Synthesis and Evaluation of Adenosine Antagonist Activity of a Series of [1,2,4]Triazolo[1,5-*c*]quinazolines

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A series of [1,2,4]triazolo[1,5-*c*]quinazolines were prepared in satisfactory yields by reaction of some derivatives of 2-aminobenzohydrazide with several hydrochlorides of aromatic amidines, and their binding affinities for the recombinant human adenosine A_{2A} and A_{2B} receptors were determined. None of the new compounds showed noteworthy affinity for these receptors, though a very high affinity for the A_{2A} receptor and, consequently, a high level of A_{2A}/A_{2B} selectivity was revealed for one of the synthesized compounds.

Key words triazoloquinazoline; hydrazide; synthesis; heterocycle; azo-compound

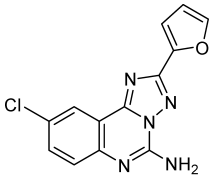
Although changes induced by xanthic bases in intracellular AMPc in respiratory tissue were initially attributed to other mechanisms, it is currently accepted that the changes are due to the competitive antagonism of these bases with receptors of the neurotransmitter adenosine.[1,2] Adenosine exerts a depressor effect with important repercussions in the functioning of the cardiovascular, renal, immune and central nervous systems. In recent years the search for series of adenosine analogues that show agonistic or antagonistic properties against adenosine receptors has intensified. These receptors belong to a superfamily of rhodopsin-like, G protein-coupled receptors (GPCRs), of which four subtypes are known and designated as A_1, A_{2A}, A_{2B} and A_3. The receptors modulate the activity of adenylate cyclase either by stimulation (A_{2A} and A_{2B}) or inhibition (A_1 and A_3) of the activity.[3] The receptors are widely distributed within organisms and are expressed in different densities at diverse cells.[2] All of the subtypes have been cloned, and each receptor can therefore be studied individually by use of recombinant systems. The A_1 and A_{2A} receptors show a high affinity for adenosine and are activated by extremely low concentrations (nanomolar) of the neurotransmitter, whereas receptors A_{2B} and A_3 show low affinity for adenosine and are only activated when the levels reach micromolar concentrations, which occurs during periods of hypoxia, inflammation or ischemia.[4,5]

In the past decade a large body of experimental evidence has been obtained that supports the idea that adenosine plays an important role in allergic asthma.[6—9] Patients with asthma and bronchitis show higher concentrations of adenosine in bronchoalveolar fluid than control subjects, suggesting that adenosine may be a marker of pulmonary inflammation. In *in vivo* assays, both inhaled adenosine and its precursor 5-adenosine monophosphate (AMP) cause bronchoconstriction in atopic and asthmatic patients, but not in normal control subjects. Furthermore, dipyridamole, which is an inhibitor of adenosine recapture, increases bronchospasms induced by adenosine in asthma sufferers, an effect that may be inhibited by theophylline, an antagonist of adenosine receptors.[10]

The presence of A_{2B} receptors has been demonstrated in bronchoalveolar mastocytes, which supports the hypothesis that adenosine participates in the physiopathology of asthma, through activation of these receptors.[8,11] This allows proposal of A_{2B} receptor antagonists as potential antiasthmatic drugs.

Present study was carried out as part of a line of investigation directed at finding potential inverse antagonist or agonists of the A_{2B} receptor, and on the basis of the strong activity reported for triazoloquinazoline CGS 15943 as an adenosine antagonist,[12] the study was focused on the synthesis of new adenosine analogues of this type, which would contribute to the characterization and validation of the A_{2B} receptor as a drug target; the structure–activity relationships (SAR) in A_{2A} and A_{2B} receptor binding assays were also compared.

A series of triazoloquinazolines **4** was synthesized by cyclization of the corresponding [1,2,4]triazoles **3** with cyanamide,[12] or with triethyl orthoformate in the case of **4j** (Chart 1). The required and amino hydrazides **2a**—**d**[13] were prepared in excellent yields (88—93%), by treatment of amino esters **1a**—**d** with hydrazine hydrate; subsequent reaction of **2a**—**d** with the amidine hydrochlorides **6** in an



CGS 15943

Fig. 1

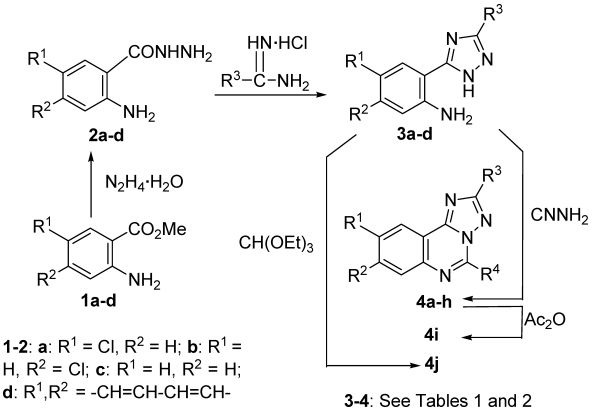


Chart 1

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ethanolic $NaOCH_3$ solution produced triazoles **3**.

Previously, the amidine hydrochlorides **6** were obtained from the corresponding imino ethers, which were prepared in turn by nucleophilic addition of methanol to the respective nitrile, with $NaBH_4$ in anhydrous MeOH being used as the basic promoter (Chart 2). Due to the reversibility of this reaction,[17,18] in those cases in which formation of the imino ether was not well favoured, imino ethers were obtained as their hydrochlorides, **5iii**—**v**, by passing a stream of dry hydrogen chloride through a solution of the nitrile in anhydrous methanol.[17] Finally iminoesters **5i**—**ii** were treated with ammonium chloride or, alternatively, hydrochlorides **5iii**—**v** were treated with a stream of ammonia, to provide amidine hydrochlorides in good yields (60—89%).

The affinities of compounds **3a**, **3d**, **3e**, and **4b**—**h** for human adenosine receptors A_{2A} and A_{2B} were determined by binding competition assays, using CGS 15943 (**4a**) as a reference standard. Results are shown in Table 3. Affinity of compounds that did not fully displace radioligand binding to the receptor is given only in terms of percentage of displacement.

The need of the triazoloquinazoline scaffold for adenosine-like activity was confirmed, the intermediate triazoles **3a**, **3d** and **3e** showed weak or nil affinity for both receptors.

In the series of triazoloquinazolines **4**, change in the location of the chlorine atom from position 9 to 8 leads to a compound, **4b**, for which there is a drastic reduction in the affinities for the receptors examined, whereas suppression of that substituent, **4c**, or replacement of the benzene ring by a naphthalene ring, **4d**, lead to compounds for which the A_{2A}/A_{2B} selectivity has been outstandingly increased, with compound **4c** showing a noteworthy 26-fold increase in the affinity for the A_{2A} receptor, with regard to the reference standard CGS 15943.

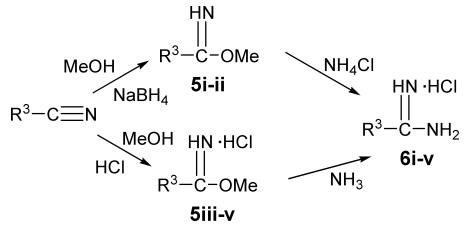
Table 1. Chemical Structures, Yields and Melting Point of Triazole Derivatives **3**

Compound	R_1	R_2	R_3	Yield (%)	mp (°C)
3a	Cl	H	2-C_4H_3O	77	248—249[12,14]
3b	H	Cl	2-C_4H_3O	58	249—251
3c	H	H	2-C_4H_3O	89	234—236[14]
3d	–CH=CH–CH=CH–		2-C_4H_3O	60	232—234
3e	Cl	H	2-C_5H_4N	68	233—235[15]
3f	Cl	H	2-C_4H_3S	65	252—255
3g	Cl	H	C_6H_5	66	240—243[14,15]
3h	Cl	H	3,4-$C_6H_3(OCH_3)_2$	82	212—214

Replacement of the furan ring in position 2 by other aromatic rings, **4e**—**h**, lead to a decrease in the affinity for both receptors, which is moderate in the case of **4f** and **4g** and much more acute for **4e** and **4h**. Conversion of the primary amine into its acetyl derivative, **4i**, or its mere suppression, **4j**, also produced a decrease in the affinity for both receptors.

Experimental

All chemicals were of reagent grade and were obtained from Aldrich Chemical Co. and used without further purification. For preparation of the hydrazides **2a**—**d**, a modified version of the method of Dacroix *et al.*[13] was followed, in which only hydrazine hydrate was used as solvent. Transforma-



i: R^3 = 2-furyl; ii: R^3 = 2-pyridyl; iii: R^3 = 2-thienyl; iv: R^3 = phenyl; v: R^3 = 3,4-dimethoxyphenyl

Chart 2

Table 3. Affinity (Percentage of Displacement at 1 μM and pK_i) of Prepared Compounds for Human Adenosine A_{2A} and A_{2B} Receptors.

Compound	Receptor			
	A_{2A}		A_{2B}	
	% displacement (1 μM)	pK_i	% displacement (1 μM)	pK_i
3a	17±4	n.c.[a]	5±2	n.c.
3d	20±2	n.c.	12±3	n.c.
3e	0±3	n.c.	0±1	n.c.
4a (CGS 15943)	100±2	8.38±0.20	91±3	7.68±0.13
4b	0±3	n.c.	18±3	n.c.
4c	100±2	9.80±0.22	85±4	7.12±0.09
4d	94±4	7.80±0.10	56±2	5.95±0.16
4e	0±2	n.c.	10±2	n.c.
4f	65±2	6.66±0.09	57±4	6.22±0.21
4g	53±6	6.44±0.11	42±4	5.85±0.17
4h	56±3	n.c.	18±4	n.c.
4i	73±2	6.50±0.08	51±3	5.75±0.14
4j	27±6	n.c.	43±3	5.76±0.11

Values represent the mean±S.E.M. of three independent assays. *a*) n.c.: not calculated due to low affinity of the compound.

Table 2. Chemical Structures, Yields and Melting Point of Triazoloquinazolines **4**

Compound	R_1	R_2	R_3	R_4	Yield (%)	mp (°C)
4a (CGS 15943)	Cl	H	2-C_4H_3O	NH_2	93	278—279[12,14]
4b	H	Cl	2-C_4H_3O	NH_2	47	275—277
4c	H	H	2-C_4H_3O	NH_2	78	290—293[12,14]
4d	–CH=CH–CH=CH–		2-C_4H_3O	NH_2	82	309—311
4e	Cl	H	2-C_5H_4N	NH_2	87	207—209
4f	Cl	H	2-C_4H_3S	NH_2	56	290—292[16]
4g	Cl	H	C_6H_5	NH_2	81	296—298[13]
4h	Cl	H	3,4-$C_6H_3(OCH_3)_2$	NH_2	57	237—239
4i	H	H	2-C_4H_3O	$NHCOCH_3$	96	206—208
4j	Cl	H	2-C_4H_3O	H	68	249—251

tion of iminoesters **5**, into amidines **6**, was achieved by following literature procedures.[17] Melting points were measured in a Reicher Kofler Thermopan and are uncorrected. Infrared spectra were recorded in a Perkin-Elmer 1640 FTIR spectrophotometer. The ^{1}H- and ^{13}C-NMR spectra were recorded in a Bruker AMX 300 spectrometer at 300 and 74 MHz, respectively, using TMS as internal standard (chemical shifts in *d* values, *J* in Hz). Mass spectra were recorded on a Kratos MS-59 spectrometer. Microanalyses were performed in a Perkin-Elmer 240B Elemental Analyser at the Microanalyses Service of the University of Santiago; all results shown were within ±0.4% of the theoretical values. All air-sensitive reactions were carried out under argon. Flash chromatography was performed on silica gel (Merck 60, 230—240 mesh) and analytical TLC on pre-coated silica gel plates (Merck 60 F_{254}, 0.25 mm).

Binding competition assays at A_{2A} and A_{2B} receptors were performed *in vitro* at human A_{2A} and A_{2B} receptors transfected in HeLa and HEK-293 cells, respectively, as previously described.[19]

Production of Triazoles 3a—h. General procedure[12] Sodium methoxide (1.1 mmol) dissolved in absolute ethanol (2.5 ml) was added to a solution of amidine hydrochloride (1 mmol) in absolute ethanol (3 ml). The mixture was stirred for 10 min under argon, then it was filtered, and the filtrate was added to a solution of the hydrazide **2** (1.1 mmol) in chlorobenzene (5 ml) and absolute ethanol (3 ml), kept at 70 °C, in an flask fitted with a Dean-Stark trap. Bath temperature was raised to 110 °C over 1 h and thereafter to 130 °C. Once distillation ceased, the water separator was removed, and a compensating addition funnel containing 4-A molecular sieves was inserted between the flask and the condenser. More chlorobenzene (2.5 ml) was added, and the mixture was heated overnight at reflux under argon. The mixture was filtered while hot, and the filtrate was cooled, left at 5 °C for 24 h, and the separated solid was collected by filtration.

5-(2-Amino-5-chlorophenyl)-3-(2-furyl)-1*H*-1,2,4-triazole (3a) Physical and spectroscopic data were consistent with data in the existing literature.[12]

5-(2-Amino-4-chlorophenyl)-3-(2-furyl)-1*H*-1,2,4-triazole (3b) White solid, mp 249—251 °C, from toluene. ^{1}H-NMR (DMSO-d_6) δ: 8.07 (1H, s, 5-H_{furyl}), 7.74 (1H, d, J=8.5 Hz, H_{phenyl}), 7.45 (1H, d, J=3.3 Hz, H_{furyl}), 6.98 (3H, br s, two D_2O exch., NH_2+3H_{phenyl}), 6.81 (1H, dd, J=3.5, 1.7 Hz, H_{furyl}), 6.71 (1H, dd, J=8.5, 2.0 Hz, 6H_{phenyl}). *Anal.* Calcd for $C_{12}H_8ClN_4O$: C, 55.29; H, 3.48; N, 21.49. Found: C, 55.52; H, 3.33; N, 21.33.

5-(2-Aminophenyl)-3-(2-furyl)-1*H*-1,2,4-triazole (3c) White solid, mp 234—236 °C, from toluene, (lit. 210—212 °C, from ethanol).[14] ^{1}H-NMR (DMSO-d_6) δ: 9.43 (1H, br s, D_2O exch., NH), 7.82 (1H, d, J=8.20 Hz, H_{phenyl}), 7.72 (1H, s, 5-H_{furyl}), 7.19 (1H, d, J=8.1 Hz, H_{phenyl}), 7.10 (1H, d, J=3.3 Hz, H_{furyl}), 6.88 (1H, d, J=8.1 Hz, H_{phenyl}), 6.75 (1H, dd, J=8.0, 7.2 Hz, H_{phenyl}), 6.65 (1H, dd, J=3.3, 1.8 Hz, H_{furyl}).

5-(3-Amino-2-naphtyl)-3-(2-furyl)-1*H*-1,2,4-triazole (3d) White solid, mp 232—234 °C. IR (KBr) cm^{-1}: 3468, 3372, 3040, 1630, 1607, 1223, 1051, 743. ^{1}H-NMR (DMSO-d_6) δ: 14.3 (1H, br s, D_2O exch., NH), 8.05 (1H, s, 5-H_{furyl}), 7.90 (1H, s), 7.73 (1H, d, J=8.1 Hz), 7.54 (1H, d, J=8.3 Hz), 7.33 (1H, t, J=7.5 Hz,), 7.23—7.11 (3H, m), 6.70 (3H two D_2O exch., m, NH_2). MS *m*/*z*: 277 (20), 276 (100), 219 (7), 168 (14), 154 (8), 140 (8), 128 (6), 127 (6), 66 (6). *Anal.* Calcd for $C_{18}H_{12}N_4O$: C, 69.55, H, 4.36; N, 20.26. Found: C, 69.52; H, 4.66; N, 20.33.

5-(2-Amino-5-chlorophenyl)-3-(2-pyridyl)-1*H*-1,2,4-triazole (3e) White solid, mp 233—235 °C, from toluene (lit. 207—209 °C).[15] IR (KBr) cm^{-1}: 3265, 1684, 1651, 1604, 1481, 808. ^{1}H-NMR (DMSO-d_6) δ: 10.34 (1H, D_2O exch., s, NH), 8.95 (1H, D_2O exch., s, NH), 8.65 (1H, d, J=5.0 Hz, 6-H_{pyr}), 8.00 (1H, d, J=7.9 Hz, H_{pyr}), 7.95—7.93 (1H, m), 7.65 (d, 1H, J=2.5 Hz), 7.56—7.52 (1H, m), 7.44 (1H, dd, J=8.7, 2.6 Hz, 4-H_{phenyl}), 7.17 (1H, d, J=8.7 Hz). ^{13}C-NMR (DMSO-d_6) δ: 165.85, 149.57, 149.21, 148.55, 144.56, 138.11, 134.25, 131.26, 126.10, 125.86, 123.15, 121.83, 121.73. MS *m*/*z*: 272 (100), 271 (26), 245 (30), 243 (90), 215 (13), 179 (17), 105(27), 79 (15), 78 (37), 75 (15).

5-(2-Amino-5-chlorophenyl)-3-(2-thienyl)-1*H*-1,2,4-triazole (3f) White solid, mp 252—255 °C, from cyclohexane. ^{1}H-NMR (DMSO-d_6) δ: 14.2 (1H, D_2O exch., br s, NH), 8.04 (1H, dd, J=3.6, 1.1 Hz, 5-$H_{thienyl}$), 7.96 (1H, dd, J=4.9, 1.2 Hz, $H_{thienyl}$), 7.80 (1H, d, J=2.4 Hz, $H_{thienyl}$), 7.33—7.28 (2H, m, H_{phenyl}+N<u>H</u>H), 6.94 (1H, d, J=8.94 Hz, H_{phenyl}), 6.89 (2H, br s, H_{arom}+NH<u>H</u>). *Anal.* Calcd for $C_{12}H_9ClN_4O$: C, 52.08; H, 3.26; N, 20.25. Found: C, 52.52; H, 3.03; N, 20.43.

5-(2-Amino-5-chlorophenyl)-3-phenyl-1*H*-1,2,4-triazole (3g) White solid, mp 254—256 °C, from toluene. (lit. 257—258 °C).[15] ^{1}H-NMR (DMSO-d_6) δ: 14.5 (1H, br s, NH), 8.07 (2H, d, J=6.9 Hz), 7.91 (1H, s), 7.52 (3H one D_2O exch., m, N<u>H</u>H), 7.16 (1H, d, J=7.1 Hz), 6.85 (3H one D_2O exch., m, NH<u>H</u>).

5-(2-Amino-5-chlorophenyl)-3-(3,4-dimethoxyphenyl)-1*H*-1,2,4-triazole (3h) White solid, mp 212—214 °C, from toluene. ^{1}H-NMR (DMSO-d_6) δ: 14.5 (1H, D_2O exch., br s, NH), 7.94 (1H, d, J=7.9 Hz), 7.67—7.66 (2H, m), 7.13—7.08 (2H, m), 6.82 (1H, d, J=8.7 Hz), 6.74 (2H D_2O exch., s, NH_2), 3.86 (3H, s, CH_3), 3.82 (3H, s, CH_3). *Anal.* Calcd for $C_{18}H_{15}ClN_4O_2$: C, 58.10; H, 4.57; N, 16.94. Found: C, 58.40; H, 4.35; N, 17.12.

Production of Triazoloquinazolines 4(a—h). General Procedure[12] 1.6 ml of 50% (w/w) aqueous cyanamide (1.7 mmol) was added to a slurry of the [1,2,4]triazole (1 mmol) in 2-propanol (5 ml), then 133 mg of 47% (w/w) aqueous sulphuric acid were added. The mixture was heated to reflux for 6 h, cooled to room temperature, and neutralized till the pH 7 by addition of 10% aqueous sodium hydroxide; after leaving overnight the mixture at 5 °C, the separated product was collected, washed with cold ethanol and dried.

5-Amino-9-chloro-2-(2-furyl)[1,2,4]triazolo[1,5-*c*]quinazoline (4a) Physical and spectroscopic data were consistent with data in the existing literature.[12,14]

5-Amino-8-chloro-2-(2-furyl)[1,2,4]triazolo[1,5-*c*]quinazoline (4b) White solid, mp 275—277 °C, ^{1}H-NMR (DMSO-d_6) δ: 8.07 (1H, d, J=0.7 Hz, 7-H), 8.55 (1H, d, J=8.6 Hz, 10-H), 7.54 (1H, d, J=3.5 Hz, 5-H_{furyl}), 6.98 (3H two D_2O exch., br s, NH_2+H_{furyl}), 6.82—6.81 (1H, m, H_{furyl}), 6.72 (1H, dd, J=8.5, 1.3 Hz, 9-H). ^{13}C-NMR (DMSO-d_6) δ: 163.12, 155.92, 149.05, 147.41, 138.38, 137.01, 129.82, 115.93, 115.25, 115.15, 113.06, 102.51. MS *m*/*z*: 263 (21), 261 (63), 204 (13), 156 (33), 154 (100), 126 (16), 95 (18), 84 (17), 66 (19). *Anal.* Calcd for $C_{13}H_8ClN_5O$: C, 54.65; H, 2.82; N, 24.51. Found: C, 54.52; H, 3.03; N, 24.33.

5-Amino-2-(2-furyl)[1,2,4]triazolo[1,5-*c*]quinazoline (4c) White solid, mp 280—283 °C, from EtOH (lit. 282—285 °C).[12,14] ^{1}H-NMR (DMSO-d_6) δ: 8.22 (1H, dd, J=7.9, 1.0 Hz, 7-H), 7.91 (1H, d, J=1.1 Hz, 5-H_{furyl}), 7.86 (2H, br s, D_2O exch., NH_2), 7.72—7.67 (1H, m, 10-H), 7.56 (1H, d, J=8.2 Hz, 8-H), 7.42—7.36 (1H, m, 9-H), 7.27 (1H, d, J=3.0 Hz, H_{furyl}), 6.73 (1H, dd, J=3.3, 1.8 Hz, H_{furyl}). ^{13}C-NMR (DMSO-d_6) δ: 155.86, 152.09, 145.80, 145.52, 145.45, 145.02, 132.61, 125.26, 123.74, 123.49, 113.47, 112.54. MS *m*/*z*: 253 (17), 252 (67), 251 (100), 145 (10), 108 (11), 84 (11), 66 (11).

5-Amino-2-(2-furyl)benzo[*g*][1,2,4]triazolo[1,5-*c*]quinazoline (4d) White solid, mp 309—311 °C, from AcOEt/MeOH. IR (KBr) cm^{-1}: 3364, 1694, 1582, 1583, 1109, 759. ^{1}H-NMR (DMSO-d_6) δ: 8.91 (1H, s, 7-H), 8.15 (1H, d, J=8.3 Hz, H_{arom}), 8.05 (1H, s, 12-H), 8.00 (1H, d, J=8.4 Hz, H_{arom}), 7.94 (1H, d, J=0.7 Hz, 5-H_{furyl}), 7.60—7.55 (1H, m, H_{arom}), 7.50 (1H, d, J=7.6 Hz, H_{arom}), 7.29 (1H, d, J=3.4 Hz, H_{furyl}), 6.75 (1H, dd, J=3.3, 1.7 Hz, H_{furyl}). ^{13}C-NMR (DMSO-d_6) δ: 163.42, 155.77, 149.45, 147.66, 139.18, 137.76, 130.07, 116.17, 115.50, 115.40, 113.30, 103.39. MS *m*/*z*: 301 (100), 272 (5), 194 (20), 167 (6), 140 (12), 108 (8), 63 (3). *Anal.* Calcd for $C_{17}H_{11}N_5O$: C, 67.77; H, 3.68; N, 23.24. Found: C, 67.92; H, 3.86; N, 23.54.

5-Amino-9-chloro-2-(2-pyridyl)[1,2,4]triazolo[1,5-*c*]quinazoline (4e) White solid, mp 207—209 °C, from AcOEt. IR (KBr) cm^{-1}: 3362, 1654, 1616, 1558, 1508, 1108, 61. ^{1}H-NMR (DMSO-d_6) δ: 8.69 (1H, d, J=4.2 Hz, 6-H_{pyr}), 8.15 (1H, d, J=2.4 Hz), 8.02 (1H, dd, J=7.7, 1.7 Hz), 7.99—7.85 (m, 2H), 7.77 (1H, d, J=8.7 Hz), 7.59—7.55 (m, 1H), 6.12 (2H D_2O exch., s, NH_2). ^{13}C-NMR (DMSO-d_6) δ: 162.73, 159.52, 153.34, 153.02, 148.91, 145.50, 137.58, 135.03, 132.08, 130.45, 125.50, 125.38, 124.98, 122.02. *Anal.* Calcd for $C_{14}H_9ClN_6O$: C, 56.67; H, 3.06; N, 28.32. Found: C, 56.60; H, 3.12; N, 28.11.

5-Amino-9-chloro-2-(2-thienyl)[1,2,4]triazolo[1,5-*c*]quinazoline (4f) White solid, mp 290—292 °C, from toluene. ^{1}H-NMR (DMSO-d_6) δ: 8.16 (1H, d, J=2.5 Hz, H_{arom}), 7.94 (2H D_2O exch., s, NH_2), 7.90 (1H, d, J=3.6 Hz, 5-$H_{thienyl}$), 7.80 (1H, d, J=5.0 Hz, 3 $H_{thienyl}$), 7.68 (1H, dd, J=8.9, 2.6 Hz, H_{arom}), 7.55 (1H, d, J=8.9 Hz, H_{arom}), 7.26 (1H, dd, J=4.8, 3.8 Hz, 4-$H_{thienyl}$). ^{13}C-NMR (DMSO-d_6) δ: 158.84, 150.96, 144.82, 143.81, 132.44, 132.25, 129.46, 128.48, 128.40, 126.98, 126.87, 122.20, 114.06. MS *m*/*z*: 301 (100), 300 (85), 178 (14), 151 (8), 137 (10), 124 (41), 102 (12), 97 (12), 84 (46), 78 (11), 69 (12). *Anal.* Calcd for $C_{13}H_8ClN_5S$: C, 51.75; H, 2.67; N, 23.21. Found: C, 51.54; H, 2.69; N, 23.38.

5-Amino-9-chloro-2-phenyl-[1,2,4]triazolo[1,5-*c*]quinazoline (4g) White solid, mp 296—298 °C, from toluene. (lit. 295—297 °C).[14] ^{1}H-NMR (DMSO-d_6) δ: 8.29—8.26 (2H, m), 8.20 (1H, d, J=2.4 Hz), 8.00 (2H, D_2O exch., s, NH_2), 7.70 (1H, dd, J=8.9, 2.6 Hz), 7.59—7.56 (4H, m). ^{13}C-NMR (DMSO-d_6) δ: 162.61, 151.01, 144.96, 143.72, 132.14, 130.60, 129.87, 128.99, 127.03, 126.87, 122.18, 114.33. MS *m*/*z*: 296 (80), 295 (100), 178 (8), 148 (8), 147 (10), 137 (10), 130 (13), 129 (13), 118 (17), 103 (15), 91

(13), 77 (17), 63 (12).

5-Amino-9-chloro-2-(3,4-dimethoxyphenyl)[1,2,4]triazolo[1,5-*c*]quinazoline (4h) White solid, mp 237—239 °C, from toluene. ^{1}H-NMR (DMSO-d_6) δ: 8.22 (1H, d, J=2.4 Hz), 7.99 (2H D_2O exch., s, NH_2), 7.89 (1H, dd, J=8.3, 1.8 Hz), 7.83 (1H, s), 7.82—7.69 (1H, m), 7.58 (1H, d, J=8.9 Hz), 7.18 (1H, d, J=8.7 Hz), 3.90 (3H, s, CH_3), 3.86 (3H, s, CH_3). MS *m*/*z*: 356 (35), 355 (100), 340 (13), 312 (17), 269 (11), 178 (23), 135 (13), 84 (32); 66 (32); 63 (15). *Anal.* Calcd for $C_{17}H_{14}ClN_5O_2$: C, 57.07; H, 4.51; N, 19.57. Found: C, 57.29; H, 4.62; N, 19.75.

5-Acetamido-2-(2-furyl)[1,2,4]triazolo[1,5-*c*]quinazoline (4i) A mixture of **4c** (0.16 g, 0.63 mmol) with 5 ml of Ac_2O and 5 ml of anhydrous pyridine was magnetically stirred under argon overnight. Solvent and excess reagent were removed at vacuum and the solid residue (0.12 g, 96%) was recrystallized from toluene; mp 206—208 °C. IR (KBr) cm^{-1}: 1739, 1613, 1528, 1508, 1476, 1425, 1368, 1314, 1227, 980, 767. ^{1}H-NMR (DMSO-d_6) δ: 8.57 (1H, d, J=7.7 Hz, 7-H), 8.14 (1H, d, J=8.1 Hz, 10-H), 8.06—7.92 (3H, m, 9-H+8-H+5-H_{furyl}), 7.37 (1H, d, J=3.3 Hz, 3-H_{furyl}), 6.76 (1H, dd, J=3.1, 1.6 Hz, 4-H_{furyl}), 2.42 (3H, s, CH_3). *Anal.* Calcd for $C_{15}H_{11}ClN_5O_2$: C, 61.43; H, 3.78; N, 23.88. Found: C, 61.60; H, 3.72; N, 23.51.

9-Chloro-2-(2-furyl)[1,2,4]triazolo[1,5-c]quinazoline (4j) Triethyl orthoformate (14 ml) and 12 N HCl (0.7 ml) were added to **3a** (0.56 g, 2.15 mmol). The mixture was stirred at room temperature for 2 d; the solvent was removed at vacuum and the obtained solid was recrystallized from toluene (0.37 g, 68%). White solid, mp 249—251 °C. IR (KBr) cm^{-1}: 3078, 1601, 1559, 1517, 1372, 1354, 1017, 910, 743. ^{1}H-NMR (DMSO-d_6) δ: 9.21 (1H, s, 5-H), 8.60 (1H, d, J=2.3 Hz, 10-H), 8.02 (1H, d, J=8.8 Hz, 7-H), 7.78 (1H, dd, J=8.8, 2.3 Hz, 8-H), 7.67 (1H, s, 5-H_{furyl}), 7.30 (1H, d, J=3.3 Hz, 3-H_{furyl}), 6.62 (1H, dd, J=3.4, 1.7 Hz, 4-H_{furyl}). ^{13}C-NMR (Cl_3CD) δ: 158.28, 150.80, 145.69, 145.35, 141.75, 138.00, 135.63, 133.29, 130.89, 123.66, 119.28, 113.29, 112.49. MS *m*/*z*: 270 (100), 269 (4), 241 (3), 179 (3), 165 (3), 163 (10), 136 (6), 122(4), 114 (4), 108 (22), 100 (5), 87 (6), 51 (3). *Anal.* Calcd for $C_{13}H_7ClN_4O$: C, 57.62; H, 2.61; N, 20.70. Found: C, 57.60; H, 2.88; N, 20.51.

Acknowledgements The authors thank the *Almirall-Prodesfarma, S. A.* for financially supporting this work.

References and Notes

1) Jacobson K. A., van Galen P. J. M., Willians M., *J. Med. Chem.*, **35**, 407—422 (1992).
2) Fredholm B. B., Ijzerman A. P., Jacobson K. A., Klotz K. N., Linden J., *Pharmacol. Rev.*, **53**, 527—552 (2001).
3) Klinger M., Freissmuth M., Nanoff C., *Cell Signal*, **14**, 99—108 (2002).
4) Olah M. E., Stiles G. L., *Ann. Rev. Pharmacol. Toxicol.*, **35**, 581—606 (1995).
5) Jacobson K. A., *Drugs Future*, **20**, 689—699 (1995).
6) Marquardt D. L., Walter L. L., Heinemann S., *J. Immunol.*, **152**, 4508—4515 (1994).
7) Auchampach J. A., Jin X., Wan T. C., Caughey G. H., Linden J., *Mol. Pharmacol.*, **52**, 846—860 (1997).
8) Feoktistov I., Biaggioni I., *Drug Dev. Res.*, **39**, 333—336 (1997).
9) Holgate S. T., *Br. J. Pharmacol.*, **145**, 1009—1015 (2005).
10) Cushley M. J., Tattersfield A. E., Holgate S. T., *Am. Rev. Resp. Dis.*, **129**, 380—384 (1984).
11) Feoktistov I., Wells J. N., Biaggioni I., *Drug Dev. Res.*, **45**, 198—206 (1998).
12) Francis J. E., Cash W. D., Psychoyos S., Ghai G., Wenk P., Friedmann R. C., Atkins C., Warren V., Furness P., Hyun J. L., Stone G. A., Desai M., Williams M., *J. Med. Chem.*, **31**, 1014—1020 (1988).
13) Dacroix B., Dubus P., Morel J., Pastour P., *Bull. Soc. Chim. France*, 621—627 (1976).
14) Gatta F., Giudice M. R., Borioni A., *J. Heterocycl. Chem.*, **30**, 11—16 (1993).
15) Francis J. E., Cash W. D., Barbaz B. S., Bernard P. S., Lovell R. A., Mazzenga G. C., Friedmann R. C., Hyun J. L., Braunwalder A. F., Loo P. S., Bennett D. A., *J. Med. Chem.*, **34**, 281—290 (1991).
16) Methanesulfonic salt of **4f**, mp 247—252 °C (reference 12).
17) Decroix B., Pastour P., *J. Chem. Res.* (*S*), 132—138 (1978).
18) Nakajima T., Izawa T., Kashwabara T., Nakajima S., Munezuka Y., *Chem. Pharm. Bull.*, **42**, 2475—248 (1994).
19) Carotti A., Cadavid M. I., Centeno N. B., Esteve C., Loza M. I., Martínez A., Nieto R., Raviña E., Sanz F., Segarra V., Sotelo E., Stefanachi A., Vidal B., *J. Med. Chem.*, **49**, 282—299 (2006).