HETEROCYCLES, Vol. 71, No. 10, 2007, pp. 2249 - 2262. © The Japan Institute of Heterocyclic Chemistry
Received, 15th May, 2007, Accepted, 3rd July, 2007, Published online, 6th July, 2007. COM-07-11110

SYNTHESIS AND FRIEDLÄNDER REACTIONS OF 5-AMINO-4-CYANO-1,3-OXAZOLES

M. Carmo Carreiras,[a,*] Ana Eleutério,[a] Catarina Dias,[a] M. Alexandra Brito,[b] Dora Brites,[b] J. Marco-Contelles,[c] and Elena Gómez-Sánchez [c]

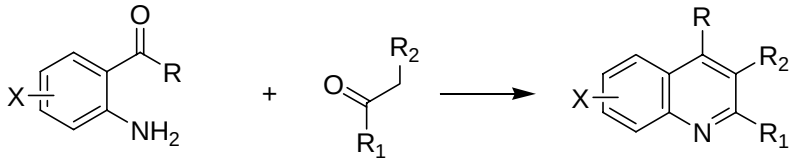
[a]Centro de Estudos de Ciências Farmacêuticas (CECF) and [b]Centro de Patogénese Molecular (UBMBE), Faculdade de Farmácia, Universidade de Lisboa, Av. das Forças Armadas, 1600-083 Lisboa, Portugal; [c]Instituto de Química Orgánica General (CSIC), C/ Juan de la Cierva 3, 28006-Madrid, Spain
mcdamaso@ff.ul.pt

Abstract – The synthesis of 2-substituted 5-amino-4-cyano-1,3-oxazoles (**1-4**, **6-11**) and the Friedländer-type reaction of compounds **1, 3, 4** is described. Compounds **13-17** are tacrine (**18**) analogues provided by the Friedländer reaction. The anti-cholinesterase activity of compounds **13**, **14**, **16** and **17** has been investigated.

INTRODUCTION

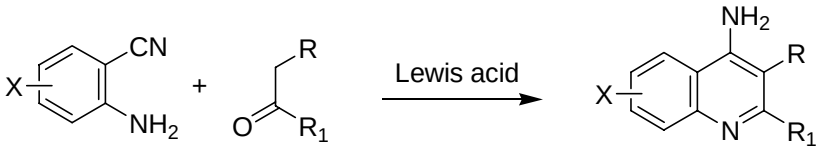
Quinolines are heterocyclic ring systems[1] present in a number of natural[2] or synthetic products endowed with interesting pharmacological or physical properties.[3] Thus, a number of methods for their synthesis have been reported and the Friedländer reaction is still one of the straightforward means for the synthesis of quinolines.[4] In its most conventional procedure, the Friedländer condensation is the base- or acid-promoted reaction followed by cyclodehydration and annulation of an aromatic *o*-amino-substituted carbonyl compound (aldehyde, ketone or derivative thereof) with an appropriately substituted carbonyl derivative containing a reactive α-methylene group (Scheme 1).[5] Friedländer annulations are generally carried out either by refluxing an aqueous or alcoholic solution of reactants in the presence of a base or by

heating a mixture of the reactants at high temperature (150-220ºC) in the absence of catalyst.[6]



Scheme 1. General procedure for the Friedländer reaction.

Regarding the *o*-aminocarbonyl component of the Friedländer reaction, the only limitation with respect to the functional groups attached to the aromatic ring concerns their stability to the basic or acidic conditions used to promote the process. In agreement with this, X can be hydrogen, alkyl, aryl, *O*-alkyl, etc. (Scheme 1). The majority of *o*-amino-substituted carbonyl compounds used in the Friedländer reaction are *o*-aminobenzaldehydes, *o*-aminoacetophenones and *o*-aminobenzophenones. Heterocyclic ring systems containing the *o*-aminocarboxaldehyde moiety have also been common partners in this type of reaction.[7] In an extension of this condensation reaction, *o*-amino-substituted benzonitriles have also been used for the synthesis of 4-aminoquinolines (Scheme 2).[8]

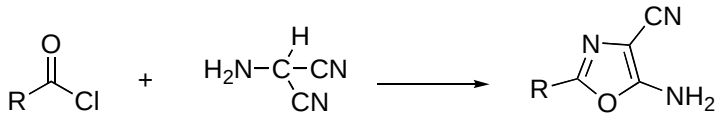


Scheme 2. Synthesis of 4-aminoquinolines.

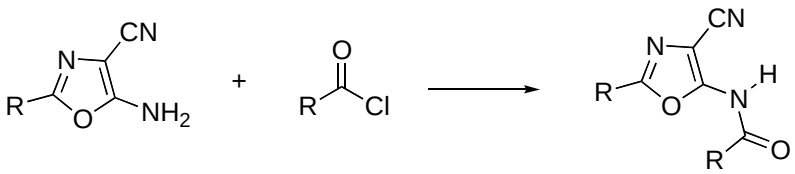
With these precedents in mind, it was surprising to ascertain that *o*-amino substituted 4- or 5-cyano 1,3-oxazoles[9] have been barely investigated as precursors in the Friedländer reaction for the synthesis of fused oxazolo[5,4-*b*]quinolines of potential biological interest.[10]

In this note we report our recent studies on the synthesis of some 2-(aryl, alkyl) substituted 1,3-oxazoles (**1**-**4**, **6**-**11**) (Schemes 3 and 4, and Table 1), as wel1 as the Friedländer reaction of compounds **1**, **3** and **4**, and the acetylcholinesterase (AChE) inhibitory activity of the resulting oxazolo[5,4-*b*]quinolines (**13**, **14**) and the cyclohepta[*b*]oxazolo[4,5-*e*]pyridines (**16**, **17**) (Scheme 5). The AChE inhibitory activity of

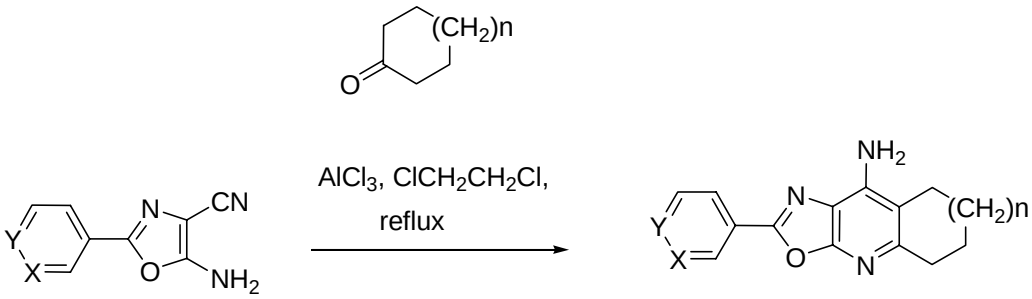
precursor **3** (Table 1) was also assessed.



Scheme 3. Synthesis of the 2-alkyl (and 2-aryl)-5-amino-4-cyano-1,3-oxazoles (**1**-**4**, **6**, **7**, **9**, **12**).



Scheme 4. Synthesis of the *N*-acylated 5-amino-1,3-oxazoles (by-products **8**, **10**, **11**).

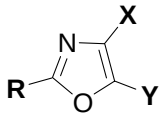


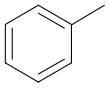
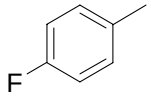
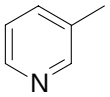
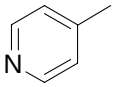
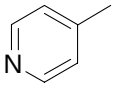
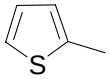
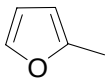
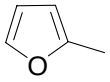
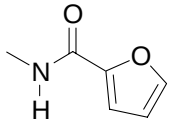
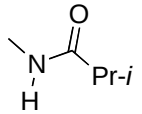
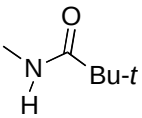
1 X= Y= CH
3 X= N; Y= CH
4 X= CH; Y= N

13 X= Y= CH (n= 1) (84%)
14 X= N; Y= CH (n= 1) (18%)
15 X= CH; Y= N (n= 1) (11%)
16 X= Y= CH (n= 2) (90%)
17 X= N; Y= CH (n= 2) (20%)

Scheme 5. Synthesis of tacrine analogues **13**-**17** by Friedländer reaction.

Table 1. Structures of the 2-alkyl (and 2-aryl)-1,3-oxazoles (**1-12**).



Compound	R	X	Y
1		CN	NH_2
2		CN	NH_2
3		CN	NH_2
4		CN	NH_2
5		CO_2H	NH_2
6		CN	NH_2
7		CN	NH_2
8		CN	
9	*i*-Pr	CN	NH_2
10	*i*-Pr	CN	
11	*t*-Bu	CN	
12	*t*-Bu	CN	NH_2

RESULTS AND DISCUSSION

The synthesis of the 1,3-oxazole precursors was attempted according to the general method reported by Freeman and Kim.[11] Thus, a solution of commercially available aminomalonitrile tosylate (AMNT) in 1-methyl-2-pyrrolidinone was reacted with different aryl or alkyl acid chlorides at room temperature (Scheme 3). The standard procedure worked suitably to afford the expected products (**1**-**3**) in moderate yields, albeit in high reaction times (around 7 days). In other circumstances, we were forced to use an excess of the acid chloride since no reaction was observed when employing a 1.5 equiv of the reagent, but under these conditions, by-products were detected which considerably lowered the yield of the expected product (**7**, **9**) or did not afford at all the required compound (**12**). Regarding the undesirable by-products, we have isolated the 5-amino-1,3-oxazole-4-carboxylic acid (**5**) (10%) (Table 1), as well as the *N*-acylated 5-amino-1,3-oxazoles **8**, **10**, and **11**, from low (7%) to high yield (82%) (Scheme 4 and Table 1).

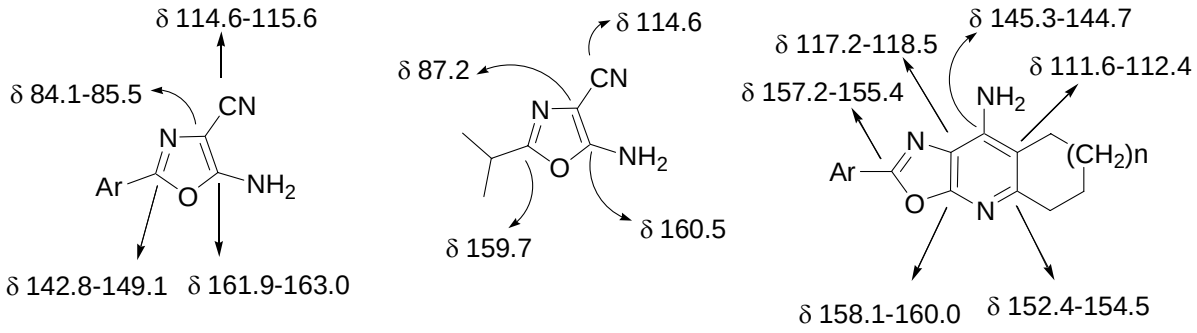


Figure 1. ^{13}C NMR (DMSO-d_6) data of the 2-alkyl (and 2-aryl)oxazolo derivatives.

The general procedure for the synthesis of compounds **1**, **7** and **9** has been previously described,[11] but their spectroscopic data has not been reported. All the known and new compounds communicated in this work showed good analytical and spectroscopic data (see **Experimental**). Moreover, the ^{13}C NMR data (see Figure 1) have been assigned by HMBC and HSQC experiments, and are in good agreement with those described for 1,3-oxazoles.[12]

The Friedländer condensation was carried out with these precursors under nitrogen using either cyclohexanone or cycloheptanone. Under the usual conditions, the reaction with 1,3-oxazoles (**1**, **3** and **4**) rendered the expected products **13**-**17** (Scheme 5) from low to good yields. However, the best results were obtained for precursor **1**, while the 3'- and 4'-pyridyl substituted compounds **3** and **4** afforded low chemical yields. In total, the investigated reactions took a long time to be completed, under reflux and

with excess of catalyst. The structures of these compounds have been established by their analytical and spectroscopic data. The more significant ^{13}C NMR data (see Figure 1) have been assigned by HMBC and HSQC experiments, and are in good agreement with those described for 1,3-oxazoles.[8]

Finally, regarding the close structural and functional relationship of compounds **13-17** (Scheme 5) with tacrine (**18**) (Figure 2), a reversible AChE inhibitor[13] formerly used for the treatment of Alzheimer`s disease,[14] the tacrine analogues **13**, **14**, **16**, **17** and precursor **3** have been submitted towards the usual pharmacological protocols in order to ascertain their inhibitory activity.

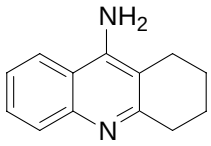
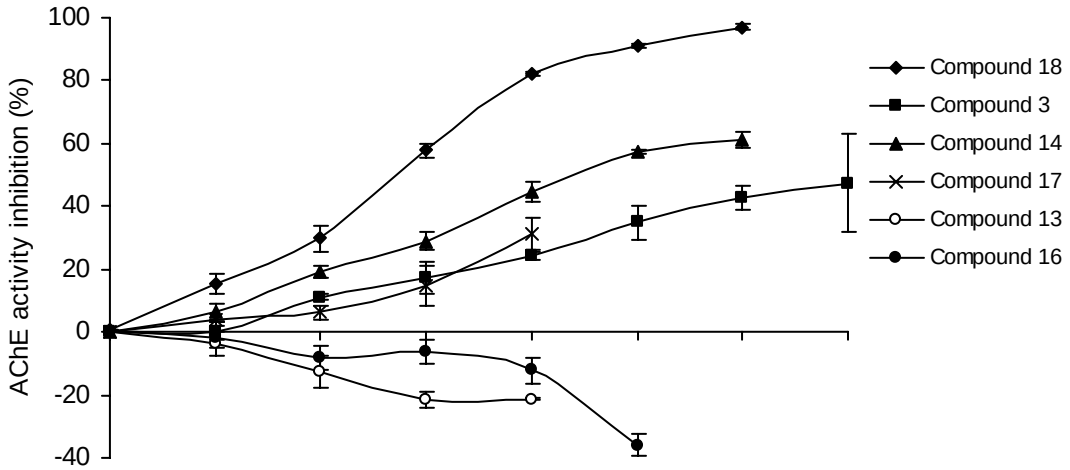


Figure 2. The structure of tacrine (18).

As expected, tacrine (**18**; Figure 2) presents a relevant AChE inhibitory activity (Figure 3).



Tacrine (**18**)	0	0.02	0.1	0.5	2.5	12.5	50	
Compound **3**	0	7.8	15.6	31.3	62.5	125	250	500
Compound **14**	0	3.9	15.6	62.5	125	250	500	
Compound **17**	0	10	30	50	70			
Compound **13**	0	10	20	30	40			
Compound **16**	0	2.5	5	10	20	40		

Figure 3. Inhibitory effects on AChE activity at different concentrations of the reference compound (tacrine; **18**) and of the tested compounds (**3**, **13**, **14**, **16** and **17**). Data are means ± SD of at least two independent experiments, performed in quadruplicate. Drugs' concentrations are presented in μM.

In fact, 0.5 μM tacrine inhibited AChE activity by more than 50%, while 50 μM nearly completely (>97%) repressed the enzyme activity. The AChE activity of the test compounds was assayed until their solubility limit in DMSO, which was 500 μM for compounds **3** and **14**, 70 μM for compound **17**, and 40 μM for both compounds **13** and **16**. At the maximum soluble concentrations, the highest AChE activity inhibition was achieved by compound **14**, followed by compound **3**, which were able to inhibit the enzyme by nearly 60% and 50%, respectively. Despite the lower solubility of derivative **17**, AChE activity was suppressed by around 30%. Contrasting with compounds **3**, **14** and **17**, the oxazolo-derivatives **13** and **16** were devoid of any inhibitory activity, a fact that conceivably results from the different structures of these compounds.

EXPERIMENTAL

General Methods. Reactions were monitored by TLC using precoated silica gel aluminium plates containing a fluorescent indicator (Merck, 5539). Detection was done by UV (254 nm) followed by charring with sulfuric-acetic acid spray, 1% aqueous potassium permanganate solution or 0.5% phosphomolybdic acid in 95% EtOH. Anhydrous Na_2SO_4 was used to dry organic solutions during work-ups and the removal of solvents was carried out under vacuum with a rotary evaporator. Flash column chromatography was performed using silica gel 60 (230-400 mesh, Merck). Melting points were determined on a Köfler block and are uncorrected. IR spectra were obtained on a Perkin-Elmer spectrophotometer. ^{1}H and ^{13}C NMR spectra were recorded with a Varian VXR-300S or Varian Inova-400 spectrometers, using tetramethylsilane as internal standard. All the assignments for protons and carbons were in agreement with 2D COSY, HSQC, HMBC, and 1D NOESY spectra. Values with (*) can be interchanged. Elemental analyses were performed on a Carlo Erba EA 1108 apparatus.

General Method for the synthesis of 2-substituted 5-amino-4-cyano-1,3-oxazoles. According to Freeman and Kim,[11] to a stirred solution of aminomalonitrile tosylate (AMNT) and 1-methyl-2-pyrrolidinone (0.09 M), the corresponding acid chloride (1.4- 6.0 equiv, depending on the case) was added in one portion. The reaction mixture was stirred at rt until the reaction was complete. Then, the mixture was diluted with a mixture of EtOH and Et_2O (1:1), and washed with water, 10% aqueous $NaHCO_3$, and water. The organic layer was dried, the solvent was evaporated in vacuo, and the product purified by chromatography.

5-Amino-4-cyano-2-phenyl-1,3-oxazole (**1**). Following the **General Method for the synthesis of 2-substituted 5-amino-4-cyano-1,3-oxazoles**, AMNT (4.68 g, 18.1 mmol), 1-methyl-2-pyrrolidinone (48

mL) and benzoyl chloride (2.95 mL, 25.4 mmol, 1.4 equiv) were reacted at rt for 8 days. Work-up and chromatography (CH_2Cl_2-EtOAc, 6:4) afforded compound (**1**) (1.94 g, 57%) as a yellow solid: mp 230-235 ºC; IR (KBr) ν_{max} 3380, 3220, 2260, 1650, 1610, 1070 cm^{-1}; ^1H NMR (DMSO-d_6, 300 MHz) δ 7.87 (m, H2', H6', NH_2, 4 H), 7.57 (m, H3', H4', H5', 3 H); ^{13}C NMR (DMSO-d_6, 75.4 MHz) δ 161.9 (C5), 149.1 (C2), 129.5 (C4'), 128.6 (2 C, C3', C5'), 125.7 (C1'), 124.6 (2 C, C2', C6'), 114.7 (CN), 84.2 (C4); MS (EI)-*m*/*z* (%): 185 $[M]^+$ (80), 157 $[M\text{-}CO]^+$ (52), 141 $[M\text{-}(NH_2CO)]^+$ (9), 130 (27), 115 (27), 105 (100), 104 (83), 103 $[PhCN]^+$ (13), 89 (34), 77 $[Ph]^+$ (57), 63 (26), 51 (31), 44 $[NH_2CO]^+$ (15). Anal. Calcd for $C_{10}H_7N_3O$: C, 64.86; H, 3.81; N, 22.69. Found: C, 64.82; H, 3.58; N, 22.55.

5-Amino-4-cyano-2-(4`-fluorophenyl)-1,3-oxazole (**2**). Following the **General Method for the synthesis of 2-substituted 5-amino-4-cyano-1,3-oxazoles**, AMNT (4.68 g, 18.1 mmol), 1-methyl-2-pyrrolidinone (40 mL) and 4-fluorobenzoyl chloride (3.40 mL, 28.78 mmol. 1.5 equiv) were reacted at rt for 6 days. Work-up and chromatography (CH_2Cl_2) rendered compound (**2**) (1.99 g, 53%) as yellow crystals: mp 260-262 ºC; IR (KBr) ν_{max} 3320, 3170, 2210, 1650, 1600, 1490, 1415, 1230, 1050 cm^{-1}; ^1H NMR (DMSO-d_6, 300 MHz) δ 7.94 (br s, NH_2, 2 H), 7.79 (m, H2', H6', 2 H), 7.29 (m, H3', H5', 2 H); ^{13}C NMR (DMSO-d_6, 75.4 MHz) δ 163.3 (C4'), 162.6 (C5), 149.1 (C2), 127.7 (2 C, C2', C6'), 122.8 (C1'), 116.5 (2 C, C3', C5'), 115.6 (CN), 84.5 (C4); MS (EI)-*m*/*z* (%): 203 $[M]^+$ (68), 176 (4), 175 $[M\text{-}CO]^+$ (35), 159 $[M\text{-}(NH_2CO)]^+$ (7), 149 (7), 148 (11), 133 (29), 132 (10), 124 (9), 123 (100), 122 (67), 121 $[FPhCN]^+$ (15), 107 (30), 106 (8), 95 $[FPh]^+$ (46), 94 (10), 85 (10), 84 (10), 83 (10), 82 $[M\text{-}FPhCN)^+$ (6), 81 (14), 80 $[(M\text{-}CO)\text{-}FPh]^+$ (3), 75 (25), 69 (19), 44 $[NH_2CO]^+$ (16). Anal. Calcd for $C_{10}H_6FN_3O$: C, 59.12; H, 2.98; N, 20.68. Found: C, 59.40; H, 3.28; N, 20.91.

5-Amino-4-cyano-2-(3`-pyridyl)-1,3-oxazole (**3**). Following the **General Method for the synthesis of 2-substituted 5-amino-4-cyano-1,3-oxazoles**, AMNT (4.68 g, 18.1 mmol), 1-methyl-2-pyrrolidinone (40 mL) and nicotinoyl chloride hydrochloride (4.93 g, 26.88 mmol, 1.4 equiv) were reacted at rt for 7 days. Work-up and chromatography (CH_2Cl_2: MeOH, 9.8:0.2) gave compound (**3**) (1.63 g, 47%) as an yellow solid: mp 267-269ºC; IR (KBr) ν_{max} 3360, 3310, 2230, 1675, 1620, 1415, 1025 cm^{-1}; ^1H NMR (DMSO-d_6, 300 MHz) δ 8.92 (d, $J_{2',4'}$= 2.2 Hz, H2', 1 H), 8.61 (dd, $J_{6',5'}$= 4.8 Hz, $J_{6',4'}$= 1.4 Hz, H6', 1 H), 8.10 (m, H4', NH_2, 3 H), 7.51 (dd, $J_{5',4'}$= 8.0 Hz, $J_{5',6'}$= 4.8 Hz, H5', 1 H); ^{13}C NMR (DMSO-d_6, 75.4 MHz) δ 162.8 (C5), 150.7 (C6'), 147.6 (C2), 146.1 (C2'), 132.7 (C4'), 124.5 (C5'), 122.6 (C3'), 115.4 (CN), 84.9 (C4); MS (EI) *m*/*z* (%): 186 $[M]^+$ (91), 158 $[M\text{-}CO]^+$ (28), 142 $[M\text{-}(NH_2CO)]^+$ (8), 131 (28), 116 (17), 106 (100), 104 $[pyCN]^+$ (11), 95 (9), 89 (18), 82 $[M\text{-}(pyCN)]^+$ (4), 80 $[(M\text{-}CO)\text{-}py]^+$ (2), 78 (39), 69 (16), 63 (30), 54 (3), 44 $[NH_2CO]^+$ (31). Anal. Calcd for $C_9H_6N_4O$: C, 58.06; H, 3.25; N, 30.09. Found: C, 58.25; H, 3.25; N, 30.04.

5-Amino-4-cyano-2-(4`-pyridyl)-1,3-oxazole (**4**) and **5-amino-2-(4'-pyridyl)-1,3-oxazole-4-carboxylic acid** (**5**). Following the **General Method for the synthesis of 2-substituted 5-amino-4-cyano-1,3-**

oxazoles, AMNT (4.68 g, 18.5 mmol), 1-methyl-2-pyrrolidinone (36 mL) and isonicotinoyl chloride hydrochloride (7.62 g, 40.7 mmol, 2.4 equiv) were reacted at rt for 7 days. Work-up and chromatography (CH_2Cl_2: MeOH, 9.6:0.4) provided compound (**4**) (0.92 g, 27%). In the aqueous layer, after evaporation we were able to isolate compound (**5**) (0.39 g, 10%). **5-Amino-4-cyano-2-(4`-pyridyl)-1,3-oxazole** (**4**): mp 282-286ºC; IR (KBr) ν_{max} 3435, 3335, 3041, 2220, 1666, 1607, 1572, 1418, 1344, 1185, 1053 cm^{-1}; ^1H NMR δ (DMSO-d_6, 300 MHz) δ 8.65 (m, H2', H6', 2 H), 8.22 (br s, NH_2, 2 H), 7.60 (m, H3', H5', 2 H); ^{13}C NMR (DMSO-d_6, 75.4 MHz) δ 163.0 (C5), 150.8 (2 C, C2', C6'), 147.5 (C2), 132.9 (C4'), 118.9 (2 C, C3', C5'), 115.1 (CN), 85.5 (C4); MS (EI) *m*/*z* (%): 186 $[M]^+$ (100), 158 $[M\text{-}CO]^+$ (25), 143 (6), 142 $[M\text{-}NH_2CO]^+$ (7), 131 (34), 116 (13), 106 (69), 105 (38), 104 $[pyCN]^+$ (10), 89 (11), 80 (1), 78 $[py]^+$ (38), 63 (23), 62 (12), 51 (32), 50 (14), 44 $[NH_2CO]^+$ (12). Anal. Calcd for $C_9H_6N_4O$: C, 58.06; H, 3.25; N, 30.09. Found: C, 57.87; H, 3.22; N, 29.50. **5-Amino-2-(4'-pyridyl)-1,3-oxazole-4-carboxylic acid** (**5**): mp 223-225ºC; IR (KBr) ν_{max} 3456, 3397, 3170, 1696, 1600, 1550, 1477, 1423, 1382, 1300, 1217, 1000 cm^{-1}; ^1H NMR (DMSO-d_6, 300 MHz) δ 8.62 (m, H2', H6', 2 H), 7.66 (m, H3', H5', 2 H), 7.24 (br s, NH_2, 2 H); ^{13}C NMR (DMSO-d_6, 75.4 MHz) δ 161.1 (CO_2H), 159.0 (C5), 150.6 (2 C, C2', C6'), 145.7 (C2), 133.7 (C4'), 118.7 (2 C, C3', C5'), 107.4 (C_4); (EI) *m*/*z* (%): 205 $[M]^+$ (16), 204 $[M\text{-}H]^+$ (100), 187 (38), 161 (2), 106 (60), 105 (23), 104 (4), 79 (21), 78 $[py]^+$ (32), 71 (6), 63 (10), 51 (18), 50 (6), 44 $[NH_2CO]^+$ (10). Anal. Calcd for $C_9H_7N_3O_3$: C, 52.68; H, 3.43; N, 20.48. Found: C, 52.40; H, 3.34; N, 20.27.

5-Amino-4-cyano-2-(2'-thienyl)-1,3-oxazole (**6**). Following the **General Method for the synthesis of 2-substituted 5-amino-4-cyano-1,3-oxazoles**, AMNT (4.68 g, 18.1 mmol), 1-methyl-2-pyrrolidinone (36 mL) and 2-thiophenecarbonyl chloride (3.95 mL, 35.8 mmol, 1.9 equiv) were reacted at rt for 6 days. Work-up and chromatography (CH_2Cl_2: MeOH, 9.5:0.5) gave compound (**6**) (0.21 g, 6%): mp 246-249 ºC; IR (KBr) ν_{max} 3360, 3280, 3219, 3162, 2214, 1657, 1610, 1591, 1424, 1053 cm^{-1}; ^1H NMR (DMSO-d_6, 300 MHz) δ 7.96 (br s, NH_2, 2 H), 7.66 (dd, *J*= 4.6, 1.1 Hz, H5', 1 H), 7.48 (dd, *J*= 4.6, 3.6 Hz, H3', 1 H), 7.15 (dd, *J*= 3.6, 1.1 Hz, H4', 1 H); ^{13}C NMR (DMSO-d_6, 75.4 MHz) δ 162.2 (C5), 146.3 (C2), 129.0 (C5'), 128.7 (C4'), 128.2 (C2'), 127.4 (C3'), 115.4 (CN), 84,1 (C4); MS (EI) *m*/*z* (%):191 $[M]^+$ (93), 163 $[M\text{-}CO]^+$ (19), 147 $[M\text{-}(NH_2CO)]^+$ (6), 121 (22), 120 (6), 112 (13), 111 (100), 110 (37), 109 $[SC_4H_3CN]^+$ (6), 95 (18), 94 (12), 83 (7), 82 (7), 80 (1), 70 (10), 69 (24), 45 (10), 44 $[NH_2CO]^+$ (13), 39 (15). Anal. Calcd for $C_8H_5N_3OS$: C, 50.25; H, 2.64; N, 21.98. Found: C, 49.98; H, 2.52; N, 21.95.

5-Amino-4-cyano-2-(2'-furyl)-1,3-oxazole (**7**) and **4-cyano-5-(2''-furoyl)amino-2-(2'-furyl)-1,3-oxazole** (**8**). Following the **General Method for the synthesis of 2-substituted 5-amino-4-cyano-1,3-oxazoles**, AMNT (1.75 g, 6.78 mmol), 1-methyl-2-pyrrolidinone (13 mL) and 2-furoyl chloride (2.10 mL, 20.2 mmol, 2.9 equiv) were reacted at rt for 3 days. Work-up and chromatography (CH_2Cl_2: MeOH, 9.5:0.5) afforded compound (**7**) (1.06 g, 29%) and (**8**) (0.42 g, 7%). **5-Amino-4-cyano-2-(2'-furyl)-1,3-**

oxazole (**7**): yellow solid; mp 220-223ºC; IR (KBr) ν_{max} 3363, 3202, 3159, 2223, 1659, 1639, 1464, 1167, 1085, 1013 cm^{-1}; ^1H NMR (DMSO-d_6, 300 MHz) δ 7.98 (br s, NH_2, 2 H), 7.86 (d, *J*= 1.6 Hz, H5', 1 H), 6.93 (d, *J*= 3.4 Hz, H3', 1 H), 6.66 (dd, *J*= 3.4, 1.6 Hz, H4', 1 H); ^{13}C NMR (DMSO-d_6, 75.4 MHz) δ 162.1 (C5), 145.4 (C5'), 142.8 (C2), 141.3 (C2'), 115.5 (CN), 112.5 (C4'), 111.0 (C3'), 84.1 (C4); MS (EI) *m*/*z* (%): 175 $[M]^+$ (98), 147 $[M-(NH_2CO)]^+$ (3), 131 (5), 119 (4), 105 (5), 95 (100), 93 (3), 82 (1), 80 (1), 77 (25), 76 (14), 75 (5), 68 (4), 67 (59), 66 (6), 53 (10), 51 (22), 50 (17), 44 $[NH_2CO]^+$ (19), 39 (35), 38 (14). Anal. Calcd for $C_8H_5N_3O_2$: C, 54.86; H, 2.88; N, 23.99. Found: C, 54.90; H, 3.02; N, 23.79. **4-Cyano-5-(2''-furoyl)amino-2-(2'-furyl)-1,3-oxazole** (**8**): white crystals; mp 269-271 ºC; IR (KBr) ν_{max} 3436, 3253, 3123, 2239, 1682, 1636, 1616, 1583, 1547, 1521, 1474, 1305, 1195, 1178, 1083, 1062, 1016 cm^{-1}; ^1H NMR (DMSO-d_6, 300 MHz) δ 8.01 (d, *J*= 1.7 Hz, H5'', 1 H), 7.92 (d, *J*= 1.7 Hz, H5', 1 H), 7.54 (d, *J*= 3.6 Hz, H3'', 1 H), 7.25 (d, *J*= 3.5 Hz, H3', 1 H), 6.79 (dd, *J*= 3.6, 1.7 Hz, H4'', 1 H), 6.74 (dd, *J*= 3.5, 1.7 Hz, H4', 1 H); ^{13}C NMR (DMSO-d_6, 75.4 MHz) δ 155.1 (NHCO), 150.4 (C5), 147.9 (C5''), 146.7 (C5'), 145.5 (C2''), 140.4 (C2'), 118.0 (C3''), 113.9 (C3'), 113.4 (CN), 113.0 (C4'')*, 112.9 (C4')*, 99.8 (C_4); MS (EI) *m*/*z* (%): 269 (8), 174 (1), 131 (1), 97 (1), 96 (8), 95 (100), 94 (1), 77 (1), 67 $[C_4H_3O]^+$ (3), 66 (1), 53 (1), 51 (3), 50 (1), 39 (13). Anal. Calcd for $C_{14}H_{11}N_3O_4$: C, 58.95; H, 3.89; N, 14.73. Found: C, 58.63; H, 3.57; N, 14.45.

5-Amino-4-cyano-2-isopropyl-1,3-oxazole (**9**) and **4-cyano-5-(isobutyroyl)amino-2-isopropyl-1,3-oxazole** (**10**). Following the **General Method for the synthesis of 2-substituted 5-amino-4-cyano-1,3-oxazoles**, AMNT (2,34 g; 9,1 mmol), 1-methyl-2-pyrrolidinone (20 mL) and *i*-butyroyl chloride (3 mL, 28.1 mmol, 3.0 equiv) were reacted at rt for 3 d. Work-up and chromatography (hexane: EtOAc, 6:4) yielded compound (**9**) (0.91 g, 20%) and (**10**) (0.88 g, 12%). **5-Amino-4-cyano-2-isopropyl-1,3-oxazole** (**9**): white crystals: mp 153-155 ºC; IR (KBr) ν_{max} 3352, 3166, 2964, 2217, 1655, 1617, 1588, 1472, 1434, 1347, 1201, 1141, 1078 cm^{-1}; ^1H NMR ($CDCl_3$, 300 MHz) δ 4.87 (br s, NH_2, 2 H), 2,91 (d, *J*= 6.9 Hz, H7, 1 H), 1.27 (d, *J*= 6.9 Hz, H8, 6 H); ^{13}C NMR ($CDCl_3$, 75.4 MHz) δ 160.5 (C5), 159.7 (C2), 114.6 (CN), 87.2 (C4), 28.5 (C7), 20.3 (2 C, C8, C8'); MS (EI) *m*/*z* (%): 151 $[M]^+$ (53), 137 (8), 136 (100), 123 (1), 109 (8), 108 (5), 107 $[M-NH_2CO]^+$ (23), 106 (7), 96 (9), 82 (2), 81 (16), 80 (6), 71 (6), 70 (4), 69 (4), 66 (20), 56 (30), 55 (17), 54 (8), 53 (12), 44 $[NH_2CO]^+$ (16), 43 (36), 41 (27), 39 (25). Anal. Calcd for $C_7H_9N_3O$: C, 55.62; H, 6.00; N, 27.80. Found: C, 55.47; H, 6.20; N, 27.67. **4-Cyano-5-(isobutyroyl)amino-2-isopropyl-1,3-oxazole** (**10**): white crystals; mp 85-86ºC; IR (KBr) ν_{max} 3436, 3246, 3195, 3043, 2977, 2934, 2876, 2234, 1686, 1626, 1551, 1466, 1238, 1179, 1079 cm^{-1}; ^1H NMR ($CDCl_3$, 300 MHz) δ 8.52 (br s, NH, 1 H), 3.01 (d, *J*= 6.9 Hz, H7, 1 H), 2.66 (d, *J*= 6.9 Hz, H10, 1 H), 1.37 (d, *J*= 6.9 Hz, H8, 6 H), 1.28 (d, *J*= 6.9 Hz, H11, 6 H); ^{13}C NMR ($CDCl_3$, 75.4 MHz) δ 174.2 (NHCO), 163.6 (C2), 149.7 (C5), 112.7 (CN), 99.5 (C4), 35.4 (C10), 28.2 (C7), 19.9 (C8)*, 19.1 (2 C, C11, C11')*; MS (EI) *m*/*z* (%): 221 $[M]^+$ (18), 193 $[M-CO]^+$ (2), 178 (2), 153 (4), 152 $[M-C_3H_7CN]^+$ (27),

151 (74), 150 [(M-CO)-C_3H_7]$^+$ (2), 137 (5), 136 (54), 135 (4), 108 (4), 107 (14), 106 (3), 96 (8), 81 (5), 71 (97), 70 (10), 69 [C_3H_7CN]$^+$ (2), 66 (2), 56 (5), 54 (2), 53 (3), 44 (5), 43 [C_3H_7]$^+$ (100), 42 (5), 41 (23), 39 (9). Anal. Calcd for $C_{11}H_{15}N_3O_2$: C, 59.71; H, 6.83; N, 18.99. Found: C, 58.83; H, 8.43; N, 18.94.

2-(t-Butyl)-4-cyano-5-(2',2'-dimethylpropanoyl)amino-1,3-oxazole (**11**). Following the **General Method for the synthesis of 2-substituted 5-amino-4-cyano-1,3-oxazoles**, AMNT (3.51 g, 13.58 mmol), 1-methyl-2-pyrrolidinone (30 mL) and pivaloyl chloride (10 mL, 80.37 mmol, 6.0 equiv) were reacted at rt for 3 days. Work-up and chromatography (hexane: EtOAc, 6:4) supplied compound (**11**) (2,83 mg, 82%): white crystals; mp 170-172 ºC; IR (KBr) ν_{max} 3270, 2980, 2930, 2238, 1685, 1630, 1580, 1550, 1525, 1305, 1180, 1110 cm^{-1}; ^{1}H NMR ($CDCl_3$, 300 MHz) δ 7.94 (br s, NH, 1 H), 1.35 (s, H8, 9 H), 1.33 (s, H11, 9 H); ^{13}C NMR ($CDCl_3$, 75.4 MHz) δ 175.2 (NHCO), 165.8 (C2), 149.1 (C5), 112.6 (CN), 100.3 (C4), 39.6 (C10), 33.8 (C7), 28.1 (3 C, C11, C11', C11")*, 27,2 (3 C, C8, C8', C8")*; MS (EI) *m*/z (%): 249 [M]$^+$ (11), 166 [M-$(CH_3)_3CCN$]$^+$ (6), 165 (50), 151 (5), 150 (46), 121 (5), 85 (22), 83 (1), 58 (7), 57 [$(CH_3)_3C$]$^+$ (100), 41 (20), 39 (5). Anal. Calcd for $C_{13}H_{19}N_3O_2$: C, 62.63; H, 7.68; N, 16.85. Found: C, 62.77; H, 8.00; N, 16.57.

General Method for the Friedländer reaction. To a suspension of $AlCl_3$ (2 equiv) in dry 1,2-dichloroethane (10 mL/mmol) at rt, under nitrogen, the corresponding cycloalkanone (2 equiv), and the oxazole were added. The mixture was refluxed until the reaction was complete. Then water and 10% aqueous solution of NaOH were added until pH 9. Then, a mixture of water and EtOAc was added, and the resulting precipitate was collected. The organic layer was dried, filtered, and evaporated. The first solid, and the residue were submitted to chromatography to give the compound.

4-Amino-5,6,7,8-tetrahydro-2-phenyl-oxazolo[5,4-*b*]quinoline (**13**). Following the **General Method for the Friedländer reaction**, $AlCl_3$ (577.5 mg, 4.33 mmol) in 1,2-dichloroethane (24 mL) was reacted with cyclohexanone (447.7 μL, 4.33 mmol) and oxazole (**1**) (399.7 mg, 2.16 mmol). The mixture was refluxed for 7 d. Work-up and chromatography (CH_2Cl_2-MeOH, 9.8:0.2) provided compound (**13**) (480 mg, 84%): mp 273-275ºC; IR (KBr) ν_{max} 3400, 3340, 3220, 2930, 2860, 1645, 1630, 1610, 1445, 1335, 1060, 1020 cm^{-1}; ^{1}H NMR (DMSO-d_6, 300 MHz) δ 8.21 (m, H2', H6', 2 H), 7.67 (m, H3', H4', H5', 3 H), 6.33 (br s, NH_2, 2 H), 2.86 (m, H8, 2 H), 2,60 (m, H5, 2H), 1.89 (m, H6, H7, 4 H); ^{13}C NMR (DMSO-d_6, 75.4 MHz) δ 158.1 (C9a)*, 157.2 (C2)*, 152.4 (C8a), 145.3 (C4), 131.1 (C4'), 129.1 (2 C, C3', C5'), 127.1 (C1'), 126.5 (2 C, C2', C6'), 117.6 (C3a), 111.8 (C4a), 32.9 (C8), 23.1 (C5), 22.5 (C7)**, 22.2 (C6)**; MS (EI) *m*/z (%): 265 [M]$^+$ (100), 264 (61), 250 (21), 249 (5), 238 (10), 237 [M-CO]$^+$ (52), 236 (9), 133 (11), 106 (12), 105 (22), 104 (13), 79 (11), 78 (7), 77 [Ph]$^+$ (23). Anal. Calcd for $C_{16}H_{15}N_3O$: C, 72.43; H, 5.70; N, 15.84. Found: C, 72.34; H, 5.71; N, 15.71.

4-Amino-5,6,7,8-tetrahydro-2-(3'-pyridyl)-oxazolo[5,4-*b*]quinoline (**14**). Following the **General Method for the Friedländer reaction**, $AlCl_3$ (334.3 mg, 2.51 mmol, 1.5 equiv) in 1,2-dichloroethane (44 mL) was reacted with cyclohexanone (252 μL; 2,43 mmol, 1.5 equiv) and oxazole (**3**) (302.5 mg, 1.62 mmol). The mixture was refluxed for 15 d. Work-up and chromatography (CH_2Cl_2-MeOH, 9.8:0.2) produced compound (**14**) (78.3 mg, 18%): yellow solid, mp 263-265 ºC; IR (KBr) ν_{max} 3413, 3329, 3215, 2927, 2856, 1642, 1624, 1465, 1425, 1335, 1284, 1062, 1015 cm^{-1}; ^1H NMR (DMSO-d_6, 300 MHz) δ 9.24 (d, $J_{2',4'}$= 2.2 Hz, H2', 1 H), 8.72 (dd, $J_{6',5'}$= 4.8 Hz, $J_{6',4'}$= 1.6 Hz, H6', 1 H), 8.41 (ddd, $J_{4',5'}$= 8.0 Hz, $J_{4',2'}$= 2.2 Hz, $J_{4',6'}$= 1.6 Hz, H4', 1 H), 7.61 (dd, $J_{5',4'}$= 8.0 Hz, $J_{5',6'}$= 4.8 Hz, H5', 1 H), 6.49 (br s, NH_2, 2 H), 3.15 (m, H8, 2 H), 2.49 (m, H5, 2 H), 1.76 (m, H6, H7, 4 H); ^{13}C NMR (DMSO-d_6, 75.4 MHz) δ 158.2 (C9a), 155.4 (C2)*, 153.1 (C8a)*, 151.8 (C6'), 147.5 (C2'), 145.9 (C4), 134.3 (C4'), 124.7 (C5')$^{\#}$, 123.6 (C3')$^{\#}$, 117.5 (C3a), 112.2 (C4a), 33.1 (C8), 23.4 (C5), 22.7 (C7)**, 22.4 (C6)**; MS (EI)-*m*/*z* (%): 266 $[M]^+$ (100), 251 (21), 238 (61), 133 (16), 119 (5), 105 (20), 78 (19), 66 (7), 51 (7). Anal. Calcd for $C_{15}H_{14}N_4O$: C, 67.65; H, 5.30; N, 21.04. Found: C, 67.34; H, 5.19; N, 20.92.

4-Amino-5,6,7,8-tetrahydro-2-(4'-pyridyl)-oxazolo[5,4-*b*]quinoline (**15**). Following the **General Method for the Friedländer reaction**, $AlCl_3$ (712.23 mg, 5.37 mmol, 4 equiv) in 1,2-dichloroethane (30 mL) was reacted with cyclohexanone (556.7 μL, 5.37 mmol, 4 equiv) and oxazole (**4**) (250 mg, 1.34 mmol). The mixture was refluxed for 17 d. Work-up and chromatography (CH_2Cl_2-MeOH, 9.98:0.02) afforded compound (**15**) (39 mg, 11 %): yellow solid, mp 317-320 ºC; IR (KBr) ν_{max} 3413, 3322, 3211, 3033, 2931, 2859, 1610, 1593, 1468, 1443, 1409, 1335, 1286 cm^{-1}; ^1H NMR (DMSO-d_6, 300 MHz) δ 8.80 (m, H2', H6', 2 H), 7.98 (m, H3', H5', 2 H), 6.38 (br s, NH_2, 2 H), 3.15 (m, H8, 2 H), 2.50 (m, H5, 2 H), 1.80 (m, H6, H7, 4 H); ^{13}C NMR (DMSO-d_6, 75.4 MHz) δ 158.9 (C9a), 155.9 (C2)*, 154.5 (C8a)*, 151.5 (2 C, C2', C6'), 146.6 (C4), 134.9 (C4'), 120.9 (2 C, C3', C5'), 118.5 (C3a), 112.8 (C4a), 33.8 (C8), 23.9 (C5), 23.3 (C7), 22.9 (C6); MS (EI) *m*/*z* (%): 266 $[M]^+$ (100), 265 (46), 251 (14), 238 (4), 134 (7), 133 (10), 79 (10), 78 $[py]^+$ (14), 77 (8), 53 (5), 52 (6), 51 (9). Anal. Calcd for $C_{15}H_{14}N_4O$: C, 67.65; H, 5.30; N, 21.04. Found: C, 67.12; H, 5.02; N, 20.79.

4-Amino-6,7,8,9-tetrahydro-2-phenyl-5*H*-cyclohepta[*e*]oxazolo[5,4-*b*]pyridine (**16**). Following the **General Method for the Friedländer reaction**, $AlCl_3$ (576.7 mg, 4.33 mmol, 4.7 equiv) in 1,2-dichloroethane (30 mL) was reacted with cycloheptanone (509.5 μL, 4.33 mmol, 4.7 equiv) and oxazole (**1**) (200 mg, 0.92 mmol). The mixture was refluxed for 7 d. Work-up and chromatography (CH_2Cl_2-MeOH, 9.8:0.2) gave compound (**16**) (543.4 mg, 90%): white crystals; mp 281-283 ºC; IR (KBr) ν_{max} 3390, 3328, 3193, 2957, 2913, 2845, 1651, 1623, 1482, 1447, 1333, 1281, 1246, 1055 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz) δ 8.06 (m, H2', H6', 2 H), 7.55 (m, H3', H4', H5', 3 H), 6.15 (br s, NH_2, 2 H), 2.88 (m, H9, 2 H), 2.71 (m, H5, 2 H), 1.77 (m, H7, 2 H), 1.54 (m, H6, H8, 4 H); MS (EI) *m*/*z* (%): 279

$[M]^+$ (100), 278 (34), 264 (28), 251 $[M\text{-}CO]^+$ (32), 250 (50), 225 (11), 119 (10), 105 (18), 104 (13), 77 $[Ph]^+$ (24), 65 (10). Anal. Calcd for $C_{17}H_{17}N_3O$: C, 73.10; H, 6.13; N, 15.04. Found: C, 73.02; H, 6.40; N, 15.07.

4-Amino-6,7,8,9-tetrahydro-2-(3'-pyridyl)-5*H*-cyclohepta[*e*]oxazolo[5,4-*b*]pyridine (**17**). Following the **General Method for the Friedländer reaction**, $AlCl_3$ (334.5 mg, 2.51 mmol, 1.5 equiv) in 1,2-dichloroethane (45 mL) was reacted with cycloheptanone (286 μL; 2.42 mmol, 1.5 equiv) and oxazole (**3**) (301.4 mg, 1.62 mmol). The mixture was refluxed for 14 d. Work-up and chromatography (CH_2Cl_2-MeOH, 9.8:0.2) rendered compound (**17**) (91.2 mg, 20%): yellow crystals; mp 274-276 ºC; IR (KBr) ν_{max} 3420, 3350, 3210, 2930, 2870, 1660, 1630, 1490, 1435, 1345, 1075, 1025 cm^{-1}; ^1H NMR (DMSO-d_6, 300 MHz, 70 ºC) δ 9.23 (d, $J_{2',4'}$= 1.9 Hz, H2', 1 H), 8.70 (dd, $J_{6',5'}$= 4.8 Hz, $J_{6',4'}$= 1.7 Hz, H6', 1 H), 8.39 (ddd, $J_{4',5'}$= 8.0 Hz, $J_{4',2'}$= 1.9 Hz, $J_{4',6'}$= 1,7 Hz, H4', 1 H), 7.58 (dd, $J_{5',4'}$= 80 Hz, $J_{5',6'}$= 4.8 Hz, H5', 1 H), 6.17 (br s, NH_2, 2 H), 2.90 (m, H9, 2 H), 2.72 (m, H5, 2 H), 1.78 (m, H7, 2 H), 1.57 (m, H6, H8, 4 H); ^{13}C NMR (DMSO-d_6, 75.4 MHz, 70ºC) δ 160.0 (C9a), 157.2 (C2)*, 155.5 (C10a)*, 151.4 (C6'), 147.2 (C2'), 144.7 (C4), 133.8 (C4'), 124.1 (C5'), 123.1 (C3'), 117.2 (C3a), 111.6 (C4a), 38.2 (C9), 31.2 (C7), 26.8 (C6)*, 26.0 (C8)*, 24.7 (C5); MS (EI) *m*/*z* (%): 280 $[M]^+$ (100), 265 (25), 252 $[M\text{-}CO]^+$ (31), 251 (46), 239 (8), 226 (10), 119 (6), 105 (8), 78 (9). Anal. Calcd for $C_{16}H_{16}N_4O$: C, 68.55; H, 5.75; N, 19.99. Found: C, 68.32; H, 5.45; N, 19.75.

Pharmacological protocol: Evaluation of biological activity. To evaluate the biological activity of compounds **3**, **13**, **14**, **16** and **17** AChE inhibition was assayed in comparison with tacrine (**18**) as reference compound. The inhibitory potency against AChE was determined based on an adaptation of the method of Ellman *et al*. (1961),[15] using AChE from bovine erythrocytes and acethylthiocholine iodide (Sigma-Aldrich, St Louis, MO, USA) as substrate. The enzyme activity was measured by registering the absorbance increase due to substrate hydrolysis by AChE, and reaction of the produced thiol with DTNB (Sigma) yielding a yellow colored 5-thio-2-nitro-benzoic acid (TNB). Briefly, the reaction was performed in a 3 mL final volume by addition of 0.025 units AChE, 333 μM DTNB, 30 μL of each dilution of the test compounds in dimethylsulfoxide (DMSO), or vehicle, and 0.5 mM acethylthiocholine iodide to 100 mM phosphate-buffered solution, pH 8.0. After 15 min incubation at 25ºC, the reaction was stopped by addition of 33 μM eserine (Fluka, Schweiz, Switzerland). The color production was measured against a blank performed with no addition of substrate, in a quartz cuvette of 1 cm path-length, at 412 nm, in a Unicam UV2 spectrophotometer (Unicam Limited, UV2, Cambridge, UK), at 25ºC. Inhibition curves were performed with several concentrations of each compound up to their solubility limit in DMSO. The percent of inhibition by the test compounds was calculated considering the absorbance value obtained for vehicle as 100% AChE activity. Results are presented as means ± SD of at least two independent experiments, performed in quadruplicate.

ACKNOWLEDGEMENTS

E.G-S. thanks CSIC for a I3P fellowship.

REFERENCES

1. J. A. Joule and K. Mills, 'Heterocyclic Chemistry', Blackwell Science Ltd, Oxford, 2000; ed. by G. W. Gribble and T. L. Gilchrist, 'Comprehensive Heterocyclic Chemistry', Vol. XIII, Pergamon, London, 2001, pp. 238-260; V. Kouznetsov, L. Y. V. Méndez, and C. M. M. Gómez, *Curr. Org. Chem.*, 2005, **9**, 141; M. Abass, *Heterocycles*, 2005, **65**, 901.
2. J. P. Michael, *Nat. Prod. Rep.,* 1997, **14**, 605.
3. G. R. Newkome and W. W. Paudler, 'Contemporary Heterocyclic Chemistry', Wiley, New York, 1982, pp. 199-231; A. R. Katritzky, 'Handbook of Heterocyclic Chemistry', Pergamon Press, Oxford, 1985.
4. C.-C. Cheng and S.-J. Yan, *Org. React.,* 1982, **28**, 37.
5. J. M. Muchowski and M. L. Maddox, *Can. J. Chem.,* 2004, **82**, 461.
6. E. A. Fehnel, *J. Heterocycl. Chem.*, 1967, **4**, 565.
7. N. Desbois, J.-M. Chezal, F. Fauvelle, J.-C. Debouzy, C. Lartigue, A. Gueiffier, Y. Blache, E. Moreau, J.-C. Madelmont, O. Chavignon, and J.-C. Teulade, *Heterocycles,* 2005, **65**, 1121; M. Björk and S. Grivas, *Heterocycles,* 2005, **65**, 2369.
8. P. Camps, E. Gómez, D. Muñoz-Torrero, A. Badía, N. M. Vivas, X. Barril, M. Orozco, and F. J. Luque, *J. Med. Chem.,* 2001, **44**, 4733; M. T. McKenna, G. R. Proctor, L. C. Young, and A. L. Harvey, *J. Med. Chem.,* 1997, **40**, 3516; E. J. Barreiro, C. A. Camara, H. Verli, L. Brazil-Más, N. G. Castro, W. M. Cintra, Y. Aravaca, C. R. Rodríguez, and C. A. Fraga, *J. Med. Chem.,* 2003, **46**, 1144.
9. A. Dondoni, G. Fantin, M. Fogagnolo, A. Mastellari, A. Medici, and P. Pedrini, *J. Org. Chem.,* 1984, **49**, 3478.
10. J. L. Marco, C. de los Ríos, A. G. García, M. Villarroya, M. C. Carreiras, C. Martins, A. Eleuterio, A. Morreale, M. Orozco, and F. J. Luque, *Bioorg. Med. Chem.,* 2004, **12**, 2199.
11. F. Freeman and S. H.-L. Kim, *Tetrahedron Lett.,* 1989, **30**, 2631.
12. C. A. Maryanoff, 'The Chemistry of Heterocyclic Compounds: Oxazoles', Vol. 45, ed. by I. J. Turchi, John Wiley & Sons, New York, 1986, pp. 343-360.
13. D. K. Lahiri, J. T. Rogers, N. H. Greig, and K. Sambamurti, *Curr. Pharm. Des.,* 2004, **10**, 3111.
14. E. Scarpini, P. Scheltens, and H. Feldman, *Lancet Neurol.,* 2003, **2**, 539.
15. G. L. Ellman, K. D. Courtney, V. Jr. Andres, and R. M. Featherstone, *Biochem. Pharmacol.,* 1961, **7**, 88.