with the observed X-ray structure of nutlin-2 complexed with MDM2. Figure 12 shows molecule A in part A docked to MDM2 in part B; part C shows the p53 N-terminus interaction with MDM2. Note that in our docking pose, the benzyl and methyl(3-indolyl) side chains in part B bind to the same hydrophobic pockets in MDM2 as the Phe and Trp side chains in p53, whereas the isobutyl side chain binds slightly differently, in a shallow hydrophobic pocket near the deeper hydrophobic pocket in MDM2 to which the p53 Ile side chain binds. An Ile-like side chain might be better for this binding site than the Leu-like side chain.

20 The GLIDE program was also used to dock molecule D in Figure 13 to Bcl-xL. The Bcl-xL structure employed was obtained from the PDB (www.rcsb.org, PDB ID: 2YZJ; Bcl-xL complexed with ABT-737; 2.2 Å resolution). ABT-737 binds with subnanomolar affinity to Bcl-xL (Oltersdorf, 2005). ABT-737 antagonizes Bcl-2 family proteins in their interaction with pro-apoptotic proteins and induces regression of solid tumors (Wendt, 2006; Bruncko, 2007).

25 The side chains selected for molecule D in Figure 13 were ones that are consistent with the amino acid residues involved in binding of the BH3 α -helical domain of Bad to Bcl-xL. The GLIDE XP docking scores obtained for molecule D in Figure 13 and ABT 737 were comparable (-6.94 kcal/mol and -6.82 kcal/mol respectively). In Figure 13, part E, molecule D is docked to Bcl-xL and in part F, both molecule D (yellow carbon atoms) and ABT-737 (cyan
30 carbon atoms) are shown docked to Bcl-xL.

C3. QikProp calculations: Structurally analogous Hamilton terphenylene, Example 8-type, and Example 9-type scaffolds with the same side chains were compared and the log $P_{\text{octanol/water}}$ values ranged from very hydrophobic for the Hamilton terphenylene scaffold to barely outside the desirable range for the Example 8-type scaffold and to well within the
35 desirable range for the Example 9-type scaffold. The actual log $P_{\text{octanol/water}}$ values and the structures that were compared are shown in Figure 14. There were additional differences that show that the trimeric 2,5-pyrimidines are more "drug-like" than the Hamilton terphenylene scaffolds.

EXAMPLE 10 - A FACILE ITERATIVE SYNTHESIS OF 2,5-TERPYRIMIDINYLENE
40 LIBRARIES AS NON-PEPTIDIC α -HELICAL MIMICS

5 We have developed a facile iterative synthesis of 2,5-terpyrimidinylenes as structurally analogous α -helix mimics. Figure 15 shows an overlay of octa-alanine in an idealized α -helical conformation with its *ith*, *ith*+4, and *ith*+7 methyl groups highlighted as spheres and a 4,4',4''-trimethyl-2,5-terpyrimidinylene with its methyl groups highlighted as adjacent spheres.

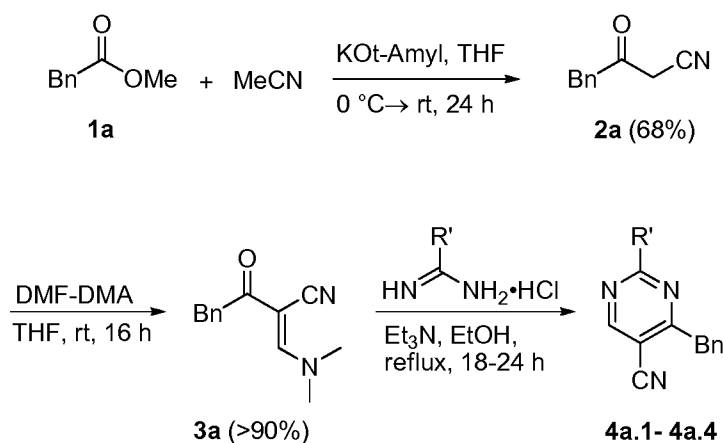
As seen with 1,4-terphenylene scaffolds [Ernst, JT, et al., *Angew. Chem., Int. Ed.* (2002) 41: 278-281; Kutzki, O, et al., *J. Am. Chem. Soc.* (2002) 124: 11838-11839], there is good
10 overlap of these positions both in orientation and distance. Residues at the *ith*+4 position are one turn plus 40° and *ith*+7 residues are 20° less than two turns of an α -helix from the *ith* position. The 2,5-terpyrimidinylene scaffold essentially replaces the phenyl making the synthesis of pyrimidine-based libraries much easier because the pyrimidine synthetic
15 chemistry is very convergent and amenable to an iterative synthetic approach.

In addition, the pyrimidine for phenyl replacement makes the resulting analogous pyrimidine scaffold much more "drug-like" according to QikProp calculations summarized in Figure 16. Structurally analogous 1,4-terphenylene and 2,5-terpyrimidinylene scaffolds show the
20 calculated log $P_{\text{octanol/water}}$ values ranged from quite hydrophobic for the 1,4-terphenylene scaffolds [Yin, H, et al., *J. Am. Chem. Soc.* (2005) 127: 10191-10196] to within the desirable range for drug-like characteristics for the analogous 2,5-terpyrimidinylene scaffold.

Pyrimidine monomers 4a.1-4a.4 were obtained in a few steps through the condensation of commercially available amidines with readily prepared α,β -unsaturated α -cyanoketones 3 (Scheme 1, below). For instance, methyl phenylacetate 1a was reacted with acetonitrile in
25 anhydrous THF in the presence of KO-*t*-amyl to obtain β -ketonitrile 2a [Yaohui, Jet al., *Org. Lett.* (2006) 8: 1161-1163]. These reactions were also carried out in presence of other bases, including NaOMe [Sorgner, K, et al., *U.S. Pat. Pub. No.* 2007/0142661], LDA, and KO-*t*-But (data not shown). While the desired products were also afforded from reactions with these bases, the isolation and purification were tedious resulting in lower yields. KO-*t*-amyl afforded
30 the products in good yields with much easier purification conditions.

Treatment of compound 2a with *N,N*-dimethylformamide dimethyl acetal (DMF-DMA) in THF gave 3a in excellent yield [Reuman, M, et al., *J. Org. Chem.* (2008) 73: 1121-1123]. The efficiency of this step yielded several α,β -unsaturated α -cyanoketones bearing hydrophobic
35 alkyl, aryl, and heteroaromatic groups for introducing diversity in the 4-position of pyrimidines. As shown in Scheme 1 (below), monomers 4a.3 and 4a.4 were isolated in higher yields and no further purification was required since these compounds precipitated from the reaction mixtures, whereas 4a.1 and 4a.2 required additional purification. The excellent yields and purities obtained when R'= Ph, bode well for the subsequent iterative synthesis of the dimeric and trimeric 2,5-pyrimidines.

40 Scheme 1. Representative synthesis of pyrimidines 4a.1-4a.4:

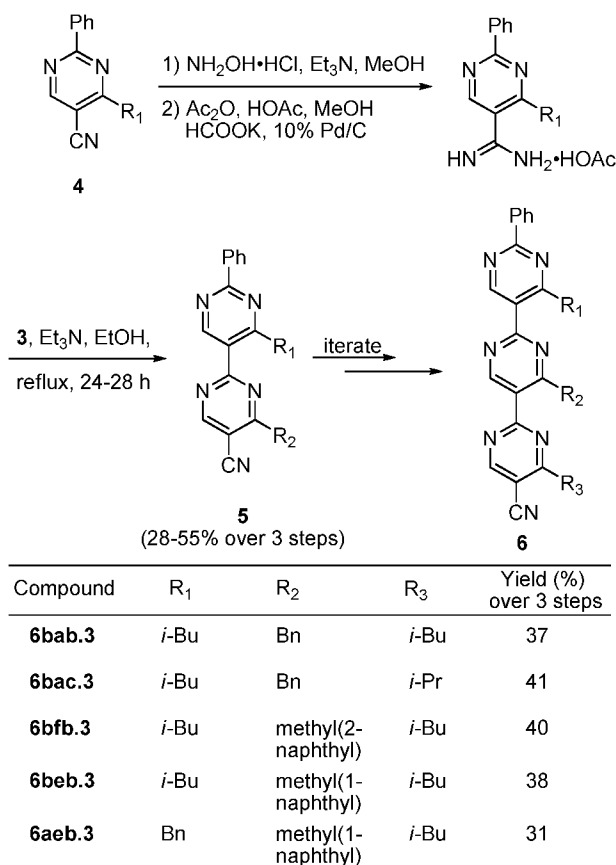


Compound	R'	Yield (%)
4a.1	H	39
4a.2	Me	63
4a.3	Ph	91
4a.4	NH ₂	83

5

Compounds 5 and 6 were synthesized via conversion of the 5- or 5'-cyano group to a 5- or 5'-carboxamidinium salt (Scheme 2). Several methods for this conversion have been reported, [Hill, MD, et al., *Chem.--Eur. J.* (2008) 14: 6836-6844; Dunn, PJ, et al., *Compr. Org. Funct. Group Transform.* (2005) II: 655-699] such as the Pinner [Balo, C, et al., *Chem. Pharm. Bull.* (2007) 55: 372-375], the thio-Pinner [Lange, UEW, et al., *Tetrahedron Lett.* (1999) 40: 7067-7070], or treatment with Na or LiHMDS followed by aqueous hydrolysis [Bruning, J. *Tetrahedron Lett.* (1997) 38: 3187-3188], but none of these alternatives were superior to the two-step hydroxylamine route [Judkins, BD, et al., *Synth. Commun.* (1996) 26: 4351-4367; Nadrah, K, et al., *Synlett* (2007) 8: 1257-1258].

15 Scheme 2. Synthesis of dimeric and trimeric 2,5-pyrimidines:



5

The literature on nitrile to amidine conversion suggests that this can be attributed to the *ortho* substitution on the pyrimidine ring [von Angerer, S. *Science of Synthesis* (2004) 16: 379-572]. An excess hydroxylamine followed by a *in situ* reduction of the resulting amidoxime was the best route to obtain the intermediate amidine salts. The *N*-hydroxylamine mediated transformation of *ortho*-substituted aryl nitrile groups to arylcarboxamidines has been reported [Basso, A, et al., *Eur. J. Org. Chem.* (2000) 23: 3887-3891], but has not been used previously to prepare oligo-2,5-pyrimidines. The resulting intermediate amidine salts were reacted with an α,β -unsaturated α -cyanoketone to yield dimers 5.

Both dimers and trimers are indicated for isolated products. The R₁, R₂, and R₃ groups of the 2,5-terpyrimidinylene scaffold were selected to mimic hydrophobic groups found to play important roles in binding the terphenylene compounds [Yin, H, et al., *J. Am. Chem. Soc.* (2005) 127: 10191-10196].

The ORTEP diagram of the 4'-benzyl-4-(1-methylethyl)-4''-(2-methylpropyl)-2''-phenyl-2,5',2',5''-terpyrimidinylene-5-carbonitrile (6bac.3) crystal structure is shown in Figure 17. This conformation places the three hydrophobic groups on the same side of the long axis of

- 5 the molecule which is the conformer needed to mimic the *ith*, *ith*+4, and *ith*+7 residues of an α -helix.

Example 10 – Materials and Methods

Starting materials, organic and inorganic reagents (ACS grade), and solvents were obtained from commercial sources and used as received unless otherwise noted. Moisture- and air-sensitive reactions were carried out under an atmosphere of argon.

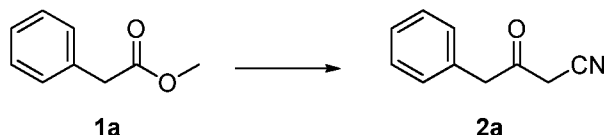
Thin layer chromatography (TLC) was performed on glass plates precoated with 0.25 mm thickness of silica gel (60 F-254) with fluorescent indicator (EMD or Whatman). Column chromatographic purification was performed using silica gel 60 Å, #70-230 mesh (Selecto Scientific). Automated flash chromatography was performed in a FlashMaster II system (Argonaut-Biotage) using Biotage silica cartridges.

^1H NMR and ^{13}C NMR spectra were obtained using a 400 MHz Varian Mercury plus instrument at 25 °C in chloroform-*d* (CDCl_3), unless otherwise indicated. Chemical shifts (δ) are reported in parts per million (ppm) relative to internal tetramethylsilane (TMS) or chloroform (δ 7.26) for ^1H NMR and chloroform (δ 77.0) for ^{13}C NMR. Multiplicity is expressed as (s = singlet, br s = broad singlet, d = doublet, t = triplet, q = quartet, or m = multiplet) and the values of coupling constants (*J*) are given in Hertz (Hz). High Resolution Mass Spectrometry (HRMS) spectra were carried out on an Agilent 1100 Series in the ESI-TOF mode. Microwave reactions were performed in a closed vessel in a Biotage Initiator I microwave reactor. Melting points (uncorrected) were determined using a Mel-Temp II®, Laboratory Devices, MA, USA.

All compounds are labeled based on the substituents as follows:

a R = Bn	f R = methyl(2-naphthyl)	1 R' = H
b R = <i>i</i> -Bu	g R = Me	2 R' = Me
c R = <i>i</i> -Pr	h R = N-Cbz 3-methyl indolyl	3 R' = Ph
d R = 2-(cyclohexyl)ethyl	i R = <i>t</i> -Bu	4 R' = NH ₂
e R = methyl(1-naphthyl)	j R = Ph	

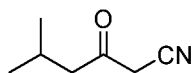
Representative Procedure for the Synthesis of β -ketonitriles 2:



- 30 3-oxo-4-phenylbutanenitrile (2a): A mixture of potassium *t*-pentylate (~1.7 M in toluene, 106 mmol, 62.3 mL) and anhydrous THF (20 mL) was cooled at 0 °C in an ice-bath under an argon atmosphere. Then anhydrous acetonitrile (106 mmol, 6 mL) and methyl ester 1a (71 mmol, 10 mL) were added simultaneously to the cold solution. The reaction mixture was

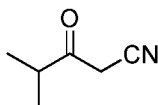
5 allowed to warm to room temperature and stirred under an argon atmosphere for 22 h. A precipitate was formed within a few minutes (3-5 min.) of stirring and the reaction mixture remained cloudy until completion. The reaction was monitored by TLC and it was stopped when the TLC indicated the consumption of the ester (1a). The mixture was filtered and the filtered cake was washed thoroughly with hexanes (80 mL). Then, the filtered residue was transferred to a separatory funnel and it was acidified to pH= ~2-3 with saturated aq. KHSO₄ solution (400 mL). An equal volume amount of DCM (400 mL) was used to extract the desired compound (2a). The organic layer was washed with brine (100 mL), dried over Na₂SO₄, and concentrated under reduced pressure to yield 2a in 68% yield, as a pale yellow oil. ¹H NMR (400 MHz, CDCl₃) δ 3.46 (s, 2H), 3.86 (s, 2H), 7.21–7.40 (m, 5H). ¹³C NMR (100 MHz, CDCl₃) δ 31.1, 49.2, 113.5, 127.9, 129.3, 129.4, 131.9, 195.1.

5-methyl-3-oxohexanenitrile (2b):



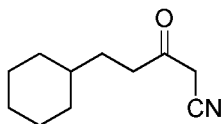
Compound 2b was prepared by the same method described for 2a. Isolated yield: 73%, yellow oil. ¹H NMR (400 MHz, CDCl₃) δ 0.95 (d, *J* = 0.85 Hz, 3H), 0.97 (d, *J* = 0.85 Hz, 3H), 3.43 (s, 2H), 2.11-2.25 (m, 1H), 2.49 (d, *J* = 6.88 Hz, 2H). ¹³C NMR (100 MHz, CDCl₃) δ 22.5, 24.7, 32.6, 51.1, 113.9, 197.2.

4-methyl-3-oxopentanenitrile (2c):



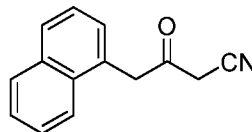
Compound 2c was prepared following the procedure described for 2a. Isolated yield: 64%, colorless oil. Spectral data is in agreement with literature (*Organic Lett.* 2006, 8(6), 1161-1163).

5-cyclohexyl-3-oxopentanenitrile (2d):



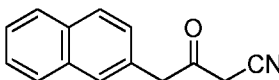
Compound 2d was prepared following the procedure described for 2a. Isolated yield: 79% yield, pale orange solid, m.p. decomposed at 152 °C. ¹H NMR (400 MHz, CDCl₃) δ 0.84-1.86 (m, 13H) 2.60 (s, 2H, *J*=7.2 Hz), 3.46 (s, 2H). ¹³C NMR (100 MHz, CDCl₃) δ 26.4, 26.6, 30.8, 37.2, 32.1, 33.2, 40.5, 114.1, 198.2. IR (solid, cm⁻¹) 3196.4, 2851.2, 1701.9, 1469.5, 1427.1, 1374.0, 1326.8, 1281.5, 1193.7, 1163.8, 1116.6, 1056.8, 962.3.

5 4-(naphthalen-1-yl)-3-oxobutanenitrile (2e):



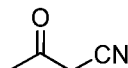
Compound 2e was prepared following the procedure described for 2a. Isolated yield: 64% yield, yellow solid. ^1H NMR (400 MHz, CDCl_3) δ 3.41 (s, 2H), 4.31 (s, 2H), 7.43 – 7.66 (m, 4H), 7.81 – 7.98 (m, 3H). ^{13}C NMR (100 MHz, CDCl_3) δ 31.2, 47.8, 113.7, 123.3, 125.9,
10 126.6, 127.5, 128.7, 129.0, 129.3, 129.4, 132.0, 134.3, 196.0.

4-(naphthalen-2-yl)-3-oxobutanenitrile (2f):



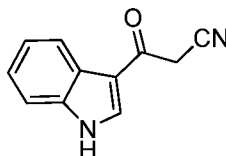
Compound 2f was prepared following the procedure described for 2a. Isolated yield: 71% yield, yellow solid. ^1H NMR (400 MHz, CDCl_3) δ 3.51 (s, 2H), 4.06 (s, 2H), 7.34 (dd, J = 1.8, 8.4, 1H), 7.51 – 7.57 (m, 2H), 7.74 (s, 1H), 7.84 – 7.93 (m, 3H). ^{13}C NMR (100 MHz, CDCl_3)
15 δ 31.4, 49.6, 113.8, 126.7, 126.9, 127.1, 127.9, 128.0, 128.8, 129.4, 129.5, 132.9, 133.7, 195.5.

3-oxobutanenitrile (2g):



20 Compound 2g was prepared following the procedure described for 2a. Isolated yield: 81%, colorless oil (compound decomposes on long standing at room temperature, thus it was used immediately without further purification). Spectral data is in agreement with literature (*Molecules* 2006, 11(5), 371-376).

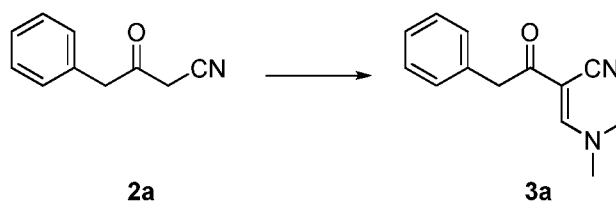
3-(1H-indol-3-yl)-3-oxopropanenitrile (2h):



25

Compound 2h was prepared following the procedure described by Radwan, M. A. A. *et. al.* Isolated yield: 95%, pale orange solid, m.p. 238 °C (lit.= 240 °C). Spectral data is in agreement with literature (*Bioorganic Med. Chem. Lett.* 2007, 15, 1206).

Representative Procedure for the Synthesis of α,β -Unsaturated α -Cyanoketones 3



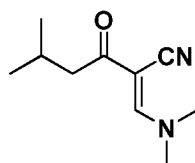
5

(*E*)-2-((dimethylamino)methylene)-3-oxo-4-phenylbutanenitrile (3a): β -ketonitrile 2a (0.75 mmol, 120 mg) was dissolved in dry THF (3 mL) under an argon atmosphere. Then *N,N*-dimethylformamide dimethyl acetal (0.98 mmol, 0.16 mL) was added and the mixture was stirred at room temperature. Progress of the reaction was monitored by TLC until complete consumption of starting material 2a was observed (16 h). The solvent was evaporated under reduced pressure and the desired compound was obtained as an *E*>>*Z* isomeric mixture. Isolated yield: >90 %, colorless needle-like crystals, m.p. 134-138 °C. ^1H NMR (400 MHz, CDCl_3) δ 3.20 (s, 3H), 3.38 (s, 3H), 3.95 (s, 2H), 7.21–7.34 (m, 5H), 7.80 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 9.0, 46.5, 48.2, 120.5, 126.9, 128.7, 129.8, 129.8, 129.9, 135.0, 158.0, 192.9. HRMS (ESI) calcd. for $\text{C}_{13}\text{H}_{14}\text{N}_2\text{O}$ $[\text{M} + \text{H}]^+$ 215.1184, found 215.1194.

15

Note: If a precipitate forms during the reaction, the mixture is cooled to 0 °C and then filtered and rinsed with cold THF to isolate the desired compound.

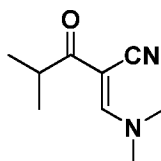
(*E*)-2-((dimethylamino)methylene)-5-methyl-3-oxohexanenitrile (3b):



Compound 3b was prepared following the procedure described for 3a. Isolated yield: >90%, pale yellow solid, m.p. 43-45 °C. ^1H NMR (400 MHz, CDCl_3) δ 0.95 (d, J = 1.48 Hz, 3H), 0.96 (d, J = 1.47 Hz, 3H), 2.13-2.25 (m, 1H), 2.54 (dd, J = 7.03 Hz, 2H), 3.24 (s, 3H), 3.40 (s, 3H), 7.82 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 22.8, 25.8, 39.0, 48.1, 48.8, 80.8, 120.6, 157.6, 195.4. HRMS (ESI) calcd. for $\text{C}_{10}\text{H}_{16}\text{N}_2\text{O}$ $[\text{M} + \text{H}]^+$ 181.1341, found 181.1333.

25

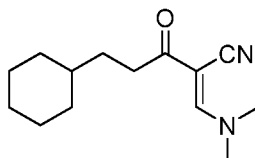
(*E*)-2-((dimethylamino)methylene)-4-methyl-3-oxopentanenitrile (3c):



Compound 3c was prepared following the procedure described for 3a. Isolated yield: >90%, hygroscopic sticky yellow solid, m.p. 40-42 °C. ^1H NMR (400 MHz, CDCl_3) δ 1.12 (d, J = 2.26 Hz, 3H), 1.14 (d, J = 2.26 Hz, 3H), 3.24 (s, 3H), 3.41 (s, 3H), 7.84 (s, 1H). ^{13}C NMR (100

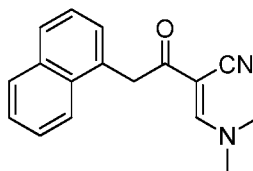
- 5 MHz, CDCl_3) δ 19.1, 37.1, 39.0, 48.1, 79.4, 120.5, 158.0, 199.8. HRMS (ESI) calcd. for $\text{C}_9\text{H}_{14}\text{N}_2\text{O}$ $[\text{M} + \text{H}]^+$ 167.1184, found 167.1184.

5-cyclohexyl-2-((dimethylamino)methylene)-3-oxopentanenitrile (3d):



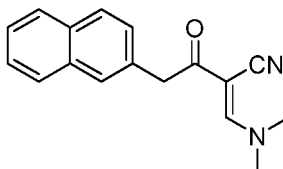
- 10 Compound 3d was prepared following the procedure described for 3a. Isolated yield: >90%, orange-brown solid, m.p. 92-94 °C. ^1H NMR (400 MHz, CDCl_3) δ 0.87-1.72 (m, 13H), 2.65 (t, 2H, $J = 8$ Hz), 3.23 (s, 3H), 3.39 (s, 3H), 7.81 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 14.1, 20.1, 26.2, 61.3, 63.1, 120.4, 157.3, 196.1. IR (solid, cm^{-1}) 3173.3, 2848.4, 2341.2, 1700.9, 1597.7, 1271.9. HRMS (ESI) calcd. for $\text{C}_{14}\text{H}_{22}\text{N}_2\text{O}$ $[\text{M} + \text{H}]^+$ 235.181, found 235.1821.

2-((dimethylamino)methylene)-4-(naphthalen-1-yl)-3-oxobutanenitrile (3e):



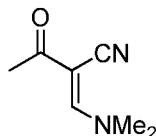
- 15 Compound 3e was prepared following the procedure described for 3a. Isolated yield: 95%, yellow solid, m.p. 121-122°C. ^1H NMR (400 MHz, CDCl_3) δ 3.17 (s, 3H), 3.41 (s, 3H), 4.43 (s, 2H), 7.40 – 7.54 (m, 4H), 7.77 (d, $J = 7.9$, 1H), 7.80 – 7.86 (m, 2H), 7.99 (d, $J = 8.2$, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 39.1, 44.1, 48.1, 80.1, 120.6, 124.6, 125.7, 125.8, 126.3, 127.9, 128.7, 128.8, 131.8, 132.6, 134.4, 158.1, 192.6. HRMS (ESI) calcd. for $\text{C}_{17}\text{H}_{16}\text{N}_2\text{O}$ $[\text{M} + \text{H}]^+$ 265.1335, found 265.1313.
- 20

2-((dimethylamino)methylene)-4-(naphthalen-2-yl)-3-oxobutanenitrile (3f):



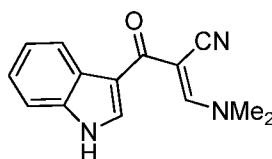
- 25 Compound 3f was prepared following the procedure described for 3a. Isolated yield: 95%, yellow solid. ^1H NMR (400 MHz, CDCl_3) δ 3.20 (s, 3H), 3.39 (s, 3H), 4.12 (s, 2H), 7.40 – 7.50 (m, 3H), 7.77 – 7.84 (m, 5H). ^{13}C NMR (100 MHz, CDCl_3) δ 39.0, 46.7, 48.2, 80.1, 120.5, 125.8, 126.2, 127.8, 128.0, 128.0, 128.3, 128.5, 132.6, 133.8, 158.0, 192.9. HRMS (ESI) calcd. for $\text{C}_{17}\text{H}_{16}\text{N}_2\text{O}$ $[\text{M} + \text{H}]^+$ 265.1335, found 265.1333.

5 2-((dimethylamino)methylene)-3-oxobutanenitrile (3g):



Compound 3g was prepared following the procedure described for 3a. Isolated yield: 80%, dark-brown waxy solid with a strong stench. ^1H NMR (400 MHz, CDCl_3) δ 2.34 (s, 3H), 3.24 (s, 3H), 3.39 (s, 3H), 7.79 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 27.7, 39.1, 48.4, 80.3, 120.6, 157.7, 193.4. HRMS (ESI) calcd. for $\text{C}_7\text{H}_{10}\text{N}_2\text{O}$ $[\text{M} + \text{H}]^+$ 139.0871, found 139.0853.

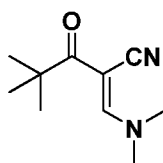
3-(dimethylamino)-2-(1H-indole-3-carbonyl)acrylonitrile (3h):



Compound 3h was prepared following the procedure described for 3a. Isolated yield: 96%, bright yellow solid, m.p. decomposed at 162 °C (Lit.= 160 °C). ^1H NMR (400 MHz, CD_3SOCD_3) δ 3.25(s, 3H), 3.34(s, 3H), 7.08–7.23 (m, 2H), 7.45 (d, $J = 7.8$, 1H), 7.98, 8.10, 8.11 (d, $J = 7.6$, 1H), 8.26, 11.74. ^{13}C NMR (100 MHz, CD_3SOCD_3) δ 40.8, 48.1, 78.3, 112.6, 115.4, 121.8, 122.4, 123.2, 127.5, 131.8, 136.5, 159.4, 182.4.

Spectral data is in agreement with literature (*Bioorganic Med. Chem.* 2007, 15, 1206).

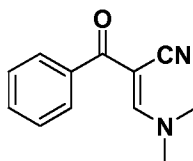
(E)-2-((dimethylamino)methylene)-4,4-dimethyl-3-oxopentanenitrile (3i):



20

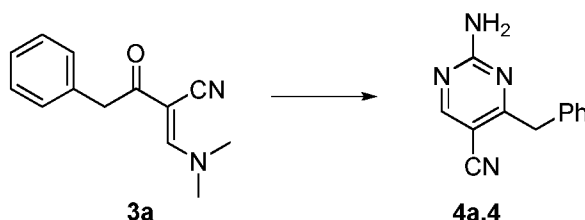
Compound 3i was prepared following the procedure described for 3a. Isolated yield: >90%, white solid, m.p. 48-49 °C. ^1H NMR (400 MHz, CDCl_3) δ 1.32 (s, 9H), 3.23 (s, 2H), 3.42 (s, 2H), 7.92 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 27.0, 39.1, 43.8, 48.5, 121.5, 160.5, 200.4. HRMS (ESI) calcd. for $\text{C}_{10}\text{H}_{16}\text{N}_2\text{O}$ $[\text{M} + \text{H}]^+$ 181.1341, found 181.1340.

25 (E)-2-benzoyl-3-(dimethylamino)acrylonitrile (3j):



- 5 Compound 3j was prepared following the procedure described for 3a. Isolated yield: >90%, white solid. ^1H NMR (400 MHz, CDCl_3) δ 3.29 (s, 3H), 3.49 (s, 3H), 7.40-7.59 (m, 3H), 7.74-7.81 (m, 2H), 7.95 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 39.2, 48.5, 120.5, 128.3, 128.4, 131.7, 138.7, 159.6, 190.5. HRMS (ESI) calcd. for $\text{C}_{12}\text{H}_{12}\text{N}_2\text{O}$ $[\text{M} + \text{H}]^+$ 201.1028, found 201.1019.

10 Representative Procedure for the Synthesis of Pyrimidines 4:



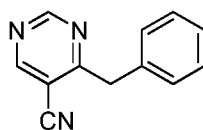
- Method A: A mixture of compound 3a (15.17 mmol, 3.25 g) and guanidine hydrochloride (30.34 mmol, 2.89 g) in ethanol (absolute, 200 proof, 10 mL) was stirred under reflux until the TLC indicated completion of the reaction. The mixture was brought to room temperature and the precipitate that formed was filtered by vacuum and rinsed with ice cold ethanol (5 mL x 3). Compound 4a.4 was isolated as colorless crystals in 83% yield. Compounds 4a.1 and 4a.2 required additional purification since these did not precipitate from the reaction mixtures.

- Method B: Microwave-assisted reactions were also done in the presence of base (e.g. Et_3N or NaOEt) to obtain the desired pyrimidines. For example, to a mixture of nitrile 3b (0.93 mmol, 200 mg) and guanidine hydrochloride (1.87 mmol, 0.17 g) in ethanol (5 mL) was added sodium ethoxide (0.93 mmol, 63.49 mg) suspended in ethanol (1 mL). The reaction mixture was placed in a microwave reactor for 40 min. at 120 $^\circ\text{C}$. A colorless crystal-like precipitate formed in the solution upon cooling to room temperature. The precipitate was filtered and rinsed with ice cold ethanol (2 x 3 mL) to isolate compound 4b.4 as needle-like crystals.

- 25 2-amino-4-benzylpyrimidine-5-carbonitrile (4a.4): Compound 4a.4 was prepared following method B. It was purified by chromatography in silica cartridge performed by FlashMaster II purification system (hexanes:ethyl acetate, 6:4), yield: 78%, white solid; recrystallized from ethanol to obtain colorless crystals, m.p. 129-132 $^\circ\text{C}$. ^1H NMR (400 MHz, CDCl_3) δ 4.10 (s, 2H), 5.58 (br s, 2H), 7.24-7.38 (m, 5H), 8.45 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 42.9, 97.8, 116.4, 127.5, 129.0, 129.5, 136.0, 162.4, 162.8, 173.5. HRMS (ESI) calcd. for $\text{C}_{12}\text{H}_{10}\text{N}_4$ $[\text{M} + \text{H}]^+$ 211.0984, found 211.0972.

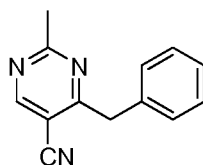
4-benzylpyrimidine-5-carbonitrile (4a.1):

5



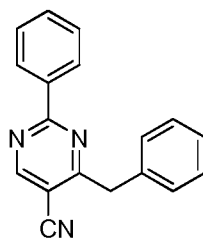
Compound 4a.1 was prepared following method A using commercially available formamidine hydrochloride. It was purified by column chromatography on silica gel (hexanes:ethyl acetate, 8:2), yield: 39%, pale yellow thick oil. ^1H NMR (400 MHz, CDCl_3) δ 4.25 (s, 2H), 7.17 – 7.26 (m, 2H), 7.26 – 7.33 (m, 3H), 8.83 (s, 1H), 9.20 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 43.1, 109.2, 114.8, 127.8, 129.2, 129.5, 135.7, 160.4, 160.5, 172.0. HRMS (ESI) calcd. for $\text{C}_{12}\text{H}_9\text{N}_3$ [M + H] $^+$ 196.0875, found 196.0868.

4-benzyl-2-methylpyrimidine-5-carbonitrile (4a.2):



Compound 4a.2 was prepared following method A using commercially available acetamidine hydrochloride. It was purified by column chromatography on silica gel (hexanes:ethyl acetate, 8:2), yield: 63%, colorless thick oil. ^1H NMR (400 MHz, CDCl_3) δ 2.79 (s, 3H), 4.26 (s, 2H), 7.24 – 7.41 (m, 5H), 8.78 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 26.9, 43.2, 105.8, 115.4, 127.6, 129.1, 129.4, 136.0, 160.4, 171.3, 171.7. HRMS (ESI) calcd. for $\text{C}_{13}\text{H}_{11}\text{N}_3$ [M + H] $^+$ 210.1031, found 210.1022.

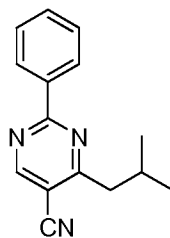
4-benzyl-2-phenylpyrimidine-5-carbonitrile (4a.3):



Compound 4a.3 prepared following method A using commercially available benzamidine hydrochloride. Isolated yield: 91%, white solid, m.p. 138-141 °C. ^1H NMR (400 MHz, CDCl_3) δ 4.37 (s, 2H), 7.27 – 7.30 (m, 1H), 7.35 (dd, J = 10.1, 4.6, 2H), 7.45 – 7.59 (m, 5H), 8.49 – 8.54 (m, 2H), 8.92 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 43.34, 105.94, 115.69, 127.59, 129.03, 129.08, 129.46, 129.53, 132.61, 136.12, 136.17, 160.82, 166.02, 171.85. HRMS (ESI) calcd. for $\text{C}_{18}\text{H}_{13}\text{N}_3$ [M + H] $^+$ 272.1188, found 272.1196.

4-isobutyl-2-phenylpyrimidine-5-carbonitrile (4b.3):

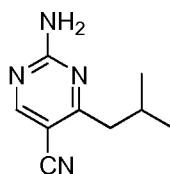
5



10

4b.3 was prepared following method A. Isolated yield: 70%, colorless crystals, m.p. 68-69 °C. ^1H NMR (400 MHz, CDCl_3) δ 1.05 (d, J = 6.7, 6H), 2.39 (m, 1H), 2.93 (d, J = 7.2, 2H), 7.49 – 7.59 (m, 3H), 8.50 – 8.53 (m, 2H), 8.93 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 22.57, 28.88, 45.71, 106.71, 115.80, 129.00, 129.38, 132.44, 136.36, 160.37, 165.72, 173.29. HRMS (ESI) calcd. for $\text{C}_{15}\text{H}_{15}\text{N}_3$ $[\text{M} + \text{H}]^+$ 238.1344, found 238.1337.

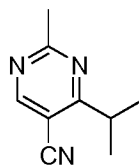
2-amino-4-isobutylpyrimidine-5-carbonitrile (4b.4):



15

4b.4 was prepared following method B. Isolated yield: 75%, white solid, m.p. 175-178 °C. ^1H NMR (400 MHz, CDCl_3) δ 0.98 (d, J = 6.7, 6H), 2.18 (m, 1H), 2.66 (d, J = 7.3, 2H), 5.72 (br s, 2H), 8.45 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 22.56, 28.86, 45.51, 98.44, 116.62, 162.06, 162.81, 174.96. HRMS (ESI) calcd. for $\text{C}_9\text{H}_{12}\text{N}_4$ $[\text{M} + \text{H}]^+$ 177.1140, found 177.1136.

4-isopropyl-2-methylpyrimidine-5-carbonitrile (4c.2):

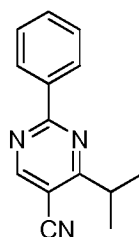


20

4c.2 was prepared following method B. It was purified by column chromatography on silica gel (hexanes:ethyl acetate, 7:3), yield: 90%, pale yellow thick oil. ^1H NMR (400 MHz, CDCl_3) δ 1.30 (d, 6H, J =5.8 Hz), 2.75 (s, 3H), 3.38 (m, 1H), 8.74 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 21.2, 29.9, 35.0, 105.0, 115.3, 160.2, 171.1, 178.2. HRMS (ESI) calcd. for $\text{C}_9\text{H}_{11}\text{N}_3$ $[\text{M} + \text{H}]^+$ 161.0953, found 161.0953.

4-isopropyl-2-phenylpyrimidine-5-carbonitrile (4c.3):

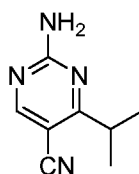
5



10

4c.3 was prepared following method B. Isolated yield: 87%, white solid, m.p. 168-170 °C. ^1H NMR (400 MHz, CDCl_3) δ 1.43 (d, $J = 3.4$, 6H), 3.48 (m, 3H), 7.53 (m, 3H), 8.53 (d, $J = 3.6$, 2H), 8.90 (s, 1H). ^{13}C NMR (400 MHz, CDCl_3) δ 21.4, 35.2, 105.2, 115.6, 128.9, 129.4, 132.4, 136.5, 160.6, 165.9, 178.2. HRMS (ESI) calcd. for $\text{C}_{14}\text{H}_{13}\text{N}_3$ $[\text{M} + \text{H}]^+$ 224.1187, found 224.1178.

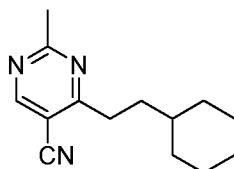
2-amino-4-isopropylpyrimidine-5-carbonitrile (4c.4):



15

4c.4 was prepared following method B. It was purified by flash column chromatography on silica gel (hexanes:ethyl acetate, 1:1), yield: 80%, off-white solid, m.p. 158-159 °C. ^1H NMR (400 MHz, CD_3COCD_3) δ 1.13 (d, $J = 6$, 6H), 3.11 (m, 1H), 6.95 (br s, 2H), 8.41 (s, 1H). ^{13}C NMR (400 MHz, CD_3COCD_3) δ 20.4, 34.3, 95.2, 116.6, 162.4, 164.2, 178.9. HRMS (ESI) calcd. for $\text{C}_8\text{H}_{10}\text{N}_4$ $[\text{M} + \text{H}]^+$ 163.0983, found 163.0981.

4-(2-cyclohexylethyl)-2-methylpyrimidine-5-carbonitrile (4d.2):



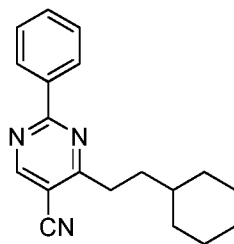
20

4d.2 was prepared following method B. It was purified by column chromatography on silica gel (hexanes:ethyl acetate, 7:3), yield: 70%, colorless thick oil. ^1H NMR (400 MHz, CDCl_3) δ 0.85-1.76, (m, 13H), 2.73 (s, 3H), 2.91 (m, 2H), 8.73 (s, 1H). ^{13}C NMR (400 MHz, CDCl_3) δ 26.4, 26.7, 26.8, 33.2, 34.7, 36.4, 37.8, 105.3, 115.2, 159.9, 170.8, 174.4. HRMS (ESI) calcd. for $\text{C}_{14}\text{H}_{19}\text{N}_3$ $[\text{M} + \text{H}]^+$ 230.1657, found 230.1646.

25

4-(2-cyclohexylethyl)-2-phenylpyrimidine-5-carbonitrile (4d.3):

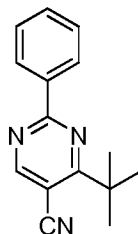
5



10

4d.3 was prepared following method B. It was purified by flash column chromatography on silica gel (hexanes:ethyl acetate, 9:1), yield: 90%, dark yellow oil. ^1H NMR (400 MHz, CDCl_3) δ 0.82-1.78 (m, 13H), 2.98 (m, 2H), 7.46 (m, 3H), 8.44 (d, 2H, $J=3.2$ Hz), 8.83 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 22.9 (23.4), 26.5 (27.0), 26.8, 27.8, 29.9 (29.6), 33.1(34.0), 36.0, 37.8, 106.0, 115.6, 129.0, 129.4, 132.4, 136.4, 160.4, 165.8, 174.5. HRMS (ESI) calcd. for $\text{C}_{19}\text{H}_{21}\text{N}_3$ $[\text{M} + \text{H}]^+$ 292.1813, found 292.1830.

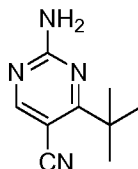
4-*tert*-butyl-2-phenylpyrimidine-5-carbonitrile (4i.3):



15

4i.3 was prepared following method A. Isolated yield: 83%, white solid, m.p. 101-103 $^\circ\text{C}$. ^1H NMR (400 MHz, CDCl_3) δ 1.60 (s, 9H), 7.49 – 7.58 (m, 3H), 8.51 – 8.55 (m, 2H), 8.94 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 28.8, 40.2, 104.0, 117.2, 128.9, 129.4, 132.4, 136.5, 162.5, 164.8, 179.3. HRMS (ESI) calcd. for $\text{C}_{15}\text{H}_{15}\text{N}_3$ $[\text{M} + \text{H}]^+$ 238.1344, found 238.1347.

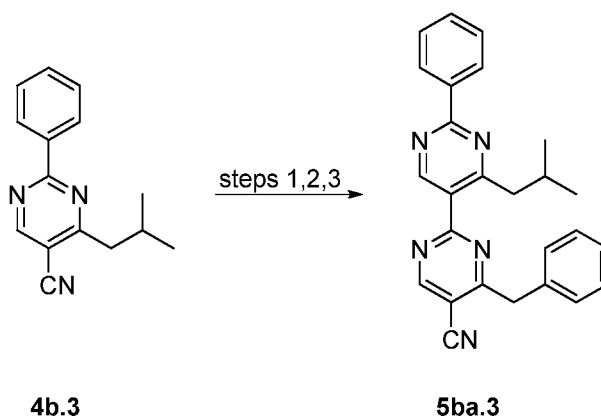
2-amino-4-*tert*-butylpyrimidine-5-carbonitrile (4i.4):



20

4i.4 was prepared following method B. Isolated yield: 75%, colorless crystals, m.p. 89-91 $^\circ\text{C}$. ^1H NMR (400 MHz, CDCl_3) δ 1.43 (s, 9H), 5.56 (br s, 2H), 8.44 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 28.5, 39.5, 95.6, 118.3, 162.4, 164.0, 181.2. HRMS (ESI) calcd. for $\text{C}_9\text{H}_{12}\text{N}_4$ $[\text{M} + \text{H}]^+$ 177.1140, found 177.1148.

Representative Procedure for the Synthesis of Dimeric 2,5-pyrimidines 5:



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4-benzyl-4'-isobutyl-2-phenyl-2,5'-bipyrimidine-5-carbonitrile (5ba.3):

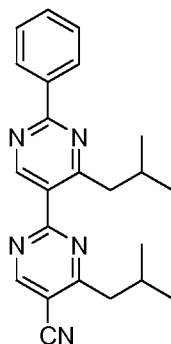
Step 1: To a mixture of compound 4b.3 (4.21 mmol, 1 g) and hydroxylamine hydrochloride (10.53 mmol, 0.72 g) in methanol (12 mL) was added triethylamine (10.53 mmol, 1.46 mL). The reaction mixture was stirred under refluxing conditions until the TLC indicated the consumption of starting material 4b.3. The solvent was removed under reduced pressure to obtain a white crude solid. The crude was dissolved in DCM (50 mL), washed with water (40 mL), dried over Na₂SO₄, and concentrated under reduced pressure to obtain intermediate amidoxime, which was used in the next step without further purification.

Step 2: Intermediate from step 1 (0.92 mmol, 0.25 g) was dissolved in glacial acetic acid (1 mL) and acetic anhydride (1.01 mmol, 95 μ L). After 5 min. of stirring, potassium formate prepared *in situ* from K₂CO₃ (5 mmol, 0.69 g), formic acid (10 mmol, 0.37 mL) in methanol (2.5 mL) was added to the mixture followed by the addition of 10% Pd/C (10 mol %, 98 mg). The reaction mixture was stirred at room temperature until the TLC indicated the consumption of starting material. The crude was filtered through Celite™ (1.5 g) and rinsed with methanol (3 mL x 3). The filtrate was concentrated under reduced pressure to obtain a yellow crude residue. To this residue, DCM (6 mL) was added and the white precipitate that formed was removed by vacuum filtration. The filtrate was concentrated under reduced pressure to obtain the crude acetate salt of carboxamidine as a yellow solid, which was used without further purification in the next step.

Step 3: To the crude carboxamidine salt dissolved in ethanol (0.8 mL) was added compound 3a (0.69 mmol, 0.147 g) and triethylamine (1.38 mmol, 0.19 mL). The reaction mixture was stirred under refluxing conditions for 2 h and then it was stirred at room temperature for 18 h. The precipitate that formed was filtered and rinsed with cold ethanol (5 mL) to yield dimer 5ba.3 in 28% yield after three steps, white solid, m.p. 144-147 °C. ¹H NMR (400 MHz, CDCl₃) δ 0.92 (d, *J* = 6.7, 6H), 2.24 – 2.35 (m, 1H), 3.19 (d, *J* = 7.1, 2H), 4.39 (s, 2H), 7.27 – 7.39 (m, 5H), 7.43 – 7.56 (m, 5H), 9.02 (s, 1H), 9.38 (s, 1H). ¹³C NMR (100 MHz, CDCl₃) δ 22.57,

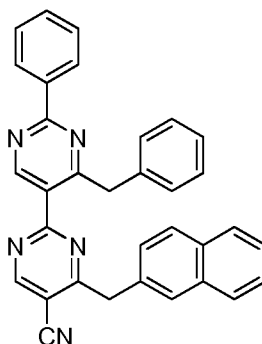
- 5 28.88, 45.71, 106.71, 115.80, 129.00, 129.38, 132.44, 136.36, 160.37, 165.72, 173.29.
HRMS (ESI) calcd. for $C_{26}H_{23}N_5$ $[M + H]^+$ 406.2032, found 406.2049.

4,4'-diisobutyl-2'-phenyl-2,5'-bipyrimidine-5-carbonitrile (5bb.3):



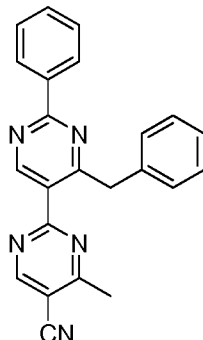
- 5bb.3 was prepared following the procedure described for 5ba.3. Isolated yield: 38% after
 10 three steps, white fluffy solid, m.p. 152-153 °C. ^1H NMR (400 MHz, $CDCl_3$) δ 0.96 (d, J = 6.7, 6H), 1.06 (d, J = 6.7, 6H), 2.24 – 2.44 (m, 2H), 2.97 (d, J = 7.2, 2H), 3.24 (d, J = 7.1, 2H), 7.50 – 7.55 (m, 3H), 8.54 – 8.59 (m, 2H), 9.02 (s, 1H), 9.41 (s, 1H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 22.64, 22.80, 28.57, 29.10, 44.97, 45.85, 107.10, 115.28, 127.69, 128.85, 128.96, 131.46, 137.44, 159.92, 160.22, 164.66, 170.29, 173.45. HRMS (ESI) calcd. for $C_{23}H_{25}N_5$ $[M$
 15 $+ H]^+$ 372.2188, found 372.2208.

4'-benzyl-4-(naphthalen-2-ylmethyl)-2'-phenyl-2,5'-bipyrimidine-5-carbonitrile (5af.3):



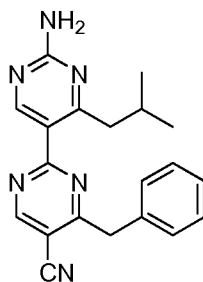
- 5af.3 was prepared following the procedure described for 5ba.3. It was purified by column
 chromatography on silica gel (hexanes:ethyl acetate, 4:1), yield: 22% after three steps, off-
 20 white solid, m.p. = 136-138 °C. ^1H NMR (400 MHz, $CDCl_3$) δ 4.51 (s, 2H), 4.70 (s, 2H), 7.08 – 7.18 (m, 5H), 7.42 – 7.54 (m, 6H), 7.76 – 7.83 (m, 4H), 8.49 – 8.55 (m, 2H), 9.00 (s, 1H), 9.47 (s, 1H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 42.4, 43.4, 106.5, 115.2, 126.4, 126.5, 126.7, 127.1, 127.2, 127.9, 128.0, 128.4, 128.9, 129.3, 131.6, 132.8, 133.1, 133.7, 137.1, 138.5, 160.4, 160.6, 165.1, 165.2, 169.0, 172.0. HRMS (ESI) calcd. for $C_{33}H_{23}N_5$ $[M + H]^+$ 490.2032, found
 25 490.2038.

- 5 4'-benzyl-4-methyl-2'-phenyl-2,5'-bipyrimidine-5-carbonitrile (5ag.3):



- 5ag.3 was prepared following the procedure described for 5ba.3. Isolated yield: 44% after three steps, tan solid, m.p. 169 °C. ¹H NMR (400 MHz, CDCl₃) δ 2.82 (s, 3H), 4.78 (s, 2H), 7.14 – 7.25 (m, 5H), 7.48 – 7.56 (m, 3H), 8.53 – 8.60 (m, 2H), 8.96 (s, 1H), 9.49 (s, 1H). ¹³C NMR (100 MHz, CDCl₃) δ 23.83, 42.63, 106.93, 115.02, 126.57, 127.13, 128.48, 128.87, 129.05, 129.36, 131.63, 137.17, 138.59, 159.93, 160.28, 164.94, 168.97, 170.49. HRMS (ESI) calcd. for C₂₃H₁₇N₅ [M + H]⁺ 364.1562, found 364.1579.

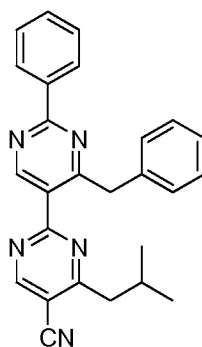
2'-amino-4-benzyl-4'-isobutyl-2,5'-bipyrimidine-5-carbonitrile (5ba.4):



- 5ba.4 was prepared following the procedure described for 5ba.3. Isolated yield: 46% after three steps, buff solid, m.p. 184-186 °C. ¹H NMR (400 MHz, CDCl₃) δ 0.85 (d, *J* = 6.7, 6H), 1.98–2.09 (m, 1H), 3.05 (d, *J* = 7.1, 2H), 4.33 (s, 2H), 5.43 (s, 2H), 7.26–7.43 (m, 5H), 8.90 (s, 1H), 9.07 (s, 1H). ¹³C NMR (100 MHz, CD₃CN) δ 21.9, 28.3, 42.8, 44.6, 105.1, 115.8, 127.4, 129.0, 129.6, 136.8, 161.0, 161.9, 163.6, 165.9, 171.5, 172.2, 174.1. HRMS (ESI) calcd. for C₂₀H₂₀N₆ [M + H]⁺ 345.1828, found 345.1820.

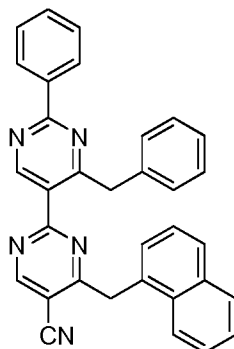
4'-benzyl-4-isobutyl-2'-phenyl-2,5'-bipyrimidine-5-carbonitrile (5ab.3):

5



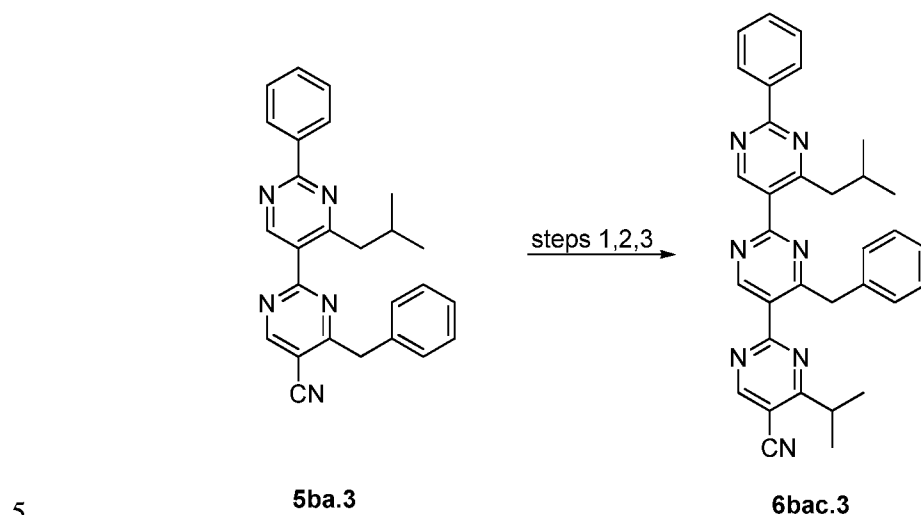
5ab.3 was prepared following the procedure described for 5ba.3. It was purified by column chromatography on silica gel (hexanes:ethyl acetate, 9:1), yield: 27% after three steps, white solid, m.p. >245 °C. ^1H NMR (400 MHz, CDCl_3) δ 1.00 (d, J = 6.7, 6H), 2.27 (m, 1H), 2.91 (d, J = 7.2, 2H), 4.77 (s, 2H), 7.24 – 7.13 (m, 5H), 7.48 – 7.55 (m, 3H), 8.51 – 8.58 (m, 2H), 8.98 (s, 1H), 9.47 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 22.57, 29.09, 42.52, 45.75, 107.22, 115.22, 126.55, 127.31, 128.48, 128.86, 129.03, 131.61, 137.18, 138.60, 160.14, 160.32, 164.89, 168.87, 173.52. HRMS (ESI) calcd. for $\text{C}_{26}\text{H}_{23}\text{N}_5$ $[\text{M} + \text{H}]^+$ 406.2032, found 406.2028.

4'-benzyl-4-(naphthalen-1-ylmethyl)-2'-phenyl-2,5'-bipyrimidine-5-carbonitrile (5ae.3):



5ae.3 was prepared following the procedure described for 5ba.3. Isolated yield: 37%, white solid, m.p. 165– 166 °C. ^1H NMR (400 MHz, CDCl_3) δ 4.45 (s, 2H), 4.84 (s, 2H), 6.88 – 6.95 (m, 2H), 7.06 – 7.11 (m, 3H), 7.42 – 7.56 (m, 7H), 7.84 (dd, J = 8.1, 19.3, 2H), 8.17 (d, J = 8.2, 1H), 8.45 – 8.52 (m, 2H), 9.01 (s, 1H), 9.33 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 40.1, 43.2, 105.0, 106.3, 115.1, 124.3, 125.5, 125.9, 126.2, 126.5, 127.3, 127.4, 127.7, 128.8, 128.9, 129.0, 129.1, 129.4, 131.6, 132.4, 134.0, 135.1, 135.6, 137.0, 160.3, 160.6, 165.1, 169.1, 172.1. HRMS (ESI) calcd. for $\text{C}_{33}\text{H}_{22}\text{N}_5$ $[\text{M} + \text{H}]^+$ 490.2026, found 490.2002.

Representative Procedure for the Synthesis of Trimeric 2,5-pyrimidines 6:



5''-cyano-4''-isopropyl-4'-phenylmethyl-4-isobutyl-2-phenylterpyrimidine (6bac.3):

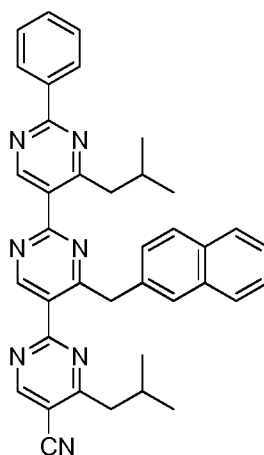
Step 1: To a mixture of compound 5ba.3 (0.6 mmol, 0.245 g) and hydroxylamine hydrochloride (1.5 mmol, 0.103 g) in methanol (8 mL) was added triethylamine (1.5 mmol, 0.21 mL). The reaction mixture was stirred under refluxing conditions until the TLC indicated the consumption of starting material 5ba.3. The solvent was removed under reduced pressure to obtain a yellow crude solid. The crude was dissolved in DCM (10 mL), washed with water (8 mL), dried over Na₂SO₄, and concentrated under vacuum to obtain an off-white, fluffy solid, which was used in the next step without further purification.

Step 2: Intermediate amidoxime obtained from step 1 (0.50 mmol, 0.22 g) was dissolved in glacial acetic acid (2 mL) and acetic anhydride (0.55 mmol, 52 μ L). After 5 min. of stirring, potassium formate prepared *in situ* from K₂CO₃ (5 mmol, 0.69 g), formic acid (10 mmol, 0.37 mL) in methanol (2.5 mL) was added to the mixture followed by the addition of 10% Pd/C (10 mol %, 89 mg). The reaction mixture was stirred at room temperature until the TLC indicated completion of the reaction. The crude was filtered through CeliteTM (1 g) and rinsed with methanol (40 mL). The filtrate was concentrated under reduced pressure to obtain a yellow crude residue. To this residue, DCM (6 mL) was added and the white precipitate that formed was removed by vacuum filtration. The filtrate was concentrated under reduced pressure to obtain the crude carboxamidinium acetate salt as a yellow solid. The crude was used without further purification in the next step.

Step 3: Crude salt from step 2 was dissolved in ethanol (3 mL); to this mixture was added compound 3c (0.41 mmol, 68.15 mg). Triethylamine was not used in this step. The reaction mixture was stirred under refluxing conditions for 28 h. The precipitate that formed was filtered and rinsed with cold ethanol (3 x 3 mL) to yield trimer 6bac.3 as an off-white solid in 41% yield after three steps, solid, m.p. 166-168 °C. ¹H NMR (400 MHz, CDCl₃) δ . 0.89 (d, *J* =

5 6.7, 6H), 1.39 (d, $J = 6.8$, 6H), 2.29 (m, 1H), 3.18 (d, $J = 7.1$, 2H), 3.53 (m, 1H), 4.80 (s, 2H), 7.16 – 7.25 (m, 5H), 7.48 – 7.54 (m, 3H), 8.52 – 8.58 (m, 2H), 9.03 (s, 1H), 9.36 (s, 1H), 9.56 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 21.3, 22.8, 28.4, 35.3, 42.2, 44.7, 106.2, 114.8, 126.8, 128.7, 128.8, 128.8, 129.4, 131.1, 137.7, 138.2, 159.5, 160.2, 160.5, 164.8, 169.0, 169.9, 178.8. HRMS (ESI) calcd. for $\text{C}_{33}\text{H}_{31}\text{N}_7$ $[\text{M} + \text{H}]^+$ 526.2719, found 526.2707.

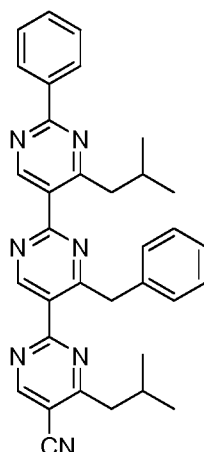
10 5''-cyano-4,4''-diisobutyl-4'-(2-naphthylmethyl)-2-phenylterpyrimidine (6.bfb.3):



6bfb.3 was prepared following the procedure described for 6bac.3. Isolated yield: 48% after three steps, off-white solid, m.p. 164-167 °C. ^1H NMR (400 MHz, CDCl_3) δ 0.83 (d, $J = 6.7$, 6H), 0.97 (d, $J = 6.7$, 6H), 2.20 – 2.32 (m, 2H), 2.93 (d, $J = 7.2$, 2H), 3.17 (d, $J = 7.1$, 2H), 4.93 (s, 2H), 7.36 – 7.46 (m, 3H), 7.49 – 7.54 (m, 3H), 7.58 (br s, 1H), 7.65 – 7.80 (m, 3H), 8.55 (m, 2H), 9.03 (s, 1H), 9.40 (s, 1H), 9.55 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) δ 22.52, 22.68, 28.52, 29.18, 42.52, 44.94, 45.78, 107.79, 114.94, 125.92, 126.39, 127.67, 127.72, 127.75, 127.83, 127.86, 128.34, 128.78, 129.05, 129.22, 132.06, 132.40, 133.63, 135.68, 153.93, 157.76, 160.23, 160.28, 164.42, 169.12, 173.88. HRMS (ESI) calcd. for $\text{C}_{38}\text{H}_{35}\text{N}_7$ $[\text{M} + \text{H}]^+$ 590.3032, found 590.3054.

5''-cyano-4,4''-diisobutyl-4'-(2-phenylmethyl)-2-phenylterpyrimidine (6bab.3):

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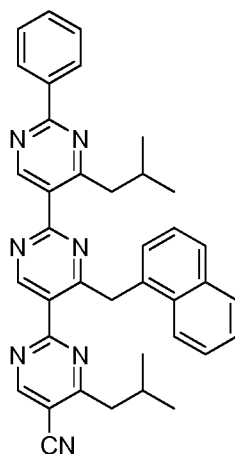


10

6bab.3 was prepared following the procedure described for 6bac.3. Isolated yield = 37% after three steps, white solid, m.p. 188-189 °C. ^1H NMR (400 MHz, CDCl_3) δ 0.90 (d, $J = 6.7$, 6H), 1.02 (d, $J = 6.7$, 6H), 2.22 – 2.36 (m, 2H), 2.95 (d, $J = 7.2$, 2H), 3.20 (d, $J = 7.1$, 2H), 4.77 (s, 2H), 7.17 – 7.25 (m, 5H), 7.49 – 7.54 (m, 3H), 8.52 – 8.58 (m, 2H), 9.03 (s, 1H), 9.38 (s, 1H), 9.52 (s, 1H). ^{13}C NMR (100 MHz, CDCl_3) 22.59, 22.80, 28.47, 29.20, 42.30, 44.71, 45.80, 107.64, 115.04, 126.79, 127.43, 128.57, 128.67, 128.79, 128.83, 129.34, 131.14, 137.76, 138.27, 159.56, 160.12, 160.26, 164.18, 164.62, 164.75, 168.98, 169.98, 173.80. HRMS (ESI) calcd. for $\text{C}_{34}\text{H}_{33}\text{N}_7$ $[\text{M} + \text{H}]^+$ 540.2876, found 540.2869.

5''-cyano-4,4''-diisobutyl-4'-(1-naphthylmethyl)-2-phenylterpyrimidine (6.beb.3):

15

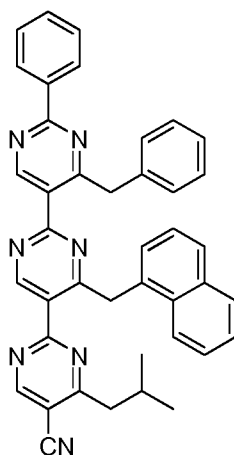


20

6beb.3 was prepared following the procedure described for 6bac.3. Isolated yield: 20% after three steps, white solid, m.p. 165– 166 °C. ^1H NMR (400 MHz, CDCl_3) δ 0.71 (d, $J = 6.7$ Hz, 6H), 0.76 (d, $J = 6.6$ Hz, 6H), 1.89 – 2.01 (m, 1H), 2.05 - 2.18 (m, 1H), 2.66 (d, $J = 7.3$ Hz, 2H), 2.95 (d, $J = 7.0$ Hz, 2H), 5.14 (s, 2H), 6.95 (d, $J = 7.0$ Hz, 1H), 7.19-7.25 (m, 1H), 7.39-7.47 (m, 5H), 7.65 (d, $J = 8.2$ Hz, 1H), 7.79 (dd, $J = 6.2, 3.3$ Hz, 1H), 7.98 (dd, $J = 6.1, 3.4$ Hz,

5 1H), 8.41-8.48 (m, 2H), 8.86 (s, 1H), 9.21 (s, 1H), 9.53 (s, 1H). ¹³C NMR (100 MHz, CDCl₃) δ 22.4, 22.7, 28.4, 29.0, 40.1, 44.5, 45.6, 107.5, 115.0, 124.0, 125.5, 126.0, 126.4, 126.7, 127.5, 127.7, 128.5, 128.8, 128.8, 129.1, 131.1, 132.3, 134.1, 134.7, 137.8, 159.5, 160.0, 160.2, 164.1, 164.5, 164.9, 169.1, 170.0, 173.8. HRMS (ESI) calcd. for C₃₈H₃₅N₇ [M + H]⁺ 590.3027, found 590.2984.

10 5"-cyano-4"-isobutyl-4'-(1-naphthylmethyl)-4-(2-phenylmethyl)-2-phenylterpyrimidine (6aeb.3):



6aeb.3 was prepared following the procedure described for 6bac.3. Isolated yield: 18% after three steps, white fluffy solid, m.p. 155–156 °C. ¹H NMR (400 MHz, CDCl₃) δ 0.85 (d, *J* = 6.7 Hz, 6H), 1.99 – 2.11 (m, 1H), 2.75 (d, *J* = 7.3 Hz, 2H), 4.47 (s, 2H), 5.21 (s, 2H), 6.99 (d, *J* = 7.0 Hz, 1H), 7.08 (ddd, *J* = 9.57, 5.70, 1.94 Hz, 5H), 7.27 (dd, *J* = 8.11, 7.11 Hz, 1H), 7.44 - 7.50 (m, 5H), 7.69 (d, *J* = 8.24 Hz, 1H), 7.80 - 7.86 (m, 1H), 7.99 - 8.05 (m, 1H), 8.45 - 8.50 (m, 2H), 8.94 (s, 1H), 9.38 (s, 1H), 9.60 (s, 1H). ¹³C NMR (100 MHz, CDCl₃) δ 22.4, 29.0, 40.1, 41.9, 45.6, 107.6, 115.0, 123.9, 125.6, 126.0, 126.2, 126.4, 126.9, 127.6, 127.7, 127.8, 128.3, 128.8, 128.9, 129.1, 129.5, 131.3, 132.3, 134.0, 134.7, 137.6, 138.8, 160.0, 160.2, 164.2, 164.4, 168.7, 169.2, 173.8. HRMS (ESI) calcd. for C₄₁H₃₃N₇ [M + H]⁺ 624.2870, found 624.2844.

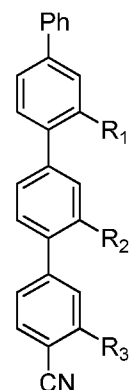
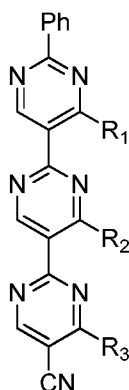
B. QikProp Calculation Parameters

Maestro (2) was used to view and build molecular models of 4,4',4"-trimethyl terpyrimidinylene and an ideal α-helix composed of 8 alanine residues. MacroModel (3) performed a conformational search of TMOP using OPLS 2005 force fields and GB/SA solvation (4). Maestro was used to superimpose the *ith*, *ith* + 4, and *ith* + 7 methyl carbons of the octa-alanine with the methyl carbons of the terpyrimidinylene. Out of 100,000 conformations generated, approximately 5,000 (5%) of the conformations aligned well with the α-helix: most of the aligned conformations exhibited the fourth lowest potential energy

- 5 calculated by MacroModel with an RMSD of 0.68 Å. PyMol (5) was used to create the image of TMOP superimposed on octa-alanine.

ClogP determination using QikProp was performed on two compounds: Compound 14 reported by Hang Y. *et al.* (J.Am.Chem.Soc. 2005, 127, 10191 – 10196) and its terpyrimidinylene scaffold analog.

- 10 1. QikProp, 3.1; Schrödinger, LLC: New York, NY, 2008.
2. Maestro, 8.5; Schrödinger, LLC: New York, NY, 2008.
3. MacroModel, 9.6; Schrödinger, LLC: New York, NY, 2008.
4. Qiu, D.; Shenkin, P. S.; Hollinger, F. P.; Still, W. C., The GB/SA Continuum Model for Solvation. A Fast Analytical Method for the Calculation of Approximate Born Radii. J. Phys. Chem. 1997, 101, (16), 3005-3014.
- 15 5. Delano, W. L. The PyMol Molecular Graphics System, Delano Scientific: Palo Alto, CA, 2002.



5

Table 1

Compound		pyrimidine	log P*		phenyl	log P*
6bab.3	R ₁	<i>i</i> -Bu	5.756	R ₁ '	<i>i</i> -Bu	7.035
	R ₂	Bn		R ₂ '	Bn	
	R ₃	<i>i</i> -Bu		R ₃ '	<i>i</i> -Bu	
6bfb.3	R ₁	<i>i</i> -Bu methyl(2-	7.326	R ₁ '	<i>i</i> -Bu methyl(2-	11.059
	R ₂	naphthyl)		R ₂ '	naphthyl)	
	R ₃	<i>i</i> -Bu		R ₃ '	<i>i</i> -Bu	
6beb.3	R ₁	<i>i</i> -Bu methyl(1-	7.035	R ₁ '	<i>i</i> -Bu methyl(1-	11.341
	R ₂	naphthyl)		R ₂ '	naphthyl)	
	R ₃	<i>i</i> -Bu		R ₃ '	<i>i</i> -Bu	
6aeb.3	R ₁	Bn methyl(1-	7.765	R ₁ '	Bn methyl(1-	12.217
	R ₂	naphthyl)		R ₂ '	naphthyl)	
	R ₃	<i>i</i> -Bu		R ₃ '	<i>i</i> -Bu	

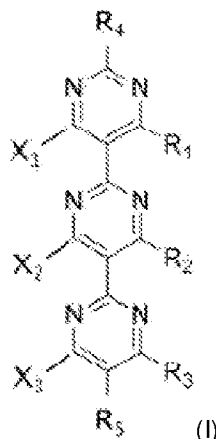
*The log P values have been calculated by Maetro 8.5

All references cited in the present application are incorporated in their entirety herein by reference to the extent not inconsistent herewith.

- 10 It will be seen that the advantages set forth above, and those made apparent from the foregoing description, are efficiently attained and since certain changes may be made in the above construction without departing from the scope of the invention, it is intended that all matters contained in the foregoing description or shown in the accompanying drawings shall be interpreted as illustrative and not in a limiting sense.
- 15 It is also to be understood that the following claims are intended to cover all of the generic and specific features of the invention herein described, and all statements of the scope of the invention which, as a matter of language, might be said to fall therebetween. Now that the invention has been described,

5 What is claimed is:

1. A compound or its pharmaceutically acceptable salt of the formula (I):

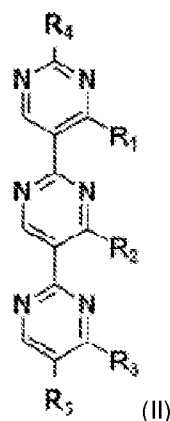


wherein X_1 , X_2 , and X_3 are independently selected from the group H and NH_2 ;

10 R_1 , R_2 and R_3 are independently selected from the group of moieties consisting of $-H$, $-C_1-C_6$ alkyl, $-C_6-C_{12}$ aryl; $-C_7-C_{18}$ alkylaryl, $-C_4-C_{18}$ alkylheterocycle, and $-C_7-C_{18}$ alkylheteroaryl, wherein one $-CH_2-$ of the alkyl may be replaced by $-S-$, and wherein the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from the group

15 R_4 and R_5 are independently selected from the group of moieties consisting of $-H$, $-C_1-C_6$ alkyl, $-C(O)O(C_1-C_6$ alkyl), $-C(O)O(C_6-C_{12}$ aryl), $-C(O)O(C_7-C_{18}$ alkylaryl), $-C(O)O(C_7-C_{18}$ alkylheteroaryl), $-SO_2(C_1-C_6$ alkyl), $-SO_2(C_6-C_{12}$ aryl), $-SO_2(C_7-C_{18}$ alkylaryl), $-SO_2(C_7-C_{18}$ alkylheteroaryl), $-C(O)NH(C_1-C_6$ alkyl), $-C(O)NH(C_6-C_{12}$ aryl); $-C(O)NH(C_7-C_{18}$ alkylaryl), and $(C_7-C_{18}$ alkylheteroaryl), wherein one $-CH_2-$ of the alkyl may be replaced by $-S-$, and wherein the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from the group consisting of C_1-C_3 alkyl, OH, SH, NH_2 , and COOH.

25 2. A compound or its pharmaceutically acceptable salt of the formula (II):



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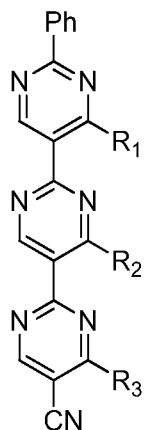
wherein R_1 , R_2 and R_3 are independently selected from the group of moieties consisting of $-H$, $-C_1-C_6$ alkyl, $-C_6-C_{12}$ aryl; $-C_7-C_{18}$ alkylaryl, $-C_4-C_{18}$ alkylheterocycle, and $-C_7-C_{18}$ alkylheteroaryl, wherein one $-CH_2-$ of the alkyl may be replaced by $-S-$, and wherein the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from the group consisting of C_1-C_3 alkyl, OH , SH , NH_2 , and $COOH$; and

wherein R_4 and R_5 are independently selected from the group of moieties consisting of $-H$, $-C_1-C_6$ alkyl, $-C(O)O(C_1-C_6$ alkyl), $-C(O)O(C_6-C_{12}$ aryl), $-C(O)O(C_7-C_{18}$ alkylaryl), $-C(O)O(C_7-C_{18}$ alkylheteroaryl), $-SO_2(C_1-C_6$ alkyl), $-SO_2(C_6-C_{12}$ aryl), $-SO_2(C_7-C_{18}$ alkylaryl), $-SO_2(C_7-C_{18}$ alkylheteroaryl), $-C(O)NH(C_1-C_6$ alkyl), $-C(O)NH(C_6-C_{12}$ aryl); $-C(O)NH(C_7-C_{18}$ alkylaryl), and $(C_7-C_{18}$ alkylheteroaryl), wherein one $-CH_2-$ of the alkyl may be replaced by $-S-$, and wherein the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from the group consisting of C_1-C_3 alkyl, OH , SH , NH_2 , and $COOH$.

25

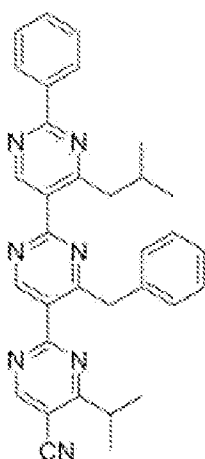
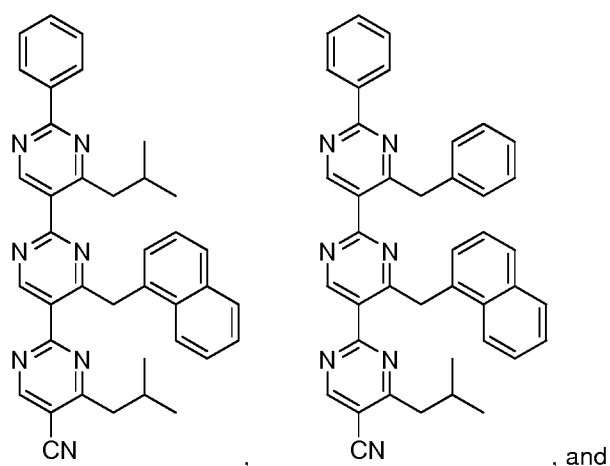
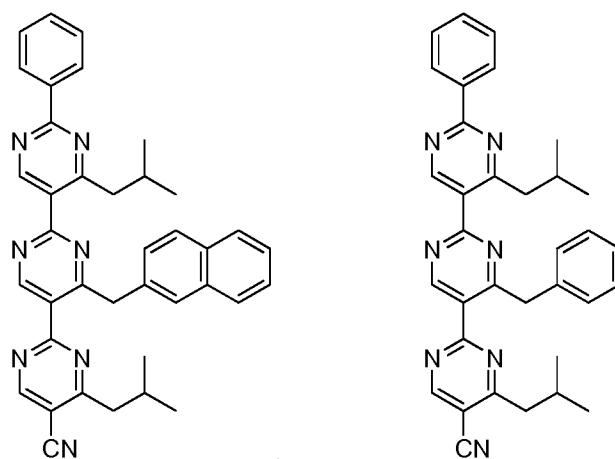
3. The compound according to claim 2 wherein at least one of R^1 , R^2 and R^3 are not H .
4. The compound according to claim 2 comprising a plurality of formula (II) subunits wherein an R_4 moiety is covalently coupled to an R_5 moiety of an adjacent subunit.
5. The compound according to claim 2 wherein each of R^1 , R^2 and R^3 independently form an amino acid residue selected from the group consisting of alanine, arginine, asparagine, aspartic acid, cysteine, glutamine, glutamic acid, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine and valine.

- 5 6. The compound according to claim 2 wherein each of R^1 , R^2 and R^3 is a moiety individually selected from the group consisting of H, CH_3 , NH_2 , $COOH$, Ph, *i*-Bu, *t*-Bu, CN, Bn, methyl(1-naphthyl), methyl(2-naphthyl), and *i*-Pr.
7. The compound according to claim 2 having the structure:



- 10 8. The compound according to claim 2 having the structure selected from the group consisting of:

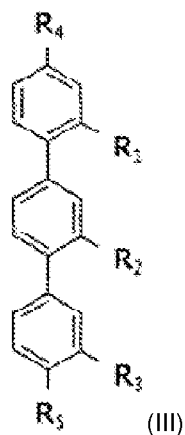
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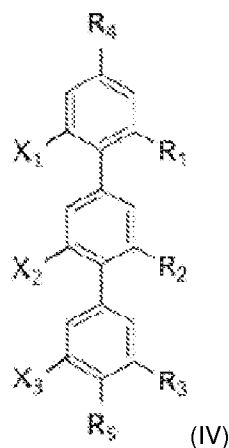
9. A method of treating a condition or disease state in a patient, the condition or disease state being modulated through the interaction of an α -helical first protein with a binding site of a second protein, the method comprising administering to a patient in

- 5 need of therapy an effective amount of one or more compounds according to claim 2, optionally in a pharmaceutically acceptable carrier, additive or excipient.
10. A method of inhibiting or disrupting the interactions between an alpha helix of a first protein and an alpha helix binding pocket of a second protein, the method comprising contacting the first protein and the second protein with a compound according to
- 10 claim 2 under conditions wherein the interactions between the alpha helix of the first protein and the alpha helix binding pocket of the second protein are inhibited or disrupted.
11. A compound or its pharmaceutically acceptable salt of the formula (III):



- 15 wherein R_1 , R_2 and R_3 are independently selected from the group of moieties consisting of $-H$, $-C_1-C_6$ alkyl, $-C_6-C_{12}$ aryl; $-C_7-C_{18}$ alkylaryl, $-C_4-C_{18}$ alkylheterocycle, and $-C_7-C_{18}$ alkylheteroaryl, wherein one $-CH_2-$ of the alkyl may be replaced by $-S-$, and wherein the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from
- 20 the group consisting of C_1-C_3 alkyl, OH , SH , NH_2 , and $COOH$; and
- wherein R_4 and R_5 are independently selected from the group of moieties consisting of $-H$, $-C_1-C_6$ alkyl, $-C(O)O(C_1-C_6$ alkyl), $-C(O)O(C_6-C_{12}$ aryl), $-C(O)O(C_7-C_{18}$ alkylaryl), $-C(O)O(C_7-C_{18}$ alkylheteroaryl), $-SO_2(C_1-C_6$ alkyl), $-SO_2(C_6-C_{12}$ aryl), $-SO_2(C_7-C_{18}$ alkylaryl), $-SO_2(C_7-C_{18}$ alkylheteroaryl), $-C(O)NH(C_1-C_6$ alkyl), $-C(O)NH(C_6-C_{12}$ aryl); $-C(O)NH(C_7-C_{18}$ alkylaryl), and $(C_7-C_{18}$ alkylheteroaryl), wherein one $-CH_2-$ of the alkyl may be replaced by $-S-$, and wherein the alkyl, aryl, or heteroaryl may be substituted with
- 25 one or two moieties selected from the group consisting of C_1-C_3 alkyl, OH , SH , NH_2 , and $COOH$.
- 30

- 5 12. The compound according to claim 12 wherein at least one of R^1 , R^2 and R^3 are not H.
13. The compound according to claim 12 comprising a plurality of formula (II) subunits wherein an R_4 moiety is covalently coupled to an R_5 moiety of an adjacent subunit.
14. The compound according to claim 12 wherein each of R^1 , R^2 and R^3 independently form an amino acid residue selected from the group consisting of alanine, arginine,
 10 asparagine, aspartic acid, cysteine, glutamine, glutamic acid, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine and valine.
15. The compound according to claim 12 wherein each of R^1 , R^2 and R^3 is a moiety independently selected from the group consisting of H, CH_3 , NH_2 , $COOH$, Ph, *i*-Bu, *t*-Bu, CN, Bn, methyl(1-naphthyl), methyl(2-naphthyl), and *i*-Pr.
 15
16. A method of treating a condition or disease state in a patient, the condition or disease state being modulated through the interaction of an α -helical first protein with a binding site of a second protein, the method comprising administering to a patient in need of therapy an effective amount of one or more compounds according to claim
 20 11, optionally in a pharmaceutically acceptable carrier, additive or excipient.
17. A method of inhibiting or disrupting the interactions between an alpha helix of a first protein and an alpha helix binding pocket of a second protein, the method comprising contacting the first protein and the second protein with a compound according to claim 11 under conditions wherein the interactions between the alpha helix of the first
 25 protein and the alpha helix binding pocket of the second protein are inhibited or disrupted.
18. A compound or its pharmaceutically acceptable salt of the formula (IV):



5 wherein X_1 , X_2 , and X_3 are independently selected from the group H and NH_2 ;

10 R_1 , R_2 and R_3 are independently selected from the group of moieties consisting of $-H$, $-C_1-C_6$ alkyl, $-C_6-C_{12}$ aryl; $-C_7-C_{18}$ alkylaryl, $-C_4-C_{18}$ alkylheterocycle, and $-C_7-C_{18}$ alkylheteroaryl, wherein one $-CH_2-$ of the alkyl may be replaced by $-S-$, and wherein the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from the group consisting of C_1-C_3 alkyl, OH, SH, NH_2 , and COOH; and

15 R_4 and R_5 are independently selected from the group of moieties consisting of $-H$, $-C_1-C_6$ alkyl, $-C(O)O(C_1-C_6$ alkyl), $-C(O)O(C_6-C_{12}$ aryl), $-C(O)O(C_7-C_{18}$ alkylaryl), $-C(O)O(C_7-C_{18}$ alkylheteroaryl), $-SO_2(C_1-C_6$ alkyl), $-SO_2(C_6-C_{12}$ aryl), $-SO_2(C_7-C_{18}$ alkylaryl), $-SO_2(C_7-C_{18}$ alkylheteroaryl), $-C(O)NH(C_1-C_6$ alkyl), $-C(O)NH(C_6-C_{12}$ aryl); $-C(O)NH(C_7-C_{18}$ alkylaryl), and $(C_7-C_{18}$ alkylheteroaryl), wherein one $-CH_2-$ of the alkyl may be replaced by $-S-$, and wherein the alkyl, aryl, or heteroaryl may be substituted with one or two moieties selected from the group consisting of C_1-C_3 alkyl, OH, SH, NH_2 , and COOH.

20

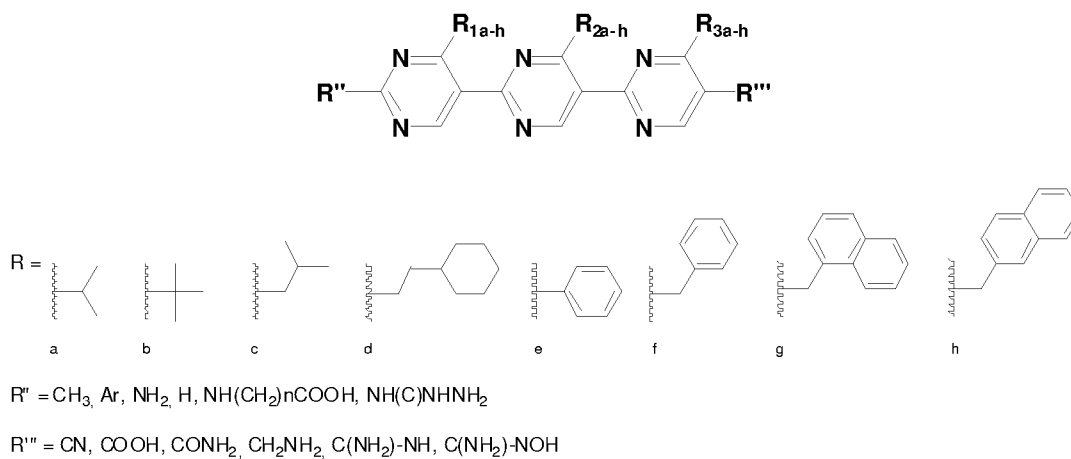


FIG. 1

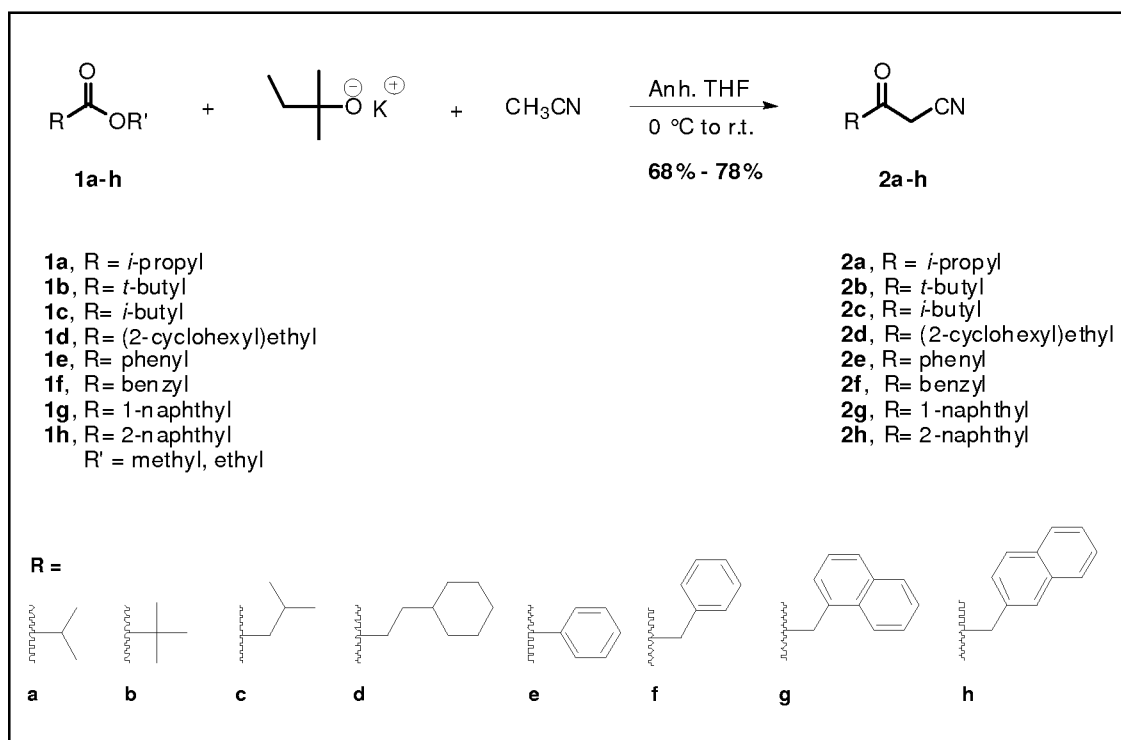


FIG. 2

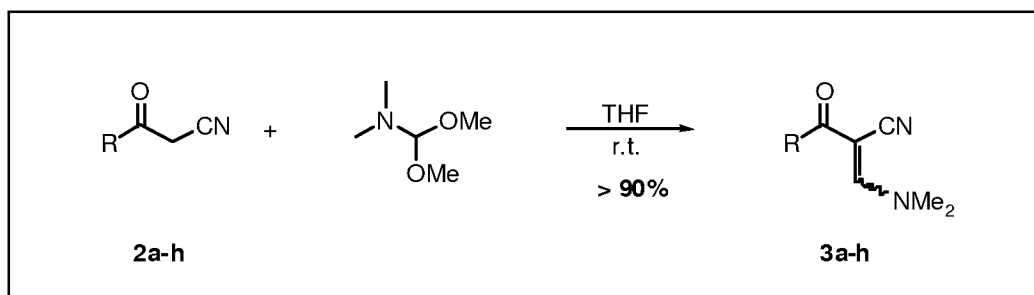


FIG. 3

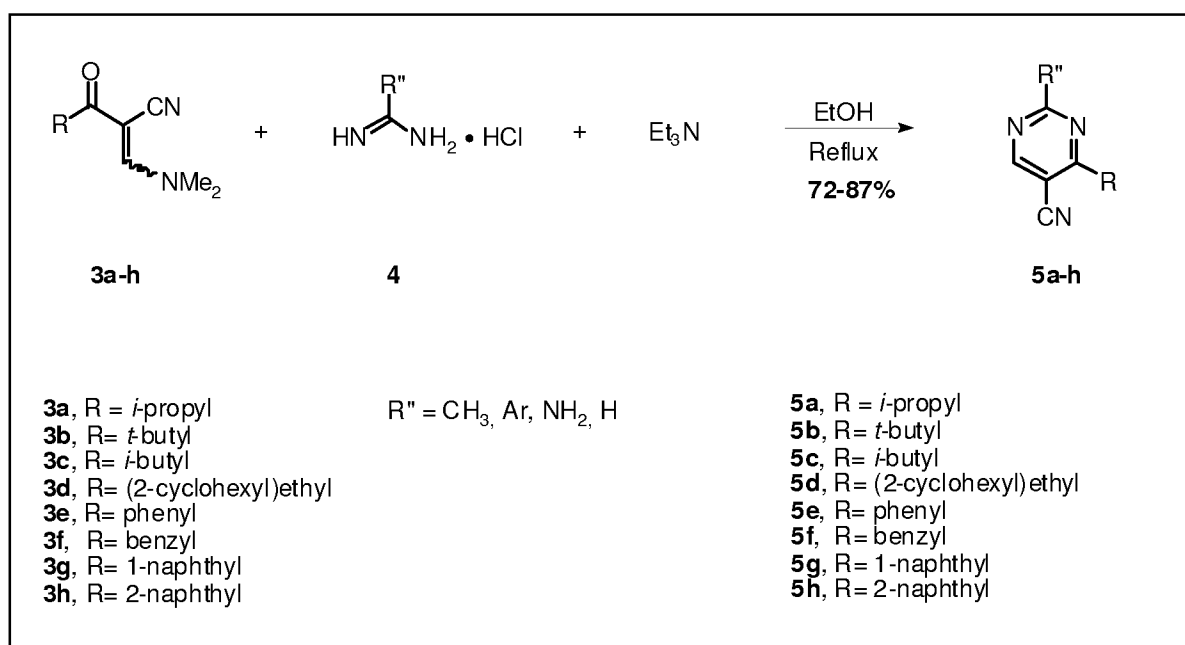


FIG. 4

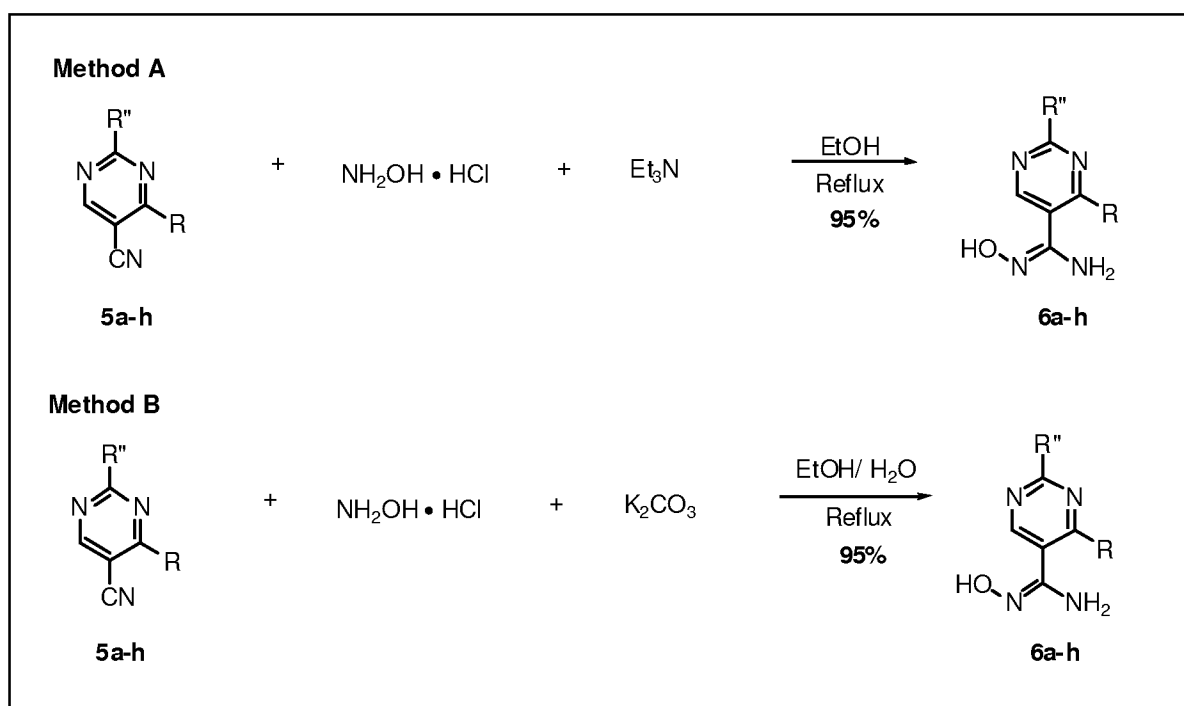


FIG. 5

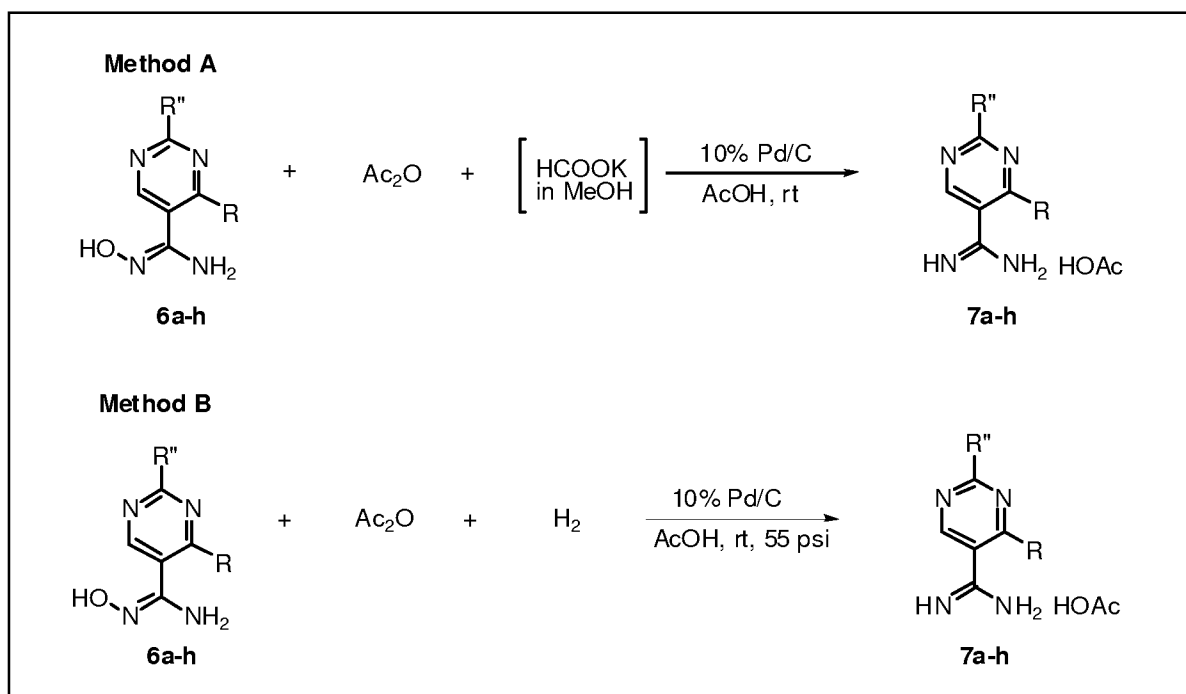


FIG. 6

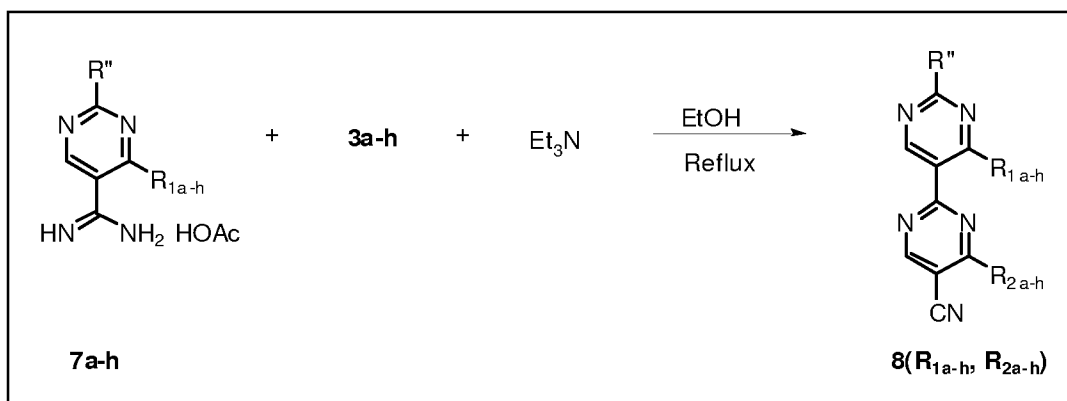


FIG. 7

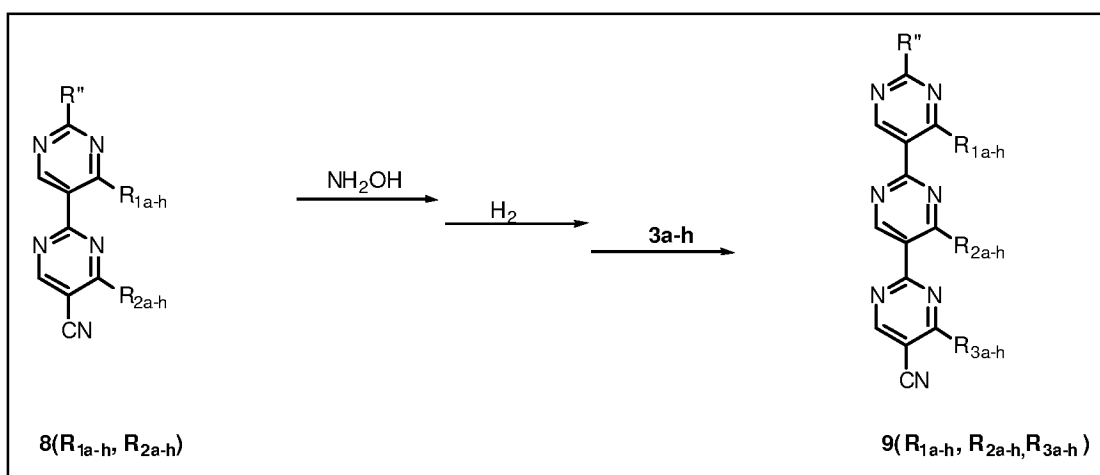
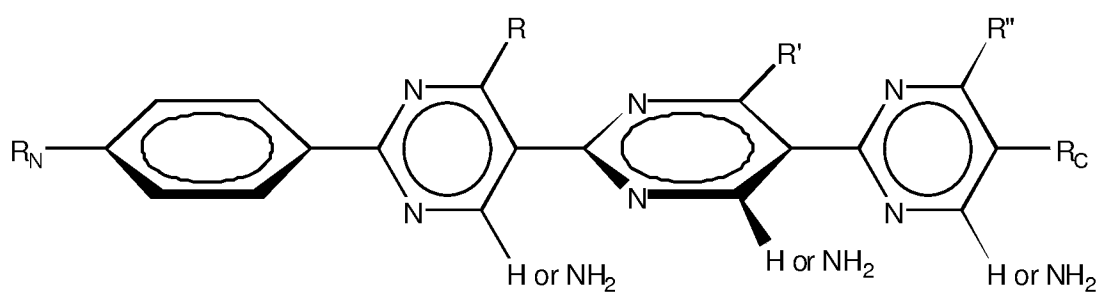


FIG. 8



$\text{R}_N, \text{R}_C = \text{CO}_2\text{H}$

$\text{R, R', and R''} = \text{CH}_3, \text{CH}_2\text{OH}, \text{CH}_2\text{CH}(\text{CH}_3)_2, \text{CH}_2\text{Ph}, \text{CH}_2(4\text{-hydroxyphenyl}), \text{or CH}_2(3\text{-indolyl})$

FIG. 9

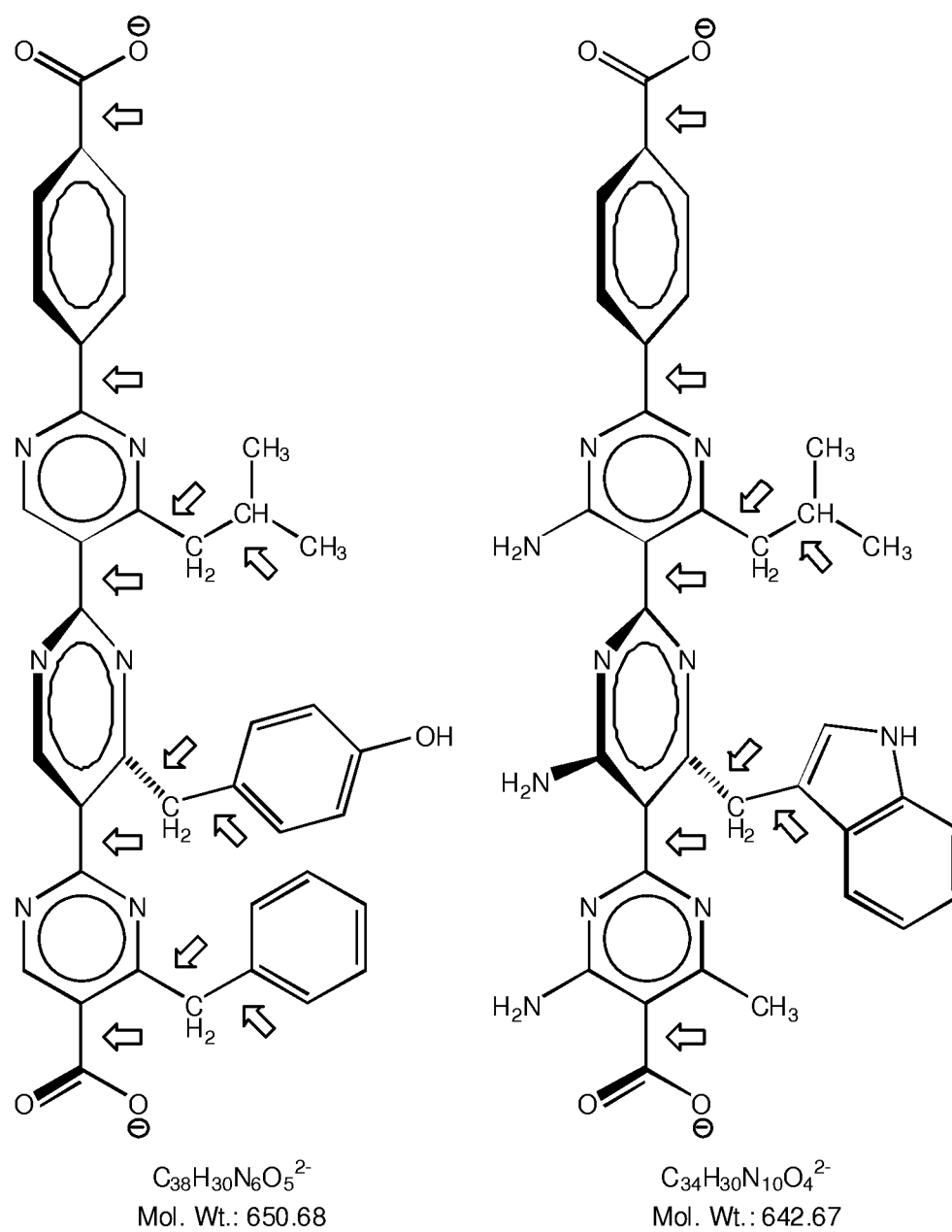


FIG. 10

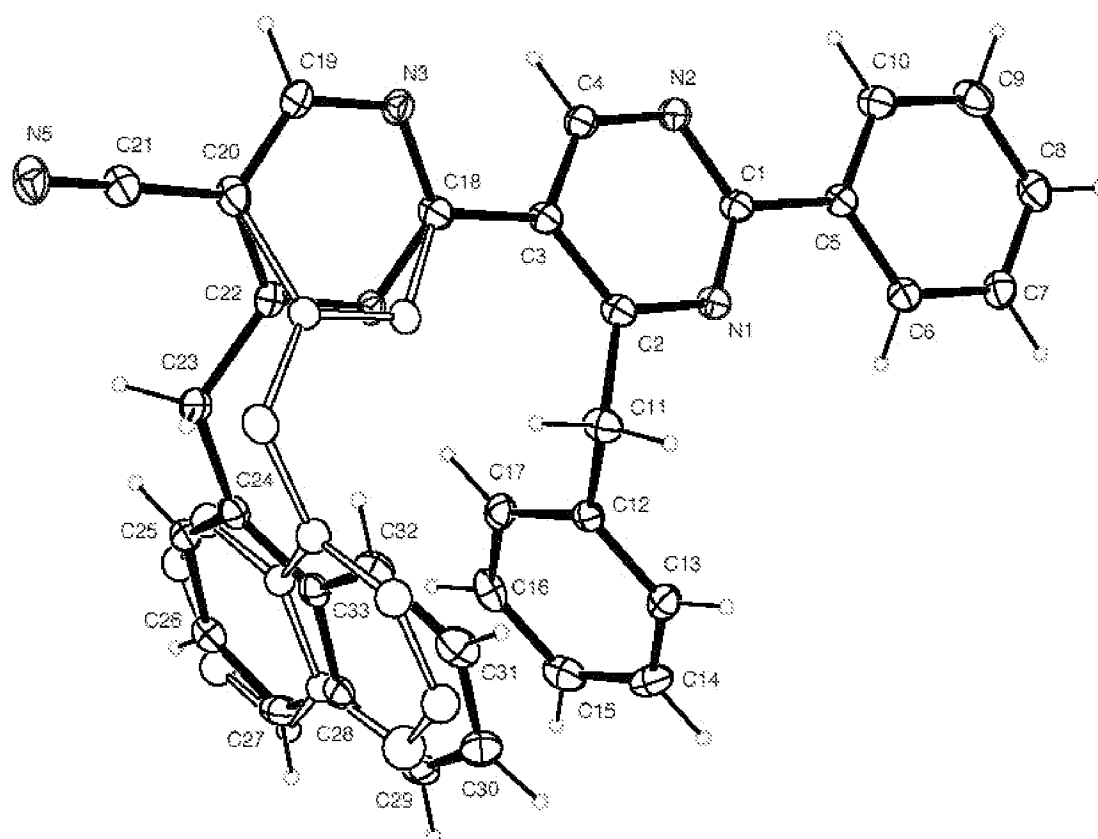


FIG. 11

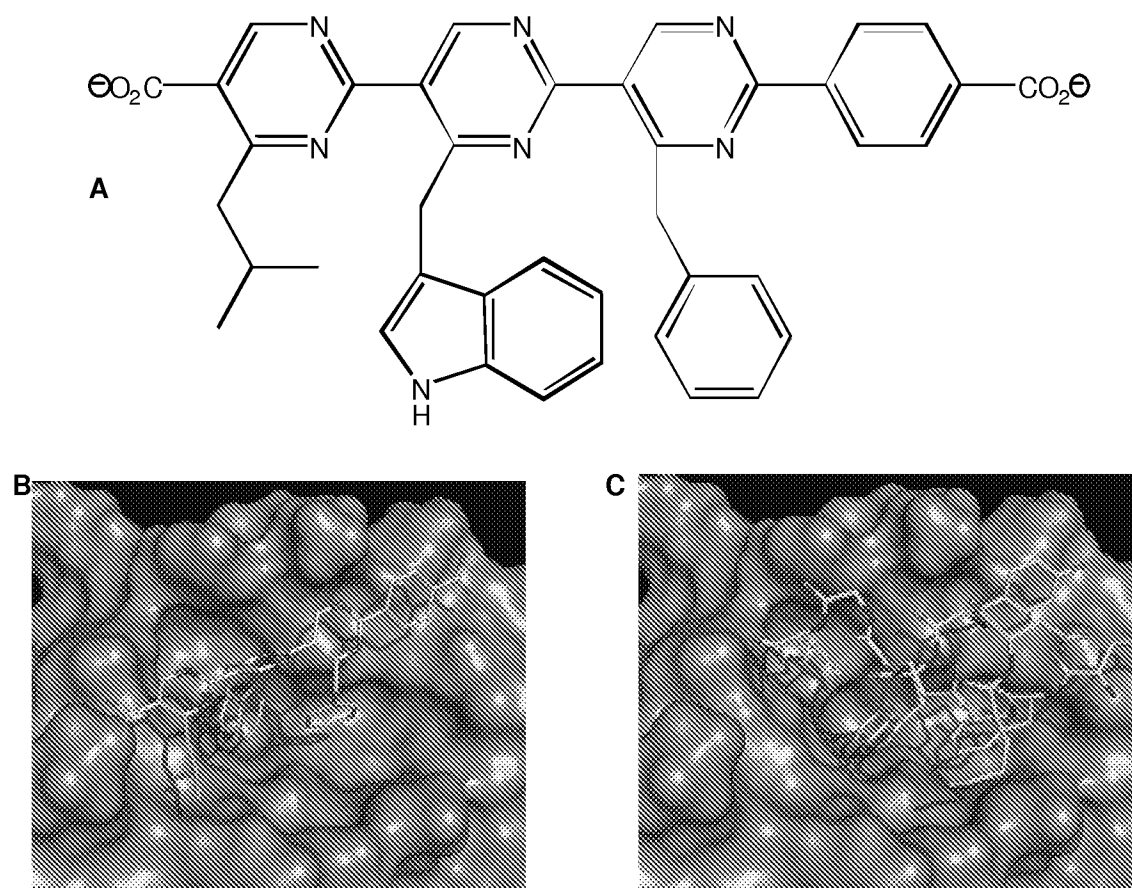


FIG. 12

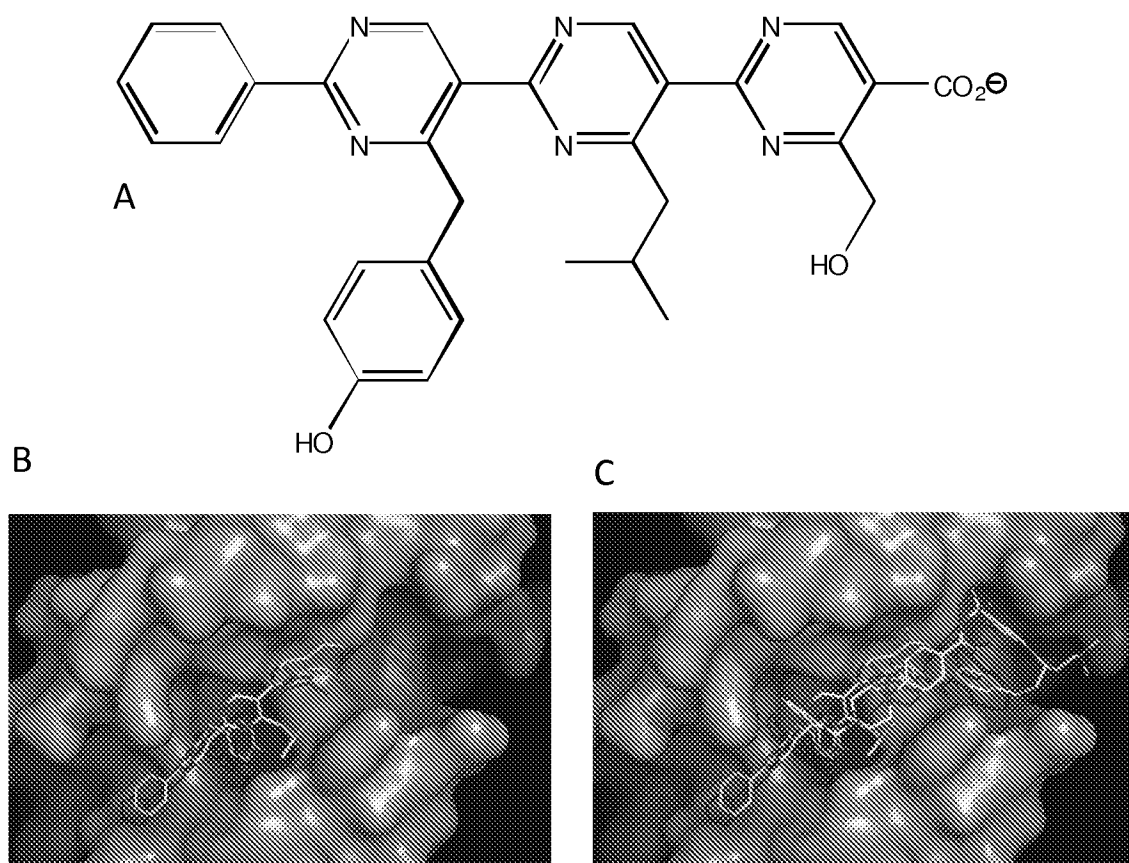
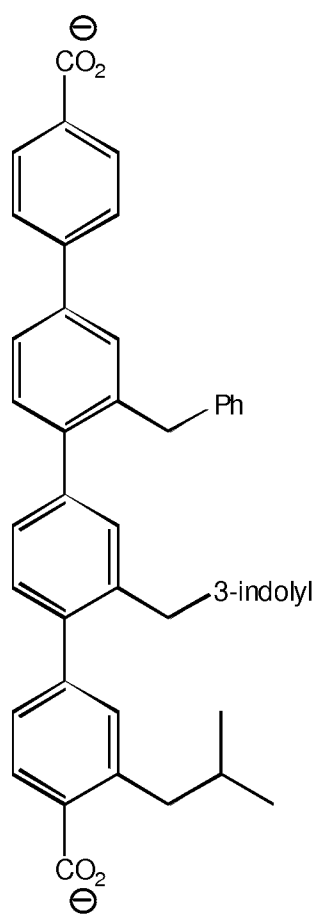
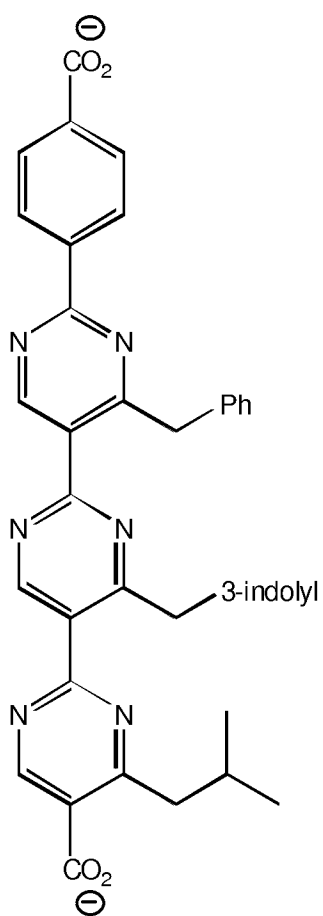


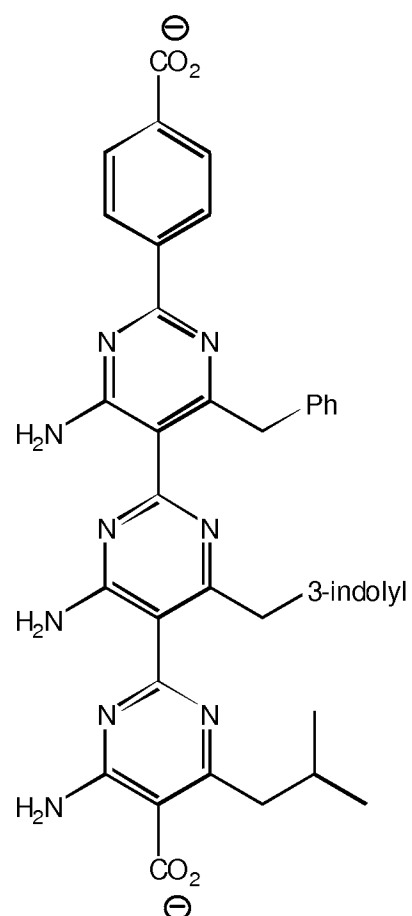
FIG. 13



$\log P_{\text{octanol/water}} = 10.2$



$\log P_{\text{octanol/water}} = 6.8$



$\log P_{\text{octanol/water}} = 5.2$

FIG. 14

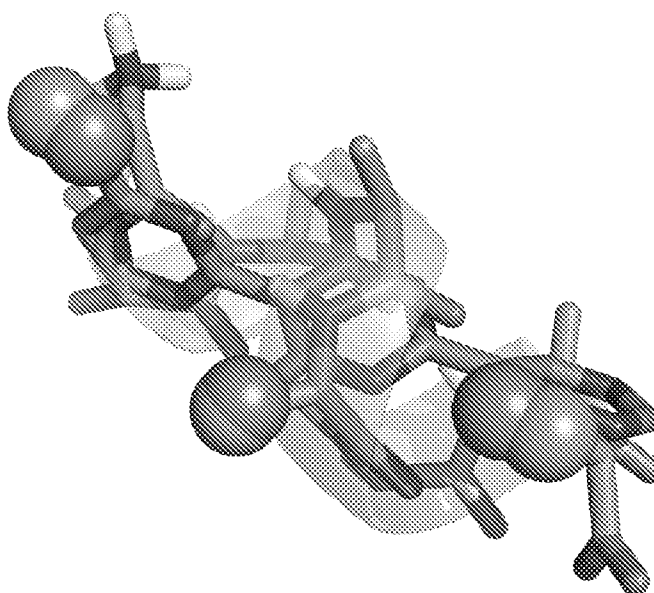


FIG. 15

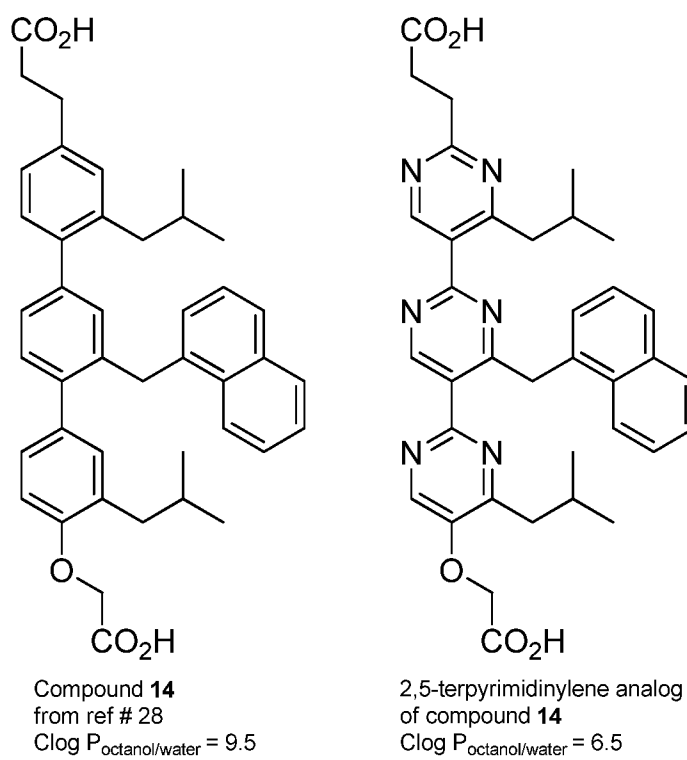


FIG. 16

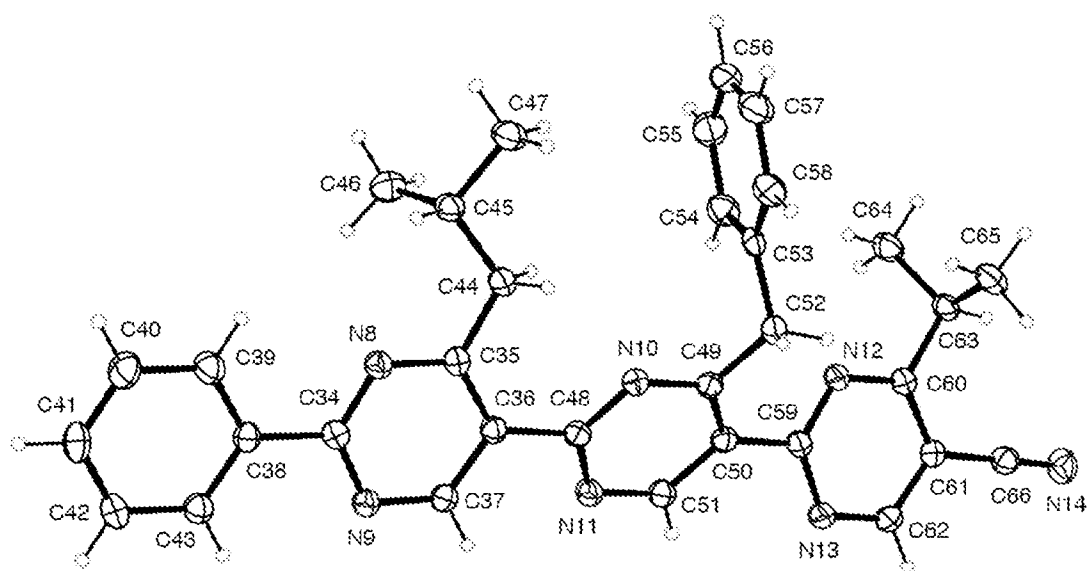


FIG. 17