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Benzimidazole Condensed Ring Systems: New Synthesis and Antineoplastic Activity of Substituted 3,4-Dihydro- and 1,2,3,4-Tetrahydro-benzo[4,5]imidazo[1,2-a]pyrimidine Derivatives

Atef Abdel-monem Abdel-hafez

Department of Pharmaceutical Medicinal Chemistry, Faculty of Pharmacy, Assiut University, Assiut 71526, Egypt

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As part of an ongoing effort to develop new antineoplastic agents, a series of substituted 3,4-dihydro- and 1,2,3,4-tetrahydro-benzo[4,5]imidazo[1,2-a]pyrimidine derivatives (**5**-**19**) were synthesized. 1,2,3,4-Tetrahydrobenzo[4,5]imidazo[1,2-a]pyrimidine-2-one derivatives (**5**-**7**) were prepared via one-pot two-component thermal cyclization reaction of 2-aminobenzimidazole **1** and *P*-substituted methyl cinnamates (**2**-**4**). Vilsmir-Haack formylation of these derivatives (**5**-**7**) afforded the 2-chloro-3-carboxaldehyde targets (**8**-**10**) followed by nucleophilic displacement of the chloro atom in the 3-carboxaldehyde compounds (**8**-**10**) to yield the remaining final targets (**11**-**19**). The structures of the synthesized derivatives (**5**-**19**) were confirmed by means of IR, ^{1}H NMR, MS and elemental analyses. The synthesized derivatives (**5**-**19**) were subjected to the National Cancer Institute (NCI) *in vitro* disease human cell screening panel assay. 2-Chloro-4-phenyl-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxyaldehyde (**8**, NCI 722731) and 4-(4-methoxyphenyl)-2-(4-methylpiperazin-1-yl)-3,4-dihydrobenzo[4,5]imidazo [1,2-a]pyrimidine-3-carboxaldehyde (**18**, NCI 722739) showed a variable degree of antineoplastic activity against some of the cell lines tested. 2-Chloro-4-(4-nitrophenyl)-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxyaldehyde (**10**, NCI 722743) exhibited good *in vitro* antineoplastic activity with subpanel disease selectivity against all the cell lines tested with $\log_{10}$ GI_{50} (M), the concentration that inhibits 50% of cell growth, values ranging from –5.08 to < -8.00.

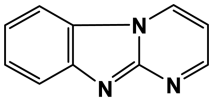
Key words: Benzimidazole, Methyl cinnamates, Pyrimido[1,2-a]benzimidazole, Azinazoles, Anticancer, Cytotoxicity

INTRODUCTION

Azinazoles including pyrido-, pyrimido-, and triazino-benzimidazole heterocycles have played a pivotal role in the development of effective pharmacophores (Frolov, 2004). The benzimidazole nucleus is an essential part of many antineoplastic derivatives (Soliman *et al.*, 1984). The pyrido[1,2-a]benzimidazole ring system has been found to exhibit anticancer properties by Badawy and Kappe (Badawy and Kappe, 1995). We became interested in the pyrimido[1,2-a]benzimidazole framework (**A**) when we tried to determine the effect of the nature of the condensed heterocycle as part of our continuation of this research. The majority of DNA intercalating antitumor drugs has a common general structure comprising a planar tricyclic and tetracyclic chromophore (Filippatos *et al.*, 1994; Matelli *et al.*, 1988; Kimura *et al.*, 1992; Abadi *et al.*, 1999; Palmer *et al.*, 1988). These structural observations led to the synthesis of a series of substituted 3,4-dihydro- and 1,2,3,4-tetrahydro-benzo[4,5]imidazo[1,2-a]pyrimidine derivatives (**5**-**19**) possessing a bridgehead nitrogen atom with a view of designing new potential anticancer agents. Thermal cyclization to form a new C-N bond is a common synthetic approach to synthesize azinazoles. Thus pyrimido [1,2-a] benzimidazoles tricyclic system is formed by the reaction of 2-aminobenzimidazole with β-diketones (Kreutzberger and Leger, 1981), 2-amino-3-ethoxycarbonyl-4,5-dihydrofuranes (Elnagdi and WamHoff, 1981), and carbon suboxide in the presence of aluminium chloride (Bousignore *et al.*, 1992). In the present study, we describe a new and efficient method for synthesizing selected

Correspondence to: Atef Abdel-monem Abdel-hafez, Department of Pharmaceutical Medicinal Chemistry, Faculty of Pharmacy, Assiut University, Assiut 71526, Egypt
Tel: 20-88-0230-3006, Fax: 20-88-0233-2776
E-mail: atef@aun.edu.eg

pyrimido[1,2-a]benzimidazole derivatives (**5-19**) by the one-pot two-component thermal condensation reaction and the evaluation of the anticancer properties of these derivatives.



(A)

MATERIALS AND METHODS

Melting points were determined on an electrothermal melting point apparatus and were uncorrected. Thin layer chromatography (TLC) was carried out on precoated silica gel TLC plates (Merck, 60 F_{254}, 2.5 × 5.0 cm, 0.25 mm layer thickness) and the spots were detected under UV light. IR spectra were recorded as KBr discs on a Shimadzu IR 200-91527 spectrophotometer. ^{1}H-NMR spectra were measured on a JNM-GX 400 FT NMR spectrophotometer (JEOL Co., Tokyo, Japan) and all chemical shifts are given in δ ppm relative to tetramethylsilane (TMS). Electron impact (EI) mass spectra were measured with JEOL-JMS-AX 505 spectrophotometer at an ionization voltage of 70 eV. Elemental analyses were performed at the microanalytical center of Toyama Medical and Pharmaceutical University, Toyama, Japan. All chemicals used are of analytical grade.

General method for the synthesis of 4-substituted-1,2,3,4-tetrahydrobenzo[4,5]imidazo[1,2-a]pyrimidin-2-one derivatives (5-7)

2-Aminobenzimidazole **1** (1 g) and 1.1 eq. of the appropriate methyl cinnamate (**2-4**) were added to a 100 mL round-bottomed flask equipped with a reflux condenser. The reaction mixture was heated for 10-15 min at 180-200°C till fusion. Before solidification, benzene (50 mL) was added to the reaction mixture to form a slurry. The precipitate was filtered, washed with benzene and the crude material was crystallized from the appropriate solvent (Table I). The physical constants are listed in Table I.

4-Phenyl-1,2,3,4-tetrahydrobenzo[4,5]imidazo[1,2-a]pyrimidin-2-one (5)

IR (KBr) υ cm^{-1}: 3405, 2815, 1663, 1629, 1595. ^{1}H-NMR (DMSO-d_6): 2.91 (1H, dd, *J* = 16.6, 3.2 Hz, H_a-3); 3.51 (1H, dd, *J* = 16.6, 7.3 Hz, H_b-3); 5.93 (1H, dd, *J* = 7.3, 3.2 Hz, H4); 6.82~7.46 (8H, m, Ar-H); 11.59 (1H, brs, NH). EI-MS *mlz* [M$^+$] 263.

4-(4-Methoxyphenyl)-1,2,3,4-tetrahydrobenzo[4,5]imidazo[1,2-a]pyrimidin-2-one (6)

IR (KBr) υ cm^{-1}: 3408, 2805, 1665, 1627, 1590. ^{1}H-NMR (DMSO-d_6): 2.89 (1H, dd, *J* = 16.6, 4.0 Hz, H_a-3); 3.43 (1H, dd, *J* = 16.4, 6.8 Hz, H_b-3); 3.71 (3H, s, OCH_3); 5.86 (1H, dd, *J*= 16.4, 6.8 Hz, H-4); 6.84~7.45 (8H, m, Ar-H); 11.70 (1H, brs, NH). EI-MS *mlz* [M$^+$] 293.

Table I. Physical data of 3,4-dihydro- and 1,2,3,4-tetrahydro-benzo[4,5]imidazo[1,2-a]pyrimidine derivatives (**5-19**)

Compound	Mp (°C)	Yield (%)	Crystallization solvent	Molecular formula[a] (Molecular weight)
5	297-298	96	$CHCl_3$/MeOH	$C_{16}H_{13}N_3O$ (263.29)
6	278-280	91	$CHCl_3$/MeOH	$C_{17}H_{15}N_3O_2$ (293.32)
7	216-218	95	$CHCl_3$/MeOH	$C_{16}H_{12}N_4O_3$ (308.29)
8	>300	75	$CHCl_3$/EtOH	$C_{17}H_{12}ClN_3O$ (309.75)
9	>300	79	$CHCl_3$/EtOH	$C_{18}H_{14}ClN_3O_2$ (339.78)
10	>300	71	$CHCl_3$/EtOH	$C_{17}H_{11}ClN_4O_3$ (354.75)
11	223-225	79	DMF	$C_{22}H_{22}N_4O$ (358.44)
12	189-190	83	DMF	$C_{23}H_{24}N_4O_2$ (388.46)
13	241-242	86	DMF	$C_{22}H_{21}N_5O_3$ (403.43)
14	209-210	93	DMF/dioxane	$C_{21}H_{20}N_4O_2$ (360.41)
15	252-253	89	DMF/dioxane	$C_{22}H_{22}N_4O_3$ (390.44)
16	272-273	84	DMF/dioxane	$C_{21}H_{19}N_5O_4$ (405.41)
17	229-230	81	DMF	$C_{22}H_{23}N_5O$ (373.45)
18	186-187	76	DMF	$C_{23}H_{25}N_5O_2$ (403.48)
19	173-174	78	DMF	$C_{22}H_{22}N_6O_3$ (418.45)

[a]Analyzed for C, H and N; results were within ± 0.4% of the theoretical values for the formulae given.

4-(4-Nitrophenyl)-1,2,3,4-tetrahydrobenzo[4,5]imidazo[1,2-a]pyrimidin-2-one (7)

IR (KBr) υ cm^{-1}: 3415, 2835, 1670, 1630, 1600. ^{1}H-NMR (DMSO-d_6): 2.97 (1H, dd, *J*= 17.0, 1.7 Hz, H_a-3); 3.62 (1H, dd, *J* = 16.6, 7.3 Hz, H_b-3); 6.16 (1H, m, H-4); 7.0 ~8.2 (8H, m, Ar-H); 11.87 (1H, brs, NH). EI-MS *mlz* [M$^+$] 308.

General method for synthesis of 2-chloro-4-substituted-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidin-3-carboxaldehyde derivatives (8-10)

Phosphorous oxychloride (0.017 mole) was gradually added to a stirred suspension of the appropriate benzo [4,5]imidazo[1,2-a]pyrimidin-2-one derivatives **5-7** (0.0075 mole) in dimethylformamide (20 mL) and the reaction mixture was stirred at 80°C for 2-3 h. After cooling and the addition of water, the product was filtered, washed with water and the crude material was crystallized from the appropriate solvent (Table I). The physical constants are listed in Table I.

2-Chloro-4-phenyl-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxaldehyde (8)

IR (KBr) υ cm^{-1}: 3435, 2860, 1695, 1638, 1606, 1580. ^{1}H-

NMR (DMSO-d_6): 3.78 (1H, d, *J* = 7.08 Hz, H-3); 6.53 (1H, brs, H-4); 7.09~8.24 (8H, m, Ar-H); 9.62 (1H, s,-CHO). El-MS *mlz* [M^+] 309.

2-Chloro-4-(4-methoxyphenyl)-3,4-dihydrobenzo[4,5] imidazo[1,2-a]pyrimidine-3-carboxaldehyde (9)
IR (KBr) υ cm^{-1}: 3450, 2875, 1697, 1640, 1605, 1578. ^{1}H-NMR (DMSO-d_6): 3.94 (1H, brs, H-3); 3.75 (3H, s, OCH_3); 6.52 (1H, brs, H-4); 7.60~8.53 (8H, m, Ar-H); 9.62 (1H, s,-CHO). El-MS *mlz* [M^+] 339.

2-Chloro-4-(4-nitrophenyl)-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimoidine-3-carboxaldehyde (10)
IR (KBr) υ cm^{-1}: 3460, 2885, 1708, 1645, 1610, 1590. ^{1}H-NMR (DMSO-d_6): 3.94 (1H, brs, H-3); 6.60 (1H,brs, H-4); 7.06~8.16 (9H, m, Ar-H); 9.63 (1H, s,-CHO). El-MS *mlz* [M^+] 354.

General method for synthesis of substituted-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidin-3-carboxaldehyde derivatives (11-19)

A mixture of the targeted 2-chloro-4-substituted-3,4-dihydrobenzo[4,5[imidazo[1,2-a]pyrimidine-3-carboxyaldehyde derivative **8-10** (0.004 mole) with the appropriate secondary amine (0.010 mole) in dimethylformamide (10 mL) was stirred at 60°C for 1 h. After cooling, the product was filtered, washed with water and the crude material was crystallized from the appropriate solvent (Table I). The physical constants are listed in Table I.

4-Phenyl-2-(piperidin-1-yl)-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxaldehyde (11)
IR (KBr) υ cm^{-1}: 3425, 2920, 1693, 1650, 1625, 1605. ^{1}H-NMR (DMSO-d_6): 1.67 (6H, m,-CH_2-CH_2-CH_2- of piperidino); 3.11 (4H, m, -CH_2-N-CH_2- of piperidino); 3.54 (1H, dd, *J* = 16.60, 7.32 Hz, H-3); 5.87 (1H, dd, *J* = 7.36, 3.68 Hz, H-4); 7.08~7.71 (9H, Ar-H); 9.03 (1H, brs, CHO). El-MS *mlz* [M^+] 358.

4-(Methoxyphenyl)-2-(piperidin-1-yl)-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxaldehyde (12)
IR (KBr) υ cm^{-1}: 3440, 2945, 1700, 1660, 1625, 1610. ^{1}H-NMR (DMSO-d_6): 1.41 (2H, m,-CH_2-C<u>H_2</u>-CH_2- of piperidino); 1.53 (4H, m,-C<u>H_2</u>-CH_2-C<u>H_2</u>- of piperidino); 3.32 (4H, m, -CH_2-N-CH_2- of piperidino); 3.52 (1H, dd, *J* = 16.60, 7.32 Hz, H-3); 3.71 (3H, s, CH_3); 5.64 (1H, dd, *J* = 7.08, 3.68 Hz, H-4); 7.11~7.79 (8H, m, Ar-H); 9.12 (1H, brs, CHO). El-MS *mlz* [M^+] 388.

4-(Nitroyphenyl)-2-(piperidin-1-yl)-3,4-dihydrobenzo[4,5] imidazo[1,2-a]pyrimidine-3-carboxaldehyde (13)
IR (KBr) υ cm^{-1}: 3485, 2940, 1705, 1658, 1629, 1610. ^{1}H-NMR (DMSO-d_6): 1.43 (2H, m,-CH_2-C<u>H_2</u>-CH_2- of piperidino); 1.56 (4H, m,-C<u>H_2</u>-CH_2-C<u>H_2</u>- of piperidino); 3.38 (4H, m, -CH_2-N-CH_2- of piperidino); 3.62 (1H, dd, *J* = 16.60, 7.32 Hz, H-3); 5.93 (1H, dd, *J* = 7.36, 3.68 Hz, H-4); 7.11~8.24 (8H, m, Ar-H); 9.54 (1H, brs, CHO). El-MS *mlz* [M^+] 403.

2-Morpholino-4-phenyl-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxaldehyde (14)
IR (KBr) υ cm^{-1}: 3428, 2935, 1695, 1645, 1630, 1600. ^{1}H-NMR (DMSO-d_6): 3.47 (4H, t, *J* = 16.42 Hz, -CH_2-N-CH_2- of morpholino); 3.55 (1H, dd, *J* = 16.42, 7.32 Hz, H-3); 3.88 (4H, t, *J* = 16.42 Hz, -CH_2-O-CH_2- of morpholino); 5.87 (1H, dd, *J* = 7.12, 3.68 Hz, H-4); 7.09~7.74 (8H, m, Ar-H); 9.02 (1H, brs, CHO). El-MS *mlz* [M^+] 360.

2-Morpholino-4-(4-Methoxyphenyl)-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxaldehyde (15)
IR (KBr) υ cm^{-1}: 3450, 2950, 1705, 1640, 1628, 1605. ^{1}H-NMR (DMSO-d_6): 3.31 (4H, t, *J* = 16.42 Hz, -CH_2-N-CH_2- of morpholino); 3.51 (1H, dd, *J* = 16.42, 7.32 Hz, H-3); 3.72 (3H, s, CH_3); 3.73 (4H, t, *J* = 16.42 Hz, -CH_2-O-CH_2- of morpholino); 5.67 (1H, dd, *J* = 7.08, 3.68 Hz, H-4); 7.14~7.82 (8H, m, Ar-H); 9.13 (1H, brs, CHO). El-MS *mlz* [M^+] 390.

2-Morpholino-4-(4.nitrophenyl)-3,4-dihydrobenzo[4,5] imidazo[1,2-a]pyrimidine-3-carboxaldehyde (16)
IR (KBr) υ cm^{-1}: 3480, 2935, 1710, 1648, 1630, 1610. ^{1}H-NMR (DMSO-d_6): 3.35 (4H, t, *J* = 16.42 Hz, -CH_2-N-CH_2- of morpholino); 3.63 (1H, dd, *J* = 16.42, 7.32 Hz, H-3); 3.77 (4H, t, *J* = 16.42 Hz, -CH_2-O-CH_2- of morpholino); 5.98 (1H, dd, *J* = 7.12, 3.68 Hz, H-4); 7.28~8.15 (8H, m, Ar-H); 9.62 (1H, brs, CHO). El-MS *mlz* [M^+] 405.

2-(4-Methylpiperazin-1-yl)-4-phenyl-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxaldehyde (17)
IR (KBr) u cm^{-1}: 3430, 2930, 1690, 1635, 1625, 1600. ^{1}H-NMR (DMSO-d_6): 2.32 (3H, s, N-CH_3); 2.49 (4H, t, *J* = 16.54 Hz, -C<u>H_2</u>-N(CH_3)-C<u>H_2</u>-); 3.13 (4H, t, *J* = 16.54 Hz, -CH_2-N-CH_2-); 3.55 (1H, dd, *J* = 16.48, 7.32 Hz, H-3); 5.86 (1H, dd, *J* = 7.32, 3.68 Hz, H-4); 7.07~7.70 (8H, m, Ar-H); 9.02 (1H, brs, CHO). El-MS *mlz* [M^+] 373.

4-(4-Methoxyphenyl)-2-(4-methylpiperazin-1-yl)-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxaldehyde (18)
IR (KBr) υ cm^{-1}: 3445, 2940, 1700, 1640, 1625, 1600. ^{1}H-NMR (DMSO-d_6): 2.18 (3H, s, N-CH_3); 2.30 (4H, t, *J* = 16.54 Hz, -C<u>H_2</u>-N(CH_3)-C<u>H_2</u>-); 3.43 (4H, t, *J* = 16.54 Hz, -CH_2-N-CH_2-); 3.48 (1H, dd, *J* = 16.48, 7.32 Hz, H-3); 5.85 (1H, dd, *J* = 7.08, 3.68 Hz, H-4); 7.06~7.69 (8H, m, Ar-H); 9.12 (1H, brs, CHO). El-MS *mlz* [M^+] 403.

2-(4-Methylpiperazin-1-yl)-4-(4-nitrophenyl)-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxaldehyde (19)

IR (KBr) υ cm^{-1}: 3480, 2930, 1700, 1645, 1625, 1605. ^{1}H-NMR (DMSO-d_6): 2.41 (3H, s, N-CH_3); 2.50 (4H, t, *J* = 16.54 Hz, -C<u>H</u>$_2$-N(CH_3)-C<u>H</u>$_2$-); 3.50 (4H, t, *J* = 16.54 Hz, -CH_2-N-CH_2-); 3.58 (1H, dd, *J* = 16.48, 7.32 Hz, H-3); 5.90 (1H, dd, *J* = 7.32, 3.68 Hz, H-4); 7.08~8.11 (8H, m, Ar-H); 9.56 (1H, brs, CHO). EI-MS *m/z* [M^+] 418.

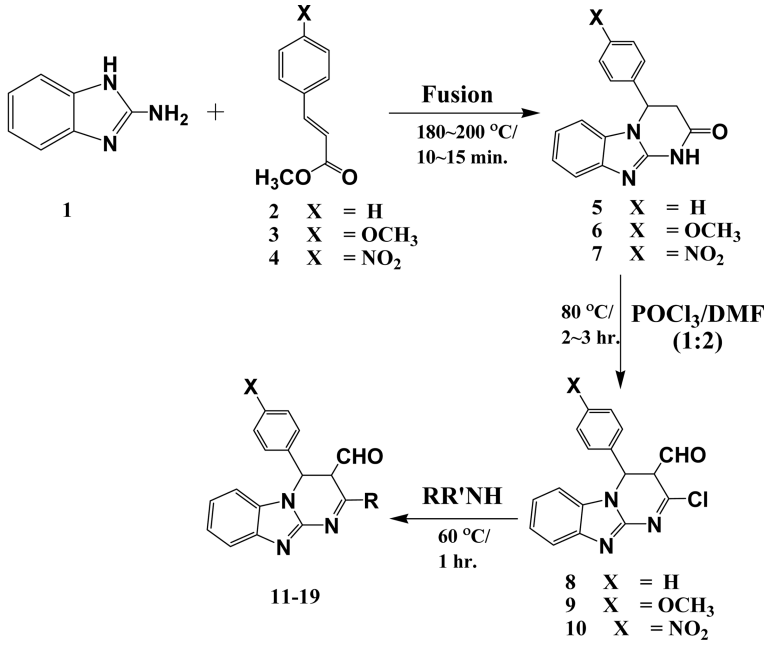
RESULTS AND DISCUSSION

Chemistry

The synthetic strategy for the synthesis to the target derivatives (**5**-**19**) is depicted in Scheme 1. Two-component condensation reactions are powerful tools for the synthesis of organic compounds, since the products are formed in a single step and diversity can be achieved by simply varying each component. In recognition of the interest in developing new approaches to the synthesis of a variety of heterocyles incorporating a benzimidazole moiety from readily obtainable inexpensive starting materials, 2-aminobenzimidazole **1** and *P*-substituted methyl cinnamates (**2**-**4**) were utilized as building blocks for a possible new route to design the pyrimido[1,2-a]benzimidazole tricyclic system. 1,2,3,4-Tetrahydrobenzo[4,5]imidazo [1,2-a]pyrimidine-2-one derivatives (**5**-**7**) were prepared via a thermal condensation reaction of 2-aminobenzoimidazole **1** and *P*-substituted methyl cinnamates (**2**-**4**) at 180-200°C. Vilsmir-Haack formylation of these derivatives (**5**-**7**) using two molar equivalent amounts of phosphorus oxychloride afforded the 2-chloro-3-carboxaldehyde targets (**8**-**10**). Nucleophilic displacement of the chloro atom in the 3-carboxaldehyde compounds (**8**-**10**) using secondary amines yielded the remaining final targets (**11**-**19**). The structure of the synthesized derivatives (**5**-**19**) was confirmed by means of IR, ^{1}H-NMR, MS and elemental analyses. The IR spectra of azinazoles (**5**-**19**) contain stretching absorption bands of C=O, C=N and C=C bonds. ^{1}H-NMR spectrum of compounds (**8**-**19**) exhibited a signal at δ 9.02-9.63 recognized as arising from aldehydic protons, in addition to characteristic signals for H-3 and H-4 protons. A broad singlet at δ 11.59-11.87 assigned to NH proton appeared at ^{1}H-NMR spectra of 1,2,3,4-tetrahydrobenzo[4,5]imidazo[1,2-a]pyrimidine-2-one derivatives (**5**-**7**). A multiplet appeared at δ 6.82-8.53 for the aromatic protons of all derivatives (**5**-**19**). The mass spectra of synthesized compounds (**5**-**19**) displayed molecular ion peaks at appropriate *m/z* values.

Antineoplastic screening

The designed targets (**5**-**19**) were selected by NCI to be primary screened for their in vitro antitumor activity according to the standard protocol of NCI (Bethesda, MD, U.S.A.; Gray and Wickstrom, 1996; Monks *et al.*, 1991). The Development Therapeutic Program (DTP), Division of Cancer Treatment and Diagnosis (DCTD) in NCI selected a three cell line panel consisting of MCF7 (breast), NCI-H460 (lung), and SF-268 (CNS) for the primary anticancer assay. Compounds which reduce the growth of any one of



Compd. No.	X	R
11	H	Piperidin-1-yl
12	OCH_3	Piperidin-1-yl
13	NO_2	Piperidin-1-yl
14	H	Morpholin-1-yl
15	OCH_3	Morpholin-1-yl
16	NO_2	Morpholin-1-yl
17	H	4-Methylpiperazin-1-yl
18	OCH_3	4-Methylpiperazin-1-yl
19	NO_2	4-Methylpiperazin-1-yl

Scheme 1. Pathway for the synthesis of the target compounds (**5**-**19**)

Table II. Inhibition of *in vitro* cancer cell lines by compounds **8**, **10**, and **18**

Compound	NCI number	Panel	Cell lines (Cytotoxicity:$\log_{10}GI_{50}$, M)[a,b]
8	722731	Leukemia	SR (-6.06)
		Renal cancer	A498 (-5.52)
		Breast cancer	HS 578T (-6.13)
10	722743	Leukemia	CCRF-CEM (-5.51)
			HL-60 (TB) (-5.48)
			K-562 (-5.74)
			MOLT-4 (-5.77)
			RPMI-8226 (-5.42)
			SR (-5.67)
		Lung cancer	HOP-62 (-5.08)
			HOP-92 (-5.26)
			NCI-H226 (-5.48)
			NCI-H23 (-5.10)
			NCI-H522 (-5.43)
		Colon cancer	COLO 205 (-5.50)
			HCT-116 (-5.43)
			HCT-15 (-5.53)
			SW-620 (-5.32)
		CNS	SF-268 (-5.16)
			SNB-75 (-5.75)
			U251 (-5.26)
		Melanoma	LOX IMVI (-5.66)
			MALME-3M (-5.09)
			M14 (-5.10)
			SK-MEL-2 (-5.22)
			SK-MEL-28 (-5.60)
			SK-MEL-5 (-5.18)
		Ovarian cancer	OVCAR-3 (-5.41)
			OVCAR-8 (-5.40)
		Renal cancer	780-0 (-5.37)
			ACHN (-5.61)
			CAKI-1 (-5.51)
			RXF-393 (-5.53)
			TK-10 (-5.34)
			UO-31 (-5.41)
		Prostate cancer	PC-3 (-5.31)
			DU-145 (-5.32)
		Breast cancer	MCF7 (-5.40)
			MDA-MB-435 (-5.32)
			BT-549 (<-8.00)
			T-47D (-5.80)
18	722739	Leukemia	CCRF-CEM (-5.64)
			HL-60 (TB) (-5.68)
			K-562 (-5.97)
			MOLT-4 (<-8.00)
			RPMI-8226 (-5.63)
			SR (-7.42)
		Lung cancer	HOP-92 (-5.32)
		Ovarian cancer	OVCAR-4 (-5.09)

[a] Data obtained from NCI *in vitro* disease-oriented tumor cell line screen.
[b] GI_{50} = compound concentration that inhibits 50% cell growth.

the cell lines to approximately 32% or less are passed on for evaluation in the full panel of 60 human tumor cell lines over a 5-log dose-range. Leukemia, lung, colon, central nervous system, melanoma, ovary, renal, prostate and breast cancer were the nine clinically isolated cancer subpanels represented by 60 human tumor cell lines. In the three cell line one dose primary anticancer assay, 2-chloro-4-phenyl-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxyaldehyde (**8**, NCI 722731), 2-chloro-4-(4-nitrophenyl)-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxyaldehyde (**10**, NCI 722743) and 4-(4-methoxyphenyl)-2-(4-methylpiperazin-1-yl)-3,4-dihydro-benzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxaldehyde (**18**, NCI 722739) were passed on for evaluation against 60 human cell lines. The results (Table I) indicated that pyrimidobenzimidazole candidate (**10**, NCI 722743) displayed good activity against all cell lines tested at $\log_{10} GI_{50}$ (M) ranging from -5.08 to < -8.00 (Fig 1). The analogue (**18**, NCI 722739) exhibited less cytotoxic activity against a few cell lines from leukemia, lung, and ovarian panels at $\log_{10} GI_{50}$ (M) ranging from -5.09 to < -8.00. In contrast, compound (**8**, NCI 722731) was active against the cell lines of leukemia, renal, and breast panels at $\log_{10} GI_{50}$ (M) ranging from -5.52 to - 6.13. All the remaining candidates (**5**-**7**, **9**, **11**-**17**, **19**) were inactive. Among this series of compounds, the intermediate (**10**, NCI 722743) appears to be the most potent, as it showed high selectivity against the BT-549 cell line from breast at $\log_{10} GI_{50}$ (M) <-8.00. Compound (**8**, NCI 722731) showed selectivity against the SR cell line from leukemia at $\log_{10} GI_{50}$ (M) <-6.06 and against the HS 578T cell line from breast at $\log_{10} GI_{50}$ (M) <-6.13. The candidate (**18**, NCI 722739) exhibited selectivity against MOLT-4, SR and K-562 cell lines from leukemia at $\log_{10} GI_{50}$ (M) <-8.00, -7.42 and -5.97, respectively.

CONCLUSIONS

A novel two-component thermal condensation reaction of 2-amionbenzimidazole **1** and *p*-substituted methyl cinnamates **2**-**4** has been developed for the efficient synthesis of pyrimido[1,2-a]benzimidazoles ring systems. It is noteworthy that manipulation of the tricyclic system pyrimidobenzimidazole led to the development of promising anticancer agents. 2-Chloro-4-phenyl-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxyaldehyde (**8**, NCI 722731) and 4-(4-methoxyphenyl)-2-(4-methylpiperazin-1-yl)-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxaldehyde (**18**, NCI 722739) showed a variable degree of antineoplastic activity against some of the cell lines tested. 2-Chloro-4-(*p*-nitrophenyl)-3,4-dihydrobenzo[4,5]imidazo[1,2-a]pyrimidine-3-carboxyaldehyde (**10**, NCI 722743) exhibited good *in vitro* antineoplastic activity with subpanel disease selectivity.

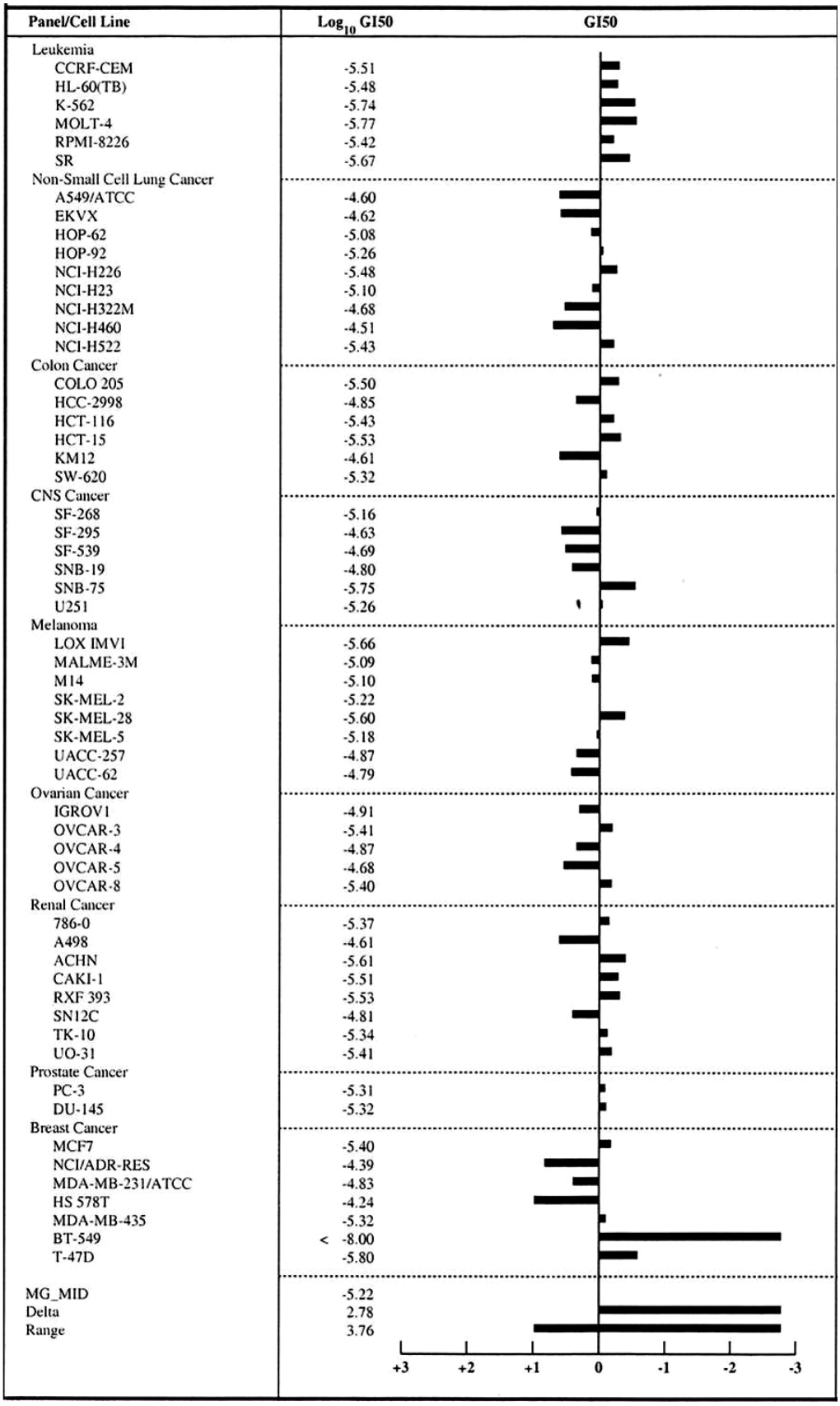


Fig. 1. The mean graphs page shows mean graphs of the principle response parameter GI_{50} from screening of compound **10** (NCI 722743)

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