

An efficient one-pot synthesis and *in vitro* antimicrobial activity of new pyridine derivatives bearing the tetrazoloquinoline nucleus

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Abstract

A new series of 2-amino-3-cyano-4-tetrazoloquinolinyipyridine derivatives has been synthesized by the one-pot cyclocondensation reaction of a tetrazolo[1,5-*a*]quinoline-4-carbaldehyde, malononitrile, a heterocyclic/aromatic methyl ketone and ammonium acetate. All the synthesized compounds were subjected to *in vitro* antimicrobial screening against a panel of pathogenic strains of bacteria and fungi. Some of the compounds were found to be equipotent or more potent than commercial antibiotics as evident from the results.

Keywords: 2-Amino-3-cyano-4-tetrazoloquinolinyipyridine, tetrazolo[1,5-*a*]quinoline-4-carbaldehyde, one-pot synthesis, antimicrobial activity, MIC

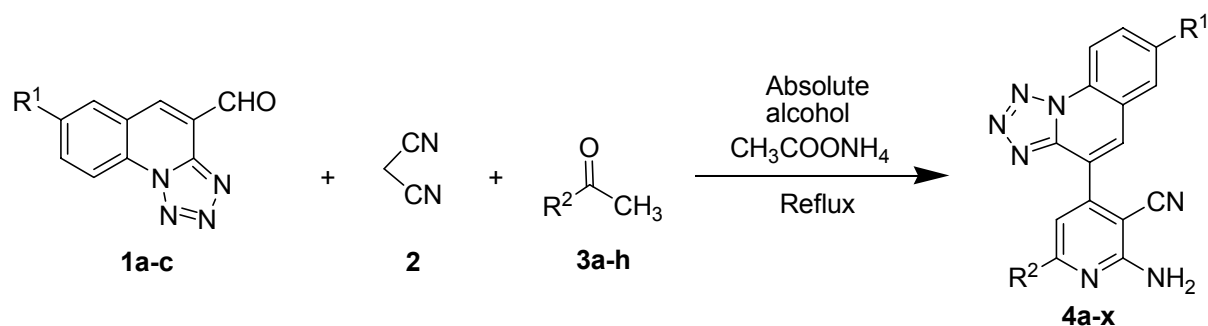
Introduction

Many naturally occurring and synthetic compounds bearing pyridine scaffold possess interesting biological properties.¹ In association with those, 2-amino-3-cyanopyridine derivatives have been identified as IKK- β inhibitors² along with its importance and utility as intermediates in preparing variety of heterocyclic compounds.³ Consequently, the synthesis of 2-amino-3-cyanopyridine derivatives keeps on attracting much interest in organic chemistry. Various routes for the synthesis of 2-amino-3-cyanopyridine derivatives have been reported using two-component as well as three-component reactions.⁴⁻⁷ Tu and coworkers have reported a facile synthesis of 2-amino-3-cyanopyridine derivatives in a one-pot reaction using aromatic aldehyde, methyl ketone, malononitrile and ammonium acetate.⁴ A literature survey shows that a number of pyridine derivatives have been synthesized using various aldehydes but not a single reference have been found where tetrazolo[1,5-*a*]quinoline-4-carbaldehyde is used. We wish to report herein this heterocyclic aldehyde which is biologically active⁸⁻¹⁰ with a view to obtaining more active heterocyclic system containing two biologically active moieties quinoline¹¹⁻¹³ and pyridine^{14,15} together. The most suitable protocol for the synthesis of functionalized organic compounds would be a one-pot reaction due to the fact that the synthesis can be performed without the

isolation of the intermediates, without discharging any functional groups in short reaction time.¹⁶ Hence, in the present investigation, we report an efficient one-pot multicomponent synthesis of 2-amino-3-cyanopyridine derivatives having tetrazoloquinoline nucleus which have also been recognized as promising new scaffold to endow good biological properties^{17,18} such as anti-inflammatory and antimicrobial activity.¹⁹⁻²²

Results and Discussion

In the present study, an effort has been made to undertake the synthesis of 2-amino-6-het/aryl-4-(7-(un)-substituted-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitriles through a one step process. For this purpose, the required tetrazolo[1,5-*a*]quinoline derivatives **1a-c** were prepared from 2-chloro-3-formylquinoline and sodium azide by our known literature process.²³ The target compounds **4a-x** were prepared in moderate to good yield (52-77%) by the reaction of tetrazolo[1,5-*a*]quinoline-4-carbaldehyde **1a-c**, malononitrile **2**, (het)aryl methyl ketone **3a-h** and ammonium acetate in absolute alcohol (Scheme 1). The formation of compounds **4a-x** may proceed *via* imine formed from ketone and ammonium acetate, imine reacts with alkylidenemalononitrile formed from Knoevenagel condensation of aldehyde and malononitrile, followed by cycloaddition, isomerization and aromatization to afford the 2-amino-3-cyano-4-tetrazoloquinolinylpyridine derivatives **4a-x**. The identity of the product was determined by IR, ¹H NMR, and ¹³C NMR spectral studies. The IR spectrum of compound **4a** exhibited absorption at 3410 cm⁻¹ (asymmetric N-H stretching) and 3314 cm⁻¹ (symmetric N-H stretching) for -NH₂, 2214 cm⁻¹ for -CN, 3015 cm⁻¹ for (aromatic C-H stretching) and 1400 to 1600 cm⁻¹ for (C=C aromatic and C=N stretching of pyridine). The ¹H NMR spectra of compound **4a** showed the absence of the aldehyde proton, moreover singlets at δ 7.06 ppm and multiplets at δ 7.20-8.71 ppm appeared for amine and aromatic protons respectively. The ¹³C NMR spectrum is in good agreement with the structure assigned. All the aromatic carbons of compounds **4a** showed signals around δ 115.7–154.3 ppm in the ¹³C NMR spectra. The signal at δ 96.0 ppm is assigned to carbon attached with carbonitrile. Besides, the structure of the compound was well confirmed by its mass spectral studies. Mass spectra of compound **4i** and **4l** gave molecular ion peak at *m/z* 378 (*M*+1) and *m/z* 368 (*M*+1) corresponding to molecular formula C₂₂H₁₅N₇ and C₂₀H₁₃N₇O respectively (Scheme 1). The elemental analysis values are in good agreement with theoretical data. Similarly, all these compounds were characterized on the basis of spectral studies. All the compounds were screened for their antibacterial and antifungal activity.



	R ¹	R ²		R ¹	R ²
4a	H	C ₆ H ₅	4m	CH ₃	2-thienyl
4b	H	4-CH ₃ C ₆ H ₄	4n	CH ₃	2-pyridyl
4c	H	4-OCH ₃ C ₆ H ₄	4o	CH ₃	3-pyridyl
4d	H	2-furyl	4p	CH ₃	4-pyridyl
4e	H	2-thienyl	4q	OCH ₃	C ₆ H ₅
4f	H	2-pyridyl	4r	OCH ₃	4-CH ₃ C ₆ H ₄
4g	H	3-pyridyl	4s	OCH ₃	4-OCH ₃ C ₆ H ₄
4h	H	4-pyridyl	4t	OCH ₃	2-furyl
4i	CH ₃	C ₆ H ₅	4u	OCH ₃	2-thienyl
4j	CH ₃	4-CH ₃ C ₆ H ₄	4v	OCH ₃	2-pyridyl
4k	CH ₃	4-OCH ₃ C ₆ H ₄	4w	OCH ₃	3-pyridyl
4l	CH ₃	2-furyl	4x	OCH ₃	4-pyridyl

Scheme 1. General synthetic route of 2-amino-3-cyano-4-tetrazoloquinolinyipyridine **4a-x**.

Antimicrobial activity

The *in vitro* antimicrobial activity of all the synthesized compounds was carried out by broth microdilution method.²⁴ Mueller Hinton broth was used as nutrient medium to grow and dilute the compound suspension for the test bacteria and Sabouraud Dextrose broth used for fungal nutrition. Inoculum size for test strain was adjusted to 10⁸ CFU [Colony Forming Unit] per milliliter by comparing the turbidity. The strains employed for the activity were procured from [MTCC – Micro Type Culture Collection] Institute of Microbial Technology, Chandigarh.

The compounds **4a-x** were screened for their antibacterial activity against *Bacillus subtilis* (MTCC 441), *Clostridium tetani* (MTCC 449), *Streptococcus pneumoniae* (MTCC 1936), *Escherichia coli* (MTCC 443), *Salmonella typhi* (MTCC 98), *Vibrio cholerae* (MTCC 3906) as well as antifungal activity against *Aspergillus fumigatus* (MTCC 3008) and *Candida albicans* (MTCC 227). DMSO was used as vehicle to get desired concentration of compounds to test upon microbial strains. The lowest concentration, which showed no visible growth after spot subculture was considered as MIC for each compound. The standard antibiotics used for

comparison in the present study were ampicillin for evaluating antibacterial activity as well as griseofulvin and nystatin for antifungal activity. The protocols are summarized in (Table 1).

Table 1. Antimicrobial activity of compounds **4a-x**

Compd.	Minimum inhibitory concentration in $\mu\text{g/mL}$							
	Gram positive bacteria			Gram negative bacteria			Fungi	
	<i>Bacillus subtilis</i>	<i>Clostridium tetani</i>	<i>Streptococcus pneumoniae</i>	<i>Escherichia coli</i>	<i>Salmonella typhi</i>	<i>Vibrio cholerae</i>	<i>Aspergillus fumigatus</i>	<i>Candida albicans</i>
	MTCC 441	MTCC 449	MTCC 1936	MTCC 443	MTCC 98	MTCC 3906	MTCC 3008	MTCC 227
4a	1000	250	500	125	125	200	1000	1000
4b	500	200	100	500	500	500	1000	500
4c	1000	150	100	125	500	250	1000	500
4d	500	250	500	500	500	250	500	250
4e	500	250	500	100	150	500	1000	500
4f	500	500	200	500	200	100	>1000	>1000
4g	250	200	500	250	500	500	250	250
4h	500	500	500	500	500	250	500	500
4i	250	250	250	100	200	250	>1000	>1000
4j	250	250	200	100	200	250	500	250
4k	250	125	100	250	500	500	1000	>1000
4l	250	100	100	200	150	500	1000	500
4m	200	500	200	62.5	100	100	1000	500
4n	150	250	250	125	200	250	>1000	1000
4o	500	500	62.5	125	125	62.5	>1000	1000
4p	250	500	125	100	150	500	1000	500
4q	125	250	250	250	125	100	1000	500
4r	100	500	500	1000	1000	500	>1000	250
4s	1000	100	500	1000	500	500	1000	500
4t	125	500	250	100	100	500	1000	200
4u	250	500	100	125	200	500	500	500
4v	250	200	500	125	500	250	500	250
4w	125	250	250	100	50	250	1000	500
4x	500	500	250	500	125	500	1000	1000
Ampi.	250	250	100	100	100	100	-	-
Grise.	-	-	-	-	-	-	100	500
Nyst.	-	-	-	-	-	-	100	100

Ampi.: Ampicillin, Grise.: Griseofulvin, Nyst.: Nystatin

An examination of the data (Table 1) reveals that amongst all the synthesized compounds **4a-x**, compound **4o** exhibited excellent activity against Gram positive bacteria *Streptococcus pneumoniae* and Gram negative bacteria *Vibrio cholerae* while compounds **4m** and **4w** are found to be highly active against Gram negative bacteria *Escherichia coli* and *Salmonella typhi* respectively as compared to standard antibiotic ampicillin.

Compounds **4m**, **4n**, **4q**, **4r**, **4t** and **4w** are found to be more potent as compared to standard antibiotic ampicillin against Gram positive bacteria *Bacillus subtilis*. In case of Gram positive bacteria *Clostridium tetani*, compounds **4b**, **4c**, **4g**, **4k**, **4l**, **4s** and **4v** are found to be more potent than ampicillin.

Antifungal study revealed that compounds **4d**, **4g**, **4j**, **4r**, **4t** and **4v** are more potent as compared to standard fungicidal griseofulvin against *Candida albicans*. Most of the compounds were not found sufficiently potent to inhibit *Aspergillus fumigatus*.

Conclusions

A series of some new 2-amino-3-cyano-4-tetrazoloquinolinylpyridine derivatives has been synthesized through a facile one-pot multicomponent reaction. This synthetic strategy allows the construction of relatively complicated nitrogen containing heterocyclic system as well as the introduction of various aromatic and heteroaromatic substitutions into 4- and 6- positions of pyridine. It can be concluded from Table 2 that compound **4o** having methyl group on tetrazoloquinoline nucleus and 3-pyridyl substitution on pyridine is highly active against *Streptococcus pneumoniae* as well as *Vibrio cholerae*. From the activity data, it is worth mentioning that minor change in molecular configuration of these compounds profoundly influences the activity.

Experimental Section

General. All the reagents were obtained commercially and used with further purification. All melting points were taken in open capillaries and are uncorrected. The monitoring of the progress of all reactions and homogeneity of the synthesized compounds was carried out by TLC. TLC was run using TLC aluminum sheets silica gel 60 F₂₅₄ (Merck). Elemental analysis (% C, H, N) was carried out by Perkin Elmer 2400 CHN elemental analyzer at Sophisticated Instrumentation Centre for Applied Research & Training (SICART), Vallabh Vidyanagar. IR spectra were recorded on a Shimadzu FTIR 8401 spectrophotometer in KBr. ¹H NMR and ¹³C NMR spectra were recorded on a Bruker Avance 400 MHz spectrometer using solvent peak as internal standard. Mass spectra were scanned on a Shimadzu LCMS 2010 spectrometer.

General procedure for the synthesis of 2-amino-6-het/aryl-4-(7-(un)-substituted-tetrazolo[1,5-*a*]quinolin-4-yl)pyridine-3-carbonitriles **4a-x**

7-(Un)substituted-tetrazolo[1,5-*a*]quinoline-4-carbaldehyde **1a-c** (5 mmole), malononitrile **2** (5 mmole), (het)aryl methyl ketone **3a-h** (5 mmole), ammonium acetate (40 mmole) and absolute alcohol (15 ml) were charged in a 50 ml round bottom flask. Then, the reaction mixture was refluxed for 2 to 2.5 hr. Progress of reaction was monitored by the TLC. After the completion of reaction, the reaction mixture was cooled to room temperature and stirred for 0.5 hr. The resulting solid was collected by filtration and washed well with absolute alcohol to obtain the pure solid sample of product **4a-x**.

2-Amino-6-phenyl-4-(tetrazolo[1,5-*a*]quinolin-4-yl)pyridine-3-carbonitrile 4a. Yield 72%, m.p. 264 °C, Anal. Calcd. for C₂₁H₁₃N₇ : C 69.41, H 3.60, N 26.98% Found: C 69.32, H 3.56, N 26.72%. IR (KBr, cm⁻¹): 3410, 3314 (NH₂), 2214 (CN), 3015 (ArC-H). ¹H NMR (400 MHz, DMSO-*d*₆): δ 7.20-8.71 (m, 11H, Ar-H), 7.06 (s, 1H, NH₂). ¹³C NMR (400 MHz, DMSO-*d*₆): δ: 96.0 (C-CN), 115.7, 116.2, 116.7, 120.0, 122.5, 123.9, 128.9, 129.2, 130.1, 130.4, 130.6, 133.0, 134.9, 137.7, 143.1, 146.9, 150.4, 154.3 (Ar-C).

2-Amino-6-(4-methylphenyl)-4-(tetrazolo[1,5-*a*]quinolin-4-yl)pyridine-3-carbonitrile 4b. Yield 65%, m.p. 276 °C, Anal. Calcd. for C₂₂H₁₅N₇ : C 70.01, H 4.00, N 25.97% Found: C 69.72, H 3.92, N 25.91%. IR (KBr, cm⁻¹): 3405, 3312 (NH₂), 2210 (CN), 3005 (ArC-H). ¹H NMR (400 MHz, DMSO-*d*₆): δ 2.394 (s, 3H, CH₃), 7.03 (s, 1H, NH₂), 7.18-8.71 (m, 10H, Ar-H). ¹³C NMR (400 MHz, DMSO-*d*₆): δ: 21.3 (CH₃), 96.0 (C-CN), 115.8, 116.8, 119.9, 122.5, 124.0, 129.1, 130.4, 133.0, 134.8, 138.3, 140.7, 143.0, 146.4, 146.8, 150.5, 154.4, 160.4, 166.9 (Ar-C).

2-Amino-6-(4-methoxyphenyl)-4-(tetrazolo[1,5-*a*]quinolin-4-yl)pyridine-3-carbonitrile 4c. Yield 66%, m.p. 256-259 °C, Anal. Calcd. for C₂₂H₁₅N₇O : C 67.16, H 3.84, N 24.92% Found: C 67.13, H 3.71, N 25.01%. IR (KBr, cm⁻¹): 3400, 3310 (NH₂), 2180 (CN), 3000 (ArC-H). ¹H NMR (400 MHz, DMSO-*d*₆): δ 3.97 (s, 3H, OCH₃), 7.10 (s, 1H, NH₂), 7.30-8.51 (m, 10H, Ar-H). ¹³C NMR (400 MHz, DMSO-*d*₆): δ: 56.3 (OCH₃), 95.8 (C-CN), 116.8, 117.5, 119.0, 123.8, 124.9, 128.1, 131.2, 133.0, 134.6, 138.4, 141.7, 143.4, 146.4, 146.2, 151.5, 154.7, 161.4, 166.0 (Ar-C).

2-Amino-6-(2-furyl)-4-(tetrazolo[1,5-*a*]quinolin-4-yl)pyridine-3-carbonitrile 4d. Yield 76%, m.p. 273 °C, Anal. Calcd. for C₁₉H₁₁N₇O : C 64.56, H 3.13, N 27.74% Found: C 64.62, H 3.11, N 27.67%. IR (KBr, cm⁻¹): 3416, 3314 (NH₂), 2190 (CN), 3005 (ArC-H). ¹H NMR (400 MHz, DMSO-*d*₆): δ 7.42 (s, 2H, NH₂), 7.52-8.61 (m, 9H, Ar-H). ¹³C NMR (400 MHz, DMSO-*d*₆): δ: 94.7 (C-CN), 116.4, 119.2, 121.7, 124.4, 128.5, 129.6, 130.1, 134.1, 136.5, 137.7, 138.4, 142.5, 146.4, 148.5, 148.7, 151.4, 153.5, 154.5 (Ar-C).

2-Amino-6-(2-thienyl)-4-(tetrazolo[1,5-*a*]quinolin-4-yl)pyridine-3-carbonitrile 4e. Yield 68%, m.p. 243-245 °C, Anal. Calcd. for C₁₉H₁₁N₇S : C 61.77, H 3.00, N 26.54% Found: C 61.66, H 2.98, N 26.49%. IR (KBr, cm⁻¹): 3414, 3312 (NH₂), 2230 (CN), 3016 (ArC-H). ¹H NMR (400 MHz, DMSO-*d*₆): δ 7.16 (s, 2H, NH₂), 7.30-8.54 (m, 9H, Ar-H). ¹³C NMR (400 MHz, DMSO-*d*₆): δ: 93.8 (C-CN), 115.4, 117.2, 120.7, 123.4, 126.9, 129.0, 130.1, 133.5, 136.5, 137.0, 137.4, 141.5, 144.8, 147.5, 148.4, 150.4, 153.8, 155.1 (Ar-C).

2-Amino-6-(2-pyridyl)-4-(tetrazolo[1,5-*a*]quinolin-4-yl)pyridine-3-carbonitrile 4f. Yield 77%, m.p. 265 °C, Anal. Calcd. for C₂₀H₁₂N₈ : C 65.92, H 3.32, N 30.75% Found: C 66.08, H 3.24, N 30.61%. IR (KBr, cm⁻¹): 3416, 3330 (NH₂), 2234 (CN), 3028 (ArC-H). ¹H NMR (400 MHz, DMSO-*d*₆): δ 7.26 (s, 2H, NH₂), 7.40-9.00 (m, 10H, Ar-H). ¹³C NMR (400 MHz, DMSO-*d*₆): δ: 98.1 (C-CN), 116.7, 117.6, 122.7, 124.4, 125.5, 128.8, 130.9, 132.1, 135.8, 136.6, 137.2, 142.7, 146.7, 148.2, 148.6, 150.3, 152.2, 157.9, 158.5 (Ar-C).

2-Amino-6-(3-pyridyl)-4-(tetrazolo[1,5-*a*]quinolin-4-yl)pyridine-3-carbonitrile 4g. Yield 69%, m.p. 270-273 °C, Anal. Calcd. for C₂₀H₁₂N₈ : C 65.92, H 3.32, N 30.75% Found: C 66.02, H 3.29, N 30.66%. IR (KBr, cm⁻¹): 3410, 3334 (NH₂), 2216 (CN), 3020 (ArC-H). ¹H NMR (400

MHz, DMSO-*d*₆): δ 7.22 (s, 2H, NH₂), 7.46-9.07 (m, 10H, Ar-H). ¹³C NMR (400 MHz, DMSO-*d*₆): δ : 98.9 (C-CN), 115.4, 118.2, 120.7, 123.4, 126.5, 128.0, 130.0, 133.1, 135.5, 136.3, 138.5, 143.5, 147.4, 148.5, 148.9, 151.3, 153.2, 156.1, 158.2 (Ar-C).

2-Amino-6-(4-pyridyl)-4-(tetrazolo[1,5-*a*]quinolin-4-yl)pyridine-3-carbonitrile 4h. Yield 72%, m.p. 258-260 °C, Anal. Calcd. for C₂₀H₁₂N₈ : C 65.92, H 3.32, N 30.75% Found: C 66.04, H 3.35, N 30.67%. IR (KBr, cm⁻¹): 3400, 3336 (NH₂), 2208 (CN), 3005 (ArC-H). ¹H NMR (400 MHz, DMSO-*d*₆): δ 7.30 (s, 2H, NH₂), 7.45-8.91 (m, 10H, Ar-H). ¹³C NMR (400 MHz, DMSO-*d*₆): δ : 97.0 (C-CN), 119.5, 121.4, 123.0, 126.7, 128.6, 129.7, 131.4, 136.4, 138.7, 141.0, 143.1, 145.5, 146.9, 148.4, 149.5, 151.2, 153.1 (Ar-C).

2-Amino-6-phenyl-4-(7-methyl-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile 4i.

Yield 71%, m.p. 274 °C, Anal. Calcd. for C₂₂H₁₅N₇ : C 70.01, H 4.00, N 25.97% Found: C 69.94, H 3.92, N 25.89%. IR (KBr, cm⁻¹): 3400, 3310 (NH₂), 2214 (CN), 3032 (ArC-H). ¹H NMR (400 MHz, DMSO-*d*₆): δ 2.50 (s, 3H, CH₃), 7.03-8.60 (m, 10H, Ar-H). ¹³C NMR (400 MHz, DMSO-*d*₆): δ : 21.3 (CH₃), 96.0 (C-CN), 115.8, 119.4, 121.5, 126.4, 126.9, 128.2, 130.9, 132.1, 134.4, 136.5, 138.8, 140.5, 144.9, 147.8, 149.3, 150.5, 153.4, 154.4 (Ar-C), MS: (M+1) 378.

2-Amino-6-(4-methylphenyl)-4-(7-methyl-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-

carbonitrile 4j. Yield 72%, m.p. 264 °C, Anal. Calcd. for C₂₃H₁₇N₇ : C 70.57, H 4.37, N 25.04% Found: C 70.42, H 4.25, N 25.01%. IR (KBr, cm⁻¹): 3405, 3300 (NH₂), 2240 (CN), 3026 (ArC-H). ¹H NMR (400 MHz, DMSO-*d*₆): δ 2.39 (s, 3H, CH₃), 2.45 (s, 3H, CH₃), 7.03 (s, 2H, NH₂), 7.18-8.71 (m, 9H, Ar-H). ¹³C NMR (400 MHz, DMSO-*d*₆): δ : 21.3 (CH₃), 23.4 (CH₃), 96.0 (C-CN), 115.8, 119.9, 122.5, 124.0, 128.9, 130.4, 130.6, 133.0, 133.3, 138.8, 139.9, 140.7, 145.0, 146.4, 146.8, 150.5, 154.4, 160.4 (Ar-C).

2-Amino-6-(4-methoxyphenyl)-4-(7-methyl-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-

carbonitrile 4k. Yield 70%, m.p. 243 °C, Anal. Calcd. for C₂₃H₁₇N₇O : C 67.80, H 4.20, N 24.06% Found: C 67.77, H 4.11, N 24.00%. IR (KBr, cm⁻¹): 3436, 3310 (NH₂), 2210 (CN), 3005 (ArC-H). ¹H NMR (400 MHz, DMSO-*d*₆): δ 2.50 (s, 3H, CH₃), 3.97 (s, 3H, OCH₃), 6.87 (s, 2H, NH₂), 7.03-8.62 (m, 9H, Ar-H). ¹³C NMR (400 MHz, DMSO-*d*₆): δ : 21.3 (CH₃), 56.3 (OCH₃), 96.4 (C-CN), 117.1, 121.9, 124.4, 126.7, 128.3, 128.9, 129.1, 132.7, 133.3, 136.0, 137.4, 138.6, 141.5, 143.0, 144.2, 145.8, 148.0, 150.4 (Ar-C).

2-Amino-6-(2-furyl)-4-(7-methyl-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile 4l.

Yield 73%, m.p. 278-280 °C, Anal. Calcd. for C₂₀H₁₃N₇O : C 65.38, H 3.56, N 26.68% Found: C 65.25, H 3.44, N 26.65%. IR (KBr, cm⁻¹): 3410, 3336 (NH₂), 2214 (CN), 3030 (ArC-H). ¹H NMR (400 MHz, DMSO-*d*₆): δ 2.50 (s, 3H, CH₃), 6.79 (s, 2H, NH₂), 7.03-8.63 (m, 8H, Ar-H). ¹³C NMR (400 MHz, DMSO-*d*₆): δ : 20.8 (CH₃), 96.7 (C-CN), 119.1, 120.4, 122.9, 124.0, 126.8, 126.9, 128.7, 130.0, 131.7, 133.2, 135.1, 136.3, 138.5, 139.9, 140.5, 141.7, 142.9, 146.1 (Ar-C), MS: (M+1) 368.

2-Amino-6-(2-thienyl)-4-(7-methyl-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile

4m. Yield 66%, m.p. 264-267 °C, Anal. Calcd. for C₂₀H₁₃N₇S : C 62.64, H 3.41, N 25.57% Found: C 62.60, H 3.39, N 25.42%. IR (KBr, cm⁻¹): 3400, 3316 (NH₂), 2230 (CN), 3005 (ArC-H). ¹H NMR (400 MHz, DMSO-*d*₆): δ 2.47 (s, 3H, CH₃), 6.84 (s, 2H, NH₂), 6.96-8.54 (m, 8H,

Ar-H). ^{13}C NMR (400 MHz, DMSO- d_6) δ : 21.6 (CH_3), 94.1 (C-CN), 114.1, 118.4, 122.6, 124.0, 126.0, 127.9, 128.7, 130.4, 131.1, 134.2, 135.1, 136.0, 137.5, 138.9, 141.0, 141.7, 142.4, 143.8 (Ar-C).

2-Amino-6-(2-pyridyl)-4-(7-methyl-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile

4n. Yield 70%, m.p. 252 °C, Anal. Calcd. for $\text{C}_{21}\text{H}_{14}\text{N}_8$: C 66.65, H 3.72, N 29.61% Found: C 65.70, H 3.64, N 29.58%. IR (KBr, cm^{-1}): 3430, 3326 (NH_2), 2230 (CN), 3016 (ArC-H). ^1H NMR (400 MHz, DMSO- d_6) δ : 2.21 (s, 3H, CH_3), 6.94 (s, 2H, NH_2), 7.23-8.78 (m, 9H, Ar-H). ^{13}C NMR (400 MHz, DMSO- d_6) δ : 21.9 (CH_3), 94.8 (C-CN), 115.6, 118.3, 121.4, 123.7, 125.5, 127.4, 130.0, 133.4, 133.9, 134.7, 135.6, 137.2, 139.0, 140.4, 144.6, 148.3, 152.0, 153.4, 154.9 (Ar-C).

2-Amino-6-(3-pyridyl)-4-(7-methyl-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile

4o. Yield 54%, m.p. 270-273 °C, Anal. Calcd. for $\text{C}_{21}\text{H}_{14}\text{N}_8$: C 66.65, H 3.72, N 29.61% Found: C 65.68, H 3.69, N 29.48%. IR (KBr, cm^{-1}): 3400, 3330 (NH_2), 2216 (CN), 3000 (ArC-H). ^1H NMR (400 MHz, DMSO- d_6) δ : 2.25 (s, 3H, CH_3), 7.04 (s, 2H, NH_2), 7.20-8.70 (m, 9H, Ar-H). ^{13}C NMR (400 MHz, DMSO- d_6) δ : 22.1 (CH_3), 95.6 (C-CN), 117.1, 119.3, 122.9, 123.0, 126.5, 128.4, 130.5, 133.0, 133.9, 134.5, 136.7, 137.0, 139.3, 141.5, 144.0, 148.1, 152.3, 154.2, 155.0 (Ar-C).

2-Amino-6-(4-pyridyl)-4-(7-methyl-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile

4p. Yield 68%, m.p. 267 °C, Anal. Calcd. for $\text{C}_{21}\text{H}_{14}\text{N}_8$: C 66.65, H 3.72, N 29.61% Found: C 65.61, H 3.80, N 29.55%. IR (KBr, cm^{-1}): 3408, 3338 (NH_2), 2236 (CN), 3018 (ArC-H). ^1H NMR (400 MHz, DMSO- d_6) δ : 2.20 (s, 3H, CH_3), 7.12 (s, 2H, NH_2), 7.26-8.77 (m, 9H, Ar-H). ^{13}C NMR (400 MHz, DMSO- d_6) δ : 21.8 (CH_3), 94.2 (C-CN), 117.1, 119.3, 122.9, 123.0, 126.5, 128.4, 130.5, 133.0, 133.9, 134.5, 136.7, 137.0, 139.3, 141.5, 144.0, 148.1, 152.3 (Ar-C).

2-Amino-6-phenyl-4-(7-methoxy-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile 4q.

Yield 62%, m.p. 243-245 °C, Anal. Calcd. for $\text{C}_{22}\text{H}_{15}\text{N}_7\text{O}$: C 67.16, H 3.84, N 24.92% Found: C 67.10, H 3.74, N 24.89%. IR (KBr, cm^{-1}): 3440, 3322 (NH_2), 2232 (CN), 3028 (ArC-H). ^1H NMR (400 MHz, DMSO- d_6) δ : 3.84 (s, 3H, OCH_3), 7.01 (s, 1H, NH_2), 7.16-8.31 (m, 10H, Ar-H). ^{13}C NMR (400 MHz, DMSO- d_6) δ : 56.8 (OCH_3), 96.0 (C-CN), 115.7, 116.2, 116.7, 120.0, 122.5, 123.9, 128.9, 129.2, 130.1, 130.4, 130.6, 133.0, 134.9, 137.7, 143.1, 146.9, 150.4, 154.3 (Ar-C).

2-Amino-6-(4-methylphenyl)-4-(7-methoxy-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile 4r.

Yield 70%, m.p. 274-276 °C, Anal. Calcd. for $\text{C}_{23}\text{H}_{17}\text{N}_7\text{O}$: C 67.80, H 4.20, N 24.06% Found: C 67.71, H 4.13, N 23.00%. IR (KBr, cm^{-1}): 3400, 3310 (NH_2), 2210 (CN), 2990 (ArC-H). ^1H NMR (400 MHz, DMSO- d_6) δ : 2.50 (s, 3H, CH_3), 3.86 (s, 3H, OCH_3), 6.91 (s, 2H, NH_2), 7.08-8.68 (m, 9H, Ar-H). ^{13}C NMR (400 MHz, DMSO- d_6) δ : 21.4 (CH_3), 55.9 (OCH_3), 97.2 (C-CN), 118.1, 120.9, 123.1, 126.7, 128.0, 128.9, 129.6, 133.1, 133.3, 136.4, 138.4, 138.6, 141.9, 143.0, 144.1, 145.3, 149.0, 152.4 (Ar-C).

2-Amino-6-(4-methoxyphenyl)-4-(7-methoxy-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile 4s.

Yield 52%, m.p. 292 °C, Anal. Calcd. for $\text{C}_{23}\text{H}_{17}\text{N}_7\text{O}_2$: C 65.24, H 4.04, N 23.15% Found: C 65.30, H 4.00, N 23.11%. IR (KBr, cm^{-1}): 3420, 3312 (NH_2), 2230 (CN), 3020

(ArC-H). ^1H NMR (400 MHz, DMSO- d_6): δ 3.90 (s, 3H, OCH₃), 3.97 (s, 3H, OCH₃), 6.98 (s, 1H, NH₂), 7.12-8.70 (9H, m, Ar-H). ^{13}C NMR (400 MHz, DMSO- d_6): δ : 54.9 (OCH₃), 56.3 (OCH₃), 95.8 ($\underline{\text{C}}$ -CN), 117.4, 121.9, 123.3, 123.6, 126.7, 128.9, 129.9, 129.6, 130.3, 133.4, 133.6, 136.9, 138.4, 139.6, 141.0, 143.5, 144.6, 145.3 (Ar-C).

2-Amino-6-(2-furyl)-4-(7-methoxy-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile

4t. Yield 70%, m.p. 266-268 °C, Anal. Calcd. for C₂₀H₁₃N₇O₂ : C 62.66, H 3.41, N 25.57% Found: C 62.60, H 3.39, N 25.52%. IR (KBr, cm⁻¹): 3440, 3326 (NH₂), 2200 (CN), 3035 (ArC--H). ^1H NMR (400 MHz, DMSO- d_6): δ 3.76 (s, 3H, OCH₃), 6.98 (s, 2H, NH₂), 7.41-8.94 (m, 8H, Ar-H). ^{13}C NMR (400 MHz, DMSO- d_6): δ : 56.1 (OCH₃), 96.3 ($\underline{\text{C}}$ -CN), 116.0, 117.8, 119.2, 120.2, 122.2, 124.4, 126.9, 129.3, 131.3, 134.1, 136.0, 138.1, 140.4, 143.8, 144.2, 146.9, 148.9, 152.1 (Ar-C).

2-Amino-6-(2-thienyl)-4-(7-methoxy-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile

4u. Yield 71%, m.p. 271-273 °C, Anal. Calcd. for C₂₀H₁₃N₇OS : C 60.14, H 3.28, N 24.54% Found: C 60.10, H 3.30, N 25.61%. IR (KBr, cm⁻¹): 3430, 3320 (NH₂), 2220 (CN), 3040 (ArC--H). ^1H NMR (400 MHz, DMSO- d_6): δ 3.45 (s, 3H, OCH₃), 7.21 (s, 2H, NH₂), 7.35-8.83 (m, 8H, Ar-H). ^{13}C NMR (400 MHz, DMSO- d_6): δ : 55.3 (OCH₃), 96.8 ($\underline{\text{C}}$ -CN), 117.8, 118.8, 119.2, 120.7, 123.2, 124.9, 126.5, 129.0, 133.3, 134.5, 136.6, 138.4, 141.4, 143.5, 144.2, 146.0, 148.2, 151.3 (Ar-C).

2-Amino-6-(2-pyridyl)-4-(7-methoxy-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile

4v. Yield 60%, m.p. 276 °C, Anal. Calcd. for C₂₁H₁₄N₈O : C 63.95, H 3.57, N 28.41% Found: C 64.02, H 3.60, N 28.38%. IR (KBr, cm⁻¹): 3410, 3318 (NH₂), 2214 (CN), 3000 (ArC-H). ^1H NMR (400 MHz, DMSO- d_6): δ 3.64 (s, 3H, OCH₃), 7.21 (s, 2H, NH₂), 7.33-8.71 (m, 9H, Ar-H). ^{13}C NMR (400 MHz, DMSO- d_6): δ : 55.4 (OCH₃), 96.7 ($\underline{\text{C}}$ -CN), 117.7, 118.0, 119.4, 120.8, 123.1, 125.9, 127.8, 129.0, 131.5, 135.4, 138.7, 140.1, 142.7, 145.6, 146.8, 148.1, 149.0, 150.4, 153.2 (Ar-C).

2-Amino-6-(3-pyridyl)-4-(7-methoxy-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile

4w. Yield 58%, m.p. 244-247 °C, Anal. Calcd. for C₂₁H₁₄N₈O : C 63.95, H 3.57, N 28.41% Found: C 63.88, H 3.57, N 28.41%. IR (KBr, cm⁻¹): 3416, 3310 (NH₂), 2228 (CN), 3020 (ArC--H). ^1H NMR (400 MHz, DMSO- d_6): δ 3.72 (s, 3H, OCH₃), 7.27 (s, 2H, NH₂), 7.30-8.74 (m, 9H, Ar-H). ^{13}C NMR (400 MHz, DMSO- d_6): δ : 56.9 (OCH₃), 97.2 ($\underline{\text{C}}$ -CN), 115.7, 117.0, 118.6, 120.2, 124.4, 126.0, 128.8, 129.7, 131.8, 136.4, 138.0, 140.4, 142.5, 143.0, 146.5, 148.8, 149.4, 151.0, 152.8 (Ar-C).

2-Amino-6-(4-pyridyl)-4-(7-methoxy-(tetrazolo[1,5-*a*]quinolin-4-yl))pyridine-3-carbonitrile

4x. Yield 70%, m.p. 258-260 °C, Anal. Calcd. for C₂₁H₁₄N₈O : C 63.95, H 3.57, N 28.41% Found: C 63.91, H 3.44, N 28.39%. IR (KBr, cm⁻¹): 3446, 3318 (NH₂), 2220 (CN), 3024 (ArC--H). ^1H NMR (400 MHz, DMSO- d_6): δ 3.85 (s, 3H, OCH₃), 7.04 (s, 2H, NH₂), 7.14-8.81 (m, 9H, Ar-H). ^{13}C NMR (400 MHz, DMSO- d_6): δ : 56.1 (OCH₃), 97.4 ($\underline{\text{C}}$ -CN), 120.4, 122.4, 125.3, 126.9, 128.3, 130.2, 131.0, 132.2, 133.3, 135.9, 137.4, 138.6, 141.6, 142.3, 144.8, 145.5, 147.5 (Ar-C).

Acknowledgements

The authors express their sincere thanks to the Department of Chemistry, S P University for providing research facilities, Vaibhav Analytical Laboratory, Ahmedabad for IR spectral analysis as well as Oxygen Healthcare Research Pvt. Ltd., Ahmedabad for providing mass spectrometry facilities. We are also thankful to Microcare Laboratory, Surat for antimicrobial activity.

References

1. Temple, C.; Rener, Jr. G. A.; Raud, W. R.; Noker, P. E. *J. Med. Chem.* **1992**, *35*, 3686.
2. Murata, T.; Shimada, M.; Sakakibara, S.; Yoshino, T.; Kadono, H.; Masuda, T.; Shimazaki, M.; Shintani, T.; Fuchikami, K.; Sakai, K.; Inbe, H.; Takeshita, K.; Niki, T.; Umeda, M.; Bacon, K. B.; Ziegelbauer, K. B.; Lowinger, T. B. *Bioorg. Med. Chem. Lett.* **2003**, *13*, 913.
3. (a) Shishoo, C. J.; Devani, M. B.; Bhadti, V. S.; Ananthan, S.; Ullas, G. V. *Tetrahedron Lett.* **1983**, *24*, 4611. (b) Doe, K.; Avasthi, K.; Pratap, R.; Bakuni, D. S.; Joshi, M. N. *Indian J. Chem.* **1990**, *29B*, 459.
4. Feng, S.; Shujiang, T.; Fang, F.; Tuanjie, L. *Arkivoc* **2005**, (i), 137.
5. Rafat, M. S.; Fathy, F. A. *J. Chem. Re. (S)*. **1997**, 294.
6. Vyas, D. H.; Tala, S. D.; Akbari, J. D.; Dhaduk, M. F.; Joshi, K. A. *Indian J. Chem.* **2009**, *48B*, 833.
7. Zhou, J. F.; Gong, G. X.; Zhu, F. X.; Zhi, S. J. *Chin. Chem. Lett.* **2009**, *20*, 37.
8. Dreikorn, B. A. U. S. Patent 3 764 681, 1973.
9. Wright, T. L. U. S. Patent 4 496 569, 1985.
10. Bekhit, A. A.; El-Sayed, O. A.; Al-Allaf, T. A. K.; Aboul-Enein, H. Y.; Kunhi, M.; Pulicat, S. M.; Al-Hussain, K.; Al-Khodairy, F.; Arif, J. *Eur. J. Med. Chem.* **2004**, *39*, 499.
11. Kumar, R. N.; Suresh, T.; Mohan, P. S. *Indian J. Chem.* **2003**, *42B*, 688.
12. Moiseev, I. K.; Zemtsova, M. N.; Trakhtenberg, P. L.; Kulikova, D. A.; Pskobkina, I.; Neschadim, G. N.; Ostapchuk, N. V. *Khim.-Farm Zh.* **1998**, *22*, 1448.
13. Strong; Lee, H. *Chem. Abstr.* **1997**, *126*, 17876 b.
14. Brahmabhatt, D. I.; Pandya, V. P.; Patel, C. N.; Patel, M. A. *Indian J. Chem.* **2005**, *44B*, 1863.
15. Cai, X. M.; Wang, Q. F.; Yan, C. G. *Synth. Commun.* **2007**, *37*, 223.
16. Boulard, L.; BouzBouz, S.; Cossy, J.; Franck, X.; Figadere, B. *Tetrahedron Lett.* **2004**, *45*, 6603.
17. Mukherjee, A.; Akhtar, M. S.; Sharma, V. L.; Seth, M.; Bhaduri, A. P.; Agnihotri, A.; Mehrotra, P. K.; Kamboj, V. P. *J. Med. Chem.* **1989**, *32*, 2297.
18. Gupta, R.; Gupta, A. K.; Paul, S. *Indian J. Chem.* **2000**, *39B*, 847.

19. Bekhit, A. A.; El-Sayed, O. A.; Aboulmagd, E.; Park, J. Y. *Eur. J. Med. Chem.* **2004**, *39*, 249.
20. Angibaud, P.; Bourdrez, X.; End, D. W. *Bio. & Med. Chem.* **2003**, *13*, 4365.
21. Bawa, S.; Kumar, S. *Indian J. Chem.* **2009**, *48B*, 142.
22. Thota, S.; Argade, A.; Singh, R.; Lu, H. H.; Huang, P., U. S. Patent 7358259B2, 2008.
23. Ladani, N. K.; Patel, M. P.; Patel, R. G. *Arkivoc* **2009**, (vii), 292.
24. National Committee for Clinical Laboratory Standards (NCCLS), 940, West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898, USA. *Performance Standards for Antimicrobial Susceptibility Testing; Twelfth Informational Supplement* (ISBN 1-56238-454 -6), **2002**, M100-S12 (M7).