European Journal of Chemistry

View Journal Online
View Article Online

Synthesis and characterization of new 4-aryl-2-(2-oxopropoxy)-6-(2,5-dichlorothiophene)nicotinonitrile and their furo[2,3-*b*]pyridine derivatives: Assessment of antioxidant and biological activity

Mahmoud Al-Refai [1,*], Mohammad Ibrahim [1], Abdullah Al-Fawwaz [2] and Armin Geyer [3]

[1] *Department of Chemistry, Faculty of Science, Al Al-Bayt University, Al-Mafraq, 25113, Jordan*
mahmoud_alrefai@aabu.edu.jo (M.R.), mohammadibrahim@aabu.edu.jo (M.I.)
[2] *Department of Biological Sciences, Faculty of Science, Al Al-Bayt University, Al-Mafraq, 25113, Jordan*
al_fawwaz@aabu.edu.jo (A.F.)
[3] *Faculty of Chemistry, Philipps University Marburg, Hans-Meerwein-Straße 4, 35032 Marburg, Germany*
geyer@staff.uni-marburg.de (A.G.)

* *Corresponding author at: Department of Chemistry, Faculty of Science, Al Al-Bayt University, Al-Mafraq, 25113, Jordan.*
Tel: +962.2.6297000/2141 Fax: +962.2.6297021 e-mail: mahmoud_alrefai@aabu.edu.jo (M. Al-Refai).

RESEARCH ARTICLE

10.5155/eurjchem.9.4.375-381.1792

Received: 27 September 2018
Received in revised form: 18 October 2018
Accepted: 20 October 2018
Published online: 31 December 2018
Printed: 31 December 2018

KEYWORDS

Pyridine
Thiophene
Antioxidant
Nicotinonitrile
Cyanopyridone
Furo[2,3-b]pyridine

ABSTRACT

A new series of furo[2,3-*b*]pyridine derivatives bearing aryl substituents were synthesized in two steps, where, the cyano-(2*H*)-pyridones (1a-l) were converted to the corresponding nicotinonitriles (2a-l), followed by the Thorpe-Ziegler ring cyclization to the furo[2,3-*b*]pyridine derivatives (3a-l). All new compounds were characterized by 1D-NMR experiments (^{1}H and ^{13}C) and 2D-NMR experiments (COSY, HMBC and HSQC), as well as ESI-MS and HR-ESI-MS data. The new compounds were screened for their antioxidant activities by 2,2-diphenyl-1-picryl-hydrazylhydrate (DPPH) free radical assay. The highest radical scavenging effect was observed for nicotinonitriles 2d, 2h and 2l and furo[2,3-*b*]pyridines 3b, 3f and 3j by methanolic solvent at 4.0 mg/mL concentration. Remarkably, all nicotinonitriles and furo[2,3-*b*]pyridine exhibited a significant radical scavenging activity after 24 and 48 hours compared with 0.5 hour.

Cite this: *Eur. J. Chem.* **2018**, *9(4)*, 375-381 **Journal website**: www.eurjchem.com

1. Introduction

Furo[2,3-*b*]pyridine containing heterocycles are of interest because they show a wide range of pharmacological activities [1-6]. The furo[2,3-*b*]pyridine scaffold present in known many naturally occurring and biologically active compounds [7,8]. The synthesis of pyridine containing derivatives with antimicrobial and antioxidant activities with no toxic effects on health is of growing concern to combat the resistance of pathogenic microorganisms to one or several problems of antibiotics [9-13].

The highly reactive chemical free radicals might form in different ways in the body and have the potential to damage cells, organelles and DNA, causing cancer, and cardiovascular and neurodegenerative diseases [14,15]. Therefore, numerous methods are used to test antioxidant activities of chemical compounds. Two free radicals that are commonly used to assess antioxidant activity in vitro are 2,2-diphenyl-1-picryl hydrazyl (DPPH) and 2,2'-azino-*bis*(3-ethylbenzothiazoline-6-sulphonic acid) (ABTS) [16]. Many investigators have studied the free radical scavenging activity based on a fixed endpoint (e.g. 30 min) [17].

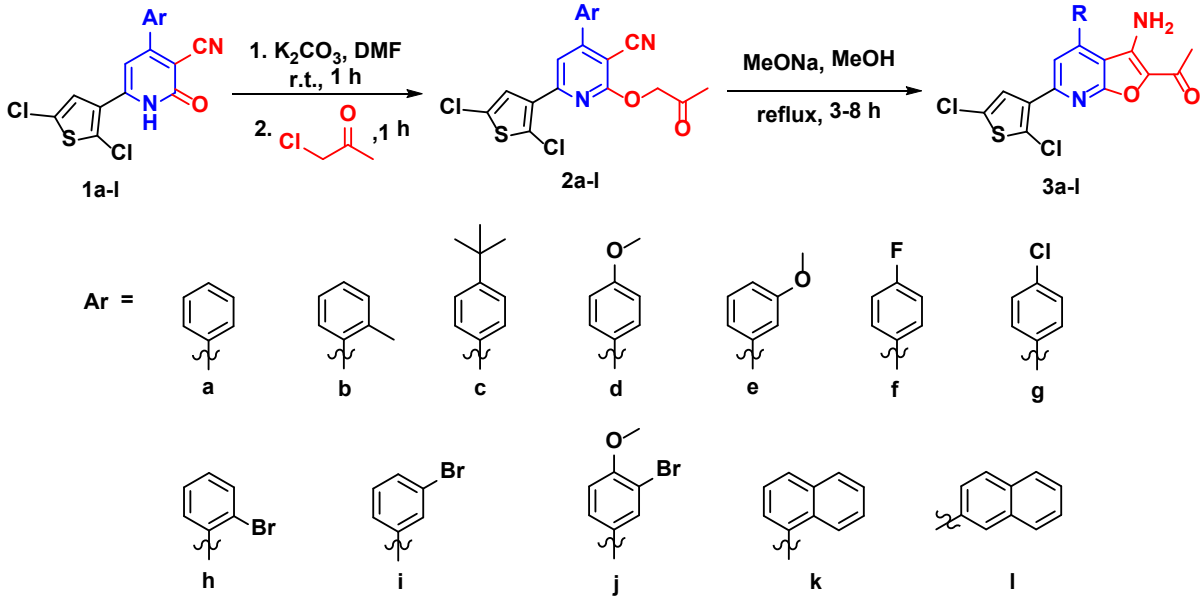
Very recently, the crystal structure and fluorescence behavior of new furo[2,3-*b*]pyridine compound and a series of a nicotinonitrile and furo[2,3-*b*]pyridine derivatives bearing alkyl moieties have been investigated by our research group [18,19]. Herein, we report the synthesis and characterization of a new series of nicotinonitriles and furo[2,3-*b*]pyridines as potent antioxidant agents (Scheme 1).

2. Experimental

2.1. Materials

4-Aryl-3-cyano-(2*H*)-pyridones (**1a-l**) were prepared according to literature procedure [20,21] and used without further purification. Solvents were dried and distilled according to standard methods.

ISSN 2153-2249 (Print) / ISSN 2153-2257 (Online)
http://dx.doi.org/10.5155/eurjchem.9.4.375-381.1792



Scheme 1. Synthesis of nicotinonitriles (**2a-l**) and furo[2,3-*b*]pyridines (**3a-l**).

2.2. Instrumentation

All purchased reagents were used as received. Solvents were dried and distilled according to standard protocols. NMR spectra, ^{1}H (300 and 500 MHz) and ^{13}C (75 and 125 MHz), were recorded in $CDCl_3$ (used as an internal standard) at 300 K on Bruker spectrometers (Bruker, Germany). Chemical shifts, δ (ppm), were determined from the centre of the respective coupling pattern (s: singlet, d: doublet, dd: doublet of doublet, t: triplet). ESI-MS and HR-ESI-MS measurements were carried out on a LTQ-FT mass spectrometer (Thermo Fisher Scientific). The reactions were monitored using thin layer chromatography (TLC) using analytical TLC plates coated with silica gel ($60F_{254}$, Merck). Preparative thin layer chromatography (PTLC) was performed at room temperature using $CHCl_3$ as the mobile phase.

2.3. Synthesis

2.3.1. General procedure for the synthesis of nicotinonitriles (2a-l)

The 4-aryl-3-cyano-(2*H*)-pyridone (**1**) (2.0 mmol), and K_2CO_3 (2.2 mmol, 1.1 equiv.) were dissolved in DMF (50 mL, dry) in a round-bottom flask equipped with a stir bar. The reaction mixture was stirred at the ambient temperature for 1 h. Chloroacetone (4.0 mmol, 2.0 equiv.) was added portion wise to the mixture and allowed to stir overnight. The reaction mixture was transferred to ice water. The precipitate formed was filtered off, washed with water, dried and then subjected to PTLC chromatography using chloroform as mobile phase to give the corresponding nicotinonitrile (**2**) (Scheme 1).

2-(2-Oxopropoxy)-6-(2, 5-dichlorothiophen-3-yl)-4-phenyl pyridine-3-carbonitrile (**2a**): Color: White solid. Yield: 97%. ^{1}H NMR (300 MHz, $CDCl_3$, δ, ppm): 2.33 (s, 3H, CH_3COCH_2O), 5.08 (s, 2H, CH_3COCH_2O), 7.21 (s, 1H, H-4'), 7.57 (m, 3H, H-3", 4", 5"), 7.58 (m, 2H, H-6", 2"), 7.66 (s, 1H, H-5). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.4 (CH_3COCH_2O), 71.2 (CH_3COCH_2O), 93.8 (C-3), 114.7 (CN-3), 116.8 (CH-5), 127.1 (CH-4'), 128.4 (CH-6", 2"), 129.2 (CH-3", 5"), 130.4 (CH-4"), 135.1 (C_q-, 3'), 135.7 (C_q-1"), 151.5 (C_q-6), 157.1 (C_q-4), 163.2 (C_q-2), 203.0 (CO). MS (+ESI, *m/z* (%)): 403 ($[M+H]^+$, 100), 405 ($[M+H+2]^+$, 66), 407 ($[M+H+4]^+$, 15), 425 ($[M+Na]^+$, 84), 427 ($[M+Na+2]^+$, 55), 429 ($[M+Na+4]^+$, 13), 827 ($[2M+Na]^+$, 15), 829 ($[2M+Na+2]^+$, 22), 830 ($[2M+Na+4]^+$, 14). HRMS (+ESI, *m/z*): 403.0070 $[M+H]^+$, 405.0040 $[M+H+2]^+$, 407.0012 $[M+H+4]^+$, (calcd. for $C_{19}H_{13}Cl_2N_2O_2S$, 403.0069). HRMS (+ESI, *m/z*): 424.9889 $[M+Na]^+$, 426.9860 $[M+Na+2]^+$, 428.9831 $[M+Na+4]^+$, (calcd. for $C_{19}H_{12}Cl_2N_2O_2SNa$, 424.9889).

(2-Oxopropoxy)-6-(2, 5-dichlorothiophen-3-yl)-4-o-tolylpyridine-3-carbonitrile (**2b**): Color: White solid. Yield: 93%. ^{1}H NMR (300 MHz, $CDCl_3$, δ, ppm): 2.29 (s, 3H, CH_3-2"), 2.31 (s, 3H, CH_3COCH_2O), 5.06 (s, 2H, CH_3COCH_2O), 7.19 (s, 1H, H-4'), 7.33 (m, 4H, H-3", 4", 5", 6"), 7.50 (s, 1H, H-5). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 19.9 (CH_3-2"), 26.3 (CH_3COCH_2O), 71.2 (CH_3COCH_2O), 95.7 (C_q-3), 114.0 (CN-3), 117.5 (CH-5), 126.2, 127.1, 128.7, 129.8, 131.0 (CH-4', 3", 4", 5", 6"), 134.0, 135.1, 135.2, 135.6 (C_q-2', 3', 5', 1'', 2"), 151.3 (C_q-6), 158.3 (C_q-4), 162.7 (C_q-2), 202.8 (CO). MS (+ESI, *m/z* (%)): 417 ($[M+H]^+$, 34), 419 ($[M+H+2]^+$, 24), 421 ($[M+H+24]^+$, 8), 439 ($[M+Na]^+$, 100), 441 ($[M+Na+2]^+$, 66), 443 ($[M+Na+4]^+$, 15), 855 ($[2M+Na]^+$, 18), 857 ($[2M+Na+2]^+$, 30), 859 ($[2M+Na+4]^+$, 17). HRMS (+ESI, *m/z*): 439.0047 $[M+Na]^+$, 441.0017 $[2M+Na+4]^+$, (calcd. for $C_{20}H_{14}Cl_2N_2O_2SNa$, 439.0045).

2-(2-Oxopropoxy)-4-(4-tert-butylphenyl)-6-(2, 5-dichloro thiophen-3-yl)pyridine-3-carbonitrile (**2c**): Color: White solid. Yield: 89%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 1.39 (s, 9H, $C(CH_3)_3$), 2.32 (s, 3H, $COCH_3$), 5.02 (s, 2H, CH_3COCH_2O), 7.20 (s, 1H, H-4'), 7.58 (d, *J* = 8.7 Hz, 2H, H-3", 5"), 7.63 (d, *J* = 8.7, 5.4 Hz, 2H, H-2", 6"), 7.66 (s, 1H, H-5). ^{13}C NMR (125 MHz, $CDCl_3$, δ, ppm): 26.4 ($COCH_3$), 31.2 ($C(CH_3)_3$), 34.9 ($C(CH_3)_3$), 71.2 (CH_3COCH_2O), 93.5 (C_q-3), 115.0 (CN), 116.7 (CH-5), 126.2 (CH-3", 5"), 126.5, 127.0 (C_q-2', 5'), 128.2 (CH-4', 2", 6"), 132.7 (C_q-1''), 135.1 (C_q-3'), 151.3 (C_q-6), 153.9 (C_q-4''), 157.0 (C_q-4), 162.6 (C_q-2), 203.1 (CO). MS (+ESI, *m/z* (%)): 460 ($[M+H]^+$, 24), 462 ($[M+H+2]^+$, 18), 482 ($[M+Na]^+$, 63), 484 ($[M+Na+2]^+$, 41), 941 ($[2M+Na]^+$, 100), 943 ($[2M+Na+2]^+$, 43). HRMS (+ESI, *m/z*): 481.0517 $[M+Na]^+$, 483.0488 $[M+Na+2]^+$, (calcd. for $C_{23}H_{20}Cl_2N_2O_2SNa$, 481.0515).

2-(2-Oxopropoxy)-6-(2, 5-dichlorothiophen-3-yl)-4-(4-methoxyphenyl)pyridine-3-carbonitrile (**2d**): Color: White solid. Yield: 93%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.22 (s, 3H, CH_3COCH_2O), 3.82 (s, 3H, OCH_3-4"), 4.97 (s, 2H, CH_3COCH_2O), 6.99 (d, *J* = 8.8 Hz, 2H, H-3", 5"), 7.11 (s, 1H, H-4'), 7.54 (s, 1H, H-5), 7.57 (d, *J* = 8.8 Hz, 2H, H-2", 6"). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.5 (CH_3COCH_2O), 55.5 (OCH_3-4"), 71.2 (CH_3CO

CH_2O), 93.2 (C-3), 114.6 (CH-3", 5"), 115.1 (CN-3), 116.5 (CH-5), 127.1 (CH-4'), 130.0 (CH-2", 6"), 127.0, 127.9, 135.2, (C_q-2', 3', 5', 1"), 151.2 (C_q-6), 156.7 (C_q-4), 161.5 (C_q-4"), 163.4 (C_q-2), 203.1 (CO). MS (+ESI, *m/z* (%)): 455 ([M+Na]$^+$, 100), 457 ([M+Na+2]$^+$, 66), 459 ([M+Na+4]$^+$, 15), 887 ([2M+Na]$^+$, 21), 889 ([2M+Na+2]$^+$, 30), 891 ([2M+Na+4]$^+$, 20). HRMS (+ESI, *m/z*): 454.9994 [M+Na]$^+$, 456.9965 [M+Na+2]$^+$, (calcd. for $C_{20}H_{14}Cl_2N_2O_3SNa$, 454.9994).

2-(2-Oxopropoxy)-6-(2, 5-dichlorothiophen-3-yl)-4-(3-methoxyphenyl)pyridine-3-carbonitrile (**2e**): Color: White solid. Yield: 95%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.33 (s, 3H, C*H*$_3$COCH$_2$O), 3.92 (s, 3H, OCH$_3$-3"), 5.09 (s, 2H, CH$_3$COC*H*$_2$O), 7.10 (ddd, *J* = 8.3, 2.4, 0.7 Hz, 1H, H-4"), 7.21 (t, *J* = 2.3 Hz, 1H, H-2"), 7.22 (s, 1H, H-4'), 7.25 (ddd, *J* = 7.6, 1.8, 1.1 Hz, 1H, H-6"), 7.49 (t, *J* = 7.9 Hz, 1H, H-5"), 7.66 (s, 1H, H-5). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.4 (*C*H$_3$COCH$_2$O), 55.5 (OCH$_3$-3"), 71.2 (CH$_3$CO*C*H$_2$O), 93.8 (C_q-3), 113.9 (CH-2"), 114.6 (CN-3), 116.1 (CH-5), 116.7 (CH-4"), 120.7 (CH-6"), 127.0 (CH-4'), 130.3 (CH-5"), 135.1 (C_q-1"), 136.9 (C_q-3'), 151.4 (C_q-6), 157.0 (C_q-4), 159.9 (C_q-3"), 163.2 (C_q-2), 202.9 (CO). MS (+ESI, *m/z* (%)): 455 ([M+Na]$^+$, 100), 457 ([M+Na+2]$^+$, 65), 459 ([M+Na+24]$^+$, 15), 887 ([2M+Na]$^+$, 20), 889 ([2M+Na+2]$^+$, 33), 891 ([2M+Na+4]$^+$, 21). HRMS (+ESI, *m/z*): 454.9996 [M+Na]$^+$, 456.9967 [M+Na+2]$^+$, (calcd. for $C_{20}H_{14}Cl_2N_2O_3SNa$, 454.9994).

2-(2-Oxopropoxy)-6-(2, 5-dichlorothiophen-3-yl)-4-(4-fluorophenyl)pyridine-3-carbonitrile (**2f**): Color: White solid. Yield: 82%. ^{1}H NMR (500 MHz, DMSO-d_6, δ, ppm): 2.18 (s, 3H, COC*H*$_3$), 5.30 (s, 2H, CH$_3$COC*H*$_2$O), 7.46 (t, *J* = 8.9 Hz, 2H, H-3", 5"), 7.71 (s, 1H, H-5), 7.78 (s, 1H, H-4'), 7.84 (dd, *J* = 8.9, 5.35 Hz, 2H, H-2", 6"). ^{13}C NMR (125 MHz, DMSO-d_6, δ, ppm): 26.4 (CO*C*H$_3$), 71.5 (CH$_3$CO*C*H$_2$O), 115.3 (CN), 93.2 (C_q-3), 116.6, 116.4 (CH-3", 5"), 117.1 (CH-5), 127.0, 126.2 (C_q-2', 5'), 128.6 (CH-4'), 131.7 (CH-2", 6"), 132.2 (C_q-1''), 135.1(C_q-3'), 151.5 (C_q-6), 155.9 (C_q-4), 162.8, 163.3 (C_q-4''), 164.8 (C_q-2), 202.8 (CO). MS (+ESI, *m/z* (%)): 421 ([M+H]$^+$, 68), 423 ([M+H+2]$^+$, 41), 443 ([M+Na]$^+$, 100), 445 ([M+Na+2]$^+$, 67), 863 ([2M+Na]$^+$, 30), 865 ([2M+Na+2]$^+$, 46), 867 ([M+Na+4]$^+$, 24). HRMS (+ESI, *m/z*): 442.9801 [M+Na]$^+$, 444.9771 [M+Na+2]$^+$, (calcd. for $C_{19}H_{11}Cl_2FN_2O_2SNa$, 442.9795).

2-(2-Oxopropoxy)-4-(4-chlorophenyl)-6-(2, 5-dichlorothiophen-3-yl)pyridine-3-carbonitrile (**2g**): Color: White solid. Yield: 86%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.32 (s, 3H, C*H*$_3$COCH$_2$O), 5.08 (s, 2H, CH$_3$COC*H*$_2$O), 7.21 (s, 1H, H-4'), 7.56 (d, *J* = 8.7 Hz, 2H, H-3", 5"), 7.62 (d, *J* = 8.7 Hz, 2H, H-2", 6"), 7.61 (s, 1H, H-5). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.3 (*C*H$_3$COCH$_2$O), 71.2 (CH$_3$CO*C*H$_2$O), 93.7 (C-3), 114.4 (CN-3), 116.4 (CH-5), 127.0 (CH-4'), 129.5 (CH-3", 5"), 129.7 (CH-2", 6"), 126.7, 127.2, 134.0, 135.0 (C_q-2', 5', 1'', 4"), 136.9 (C_q-3'), 151.7 (C_q-6), 155.8 (C_q-4), 163.3 (C_q-2), 202.5 (CO). MS (+ESI, *m/z* (%)): 437 ([M+H]$^+$, 23), 439 ([M+H+2]$^+$, 21), 441 ([M+H+24]$^+$, 8), 459 ([M+Na]$^+$, 97), 461 ([M+Na+2]$^+$, 100), 463 ([M+Na+4]$^+$, 36), 895 ([2M+Na]$^+$, 21), 897 ([2M+Na+2]$^+$, 39), 899 ([2M+Na+4]$^+$, 34). HRMS (+ESI, *m/z*): 458.9501 [M+Na]$^+$, 460.9471 [M+Na+2]$^+$, 462.9442 [M+Na+4]$^+$, (calcd. for $C_{19}H_{11}Cl_3N_2O_2SNa$, 458.9499).

2-(2-Oxopropoxy)-4-(2-bromophenyl)-6-(2, 5-dichlorothiophen-3-yl)pyridine-3-carbonitrile (**2h**): Color: White solid. Yield: 79%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.34 (s, 3H, C*H*$_3$COCH$_2$O), 5.08 (s, 2H, CH$_3$COC*H*$_2$O), 7.22 (s, 1H, H-4'), 7.40 (m, 2H, H-3", 4"), 7.50 (td, *J* = 7.9, 1.1 Hz, 1H, H-5"), 7.59 (s, 1H, H-5), 7.77 (d, *J* = 8.2 Hz, 1H, H-6''). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.4 (*C*H$_3$COCH$_2$O), 71.2 (CH$_3$CO*C*H$_2$O), 95.6 (C_q-3), 113.8 (CN-3), 117.7 (CH-5), 121.7 (C_q-2"), 127.0 (CH-4'), 127.9, 130.4, 131.3, 133.6 (CH-3", 4", 5", 6"), 134.9 (C_q-1"), 136.6 (C_q-3'), 151.3 (C_q-6), 156.5 (C_q-4), 162.5 (C_q-2), 202.9 (CO). MS (+ESI, *m/z* (%)): 481 ([M+H]$^+$, 18), 483 ([M+H+2]$^+$, 27), 485 ([M+H+24]$^+$, 13), 503 ([M+Na]$^+$, 68), 505 ([M+Na+2]$^+$, 100), 507 ([M+Na+4]$^+$, 52). HRMS (+ESI, *m/z*): 502.8994 [M+Na]$^+$, 504.8970 [M+Na+2]$^+$, 506.8943 [M+Na+4]$^+$, (calcd. for $C_{19}H_{11}BrCl_2N_2O_2SNa$, 502.8994).

2-(2-Oxopropoxy)-4-(3-bromophenyl)-6-(2, 5-dichlorothiophen-3-yl)pyridine-3-carbonitrile (**2i**): Color: White solid. Yield: 81%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.32 (s, 3H, C*H*$_3$COCH$_2$O), 5.09 (s, 2H, CH$_3$COC*H*$_2$O), 7.20 (s, 1H, H-4'), 7.45 (t, *J* = 7.9 Hz, 1H, H-5"), 7.60 (1H, s, H-5), 7.63 (ddd, *J* = 7.8, 1.7, 1.0 Hz, 1H, H-6"), 7.70 (ddd, *J* = 8.04, 1.89, 1.0 Hz, 1H, H-4"), 7.77 (d, *J* = 1.8 Hz, 1H, H-2"). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.4 (*C*H$_3$COCH$_2$O), 71.2 (CH$_3$CO*C*H$_2$O), 93.8 (C_q-3), 114.3 (CN-3), 116.5 (CH-5), 123.2 (C_q-3"), 126.9 (CH-6"), 127.0 (CH-4'), 130.7 (CH-5"), 131.3 (CH-2"), 133.4 (CH-4"), 134.9 (C_q-, 3'), 137.6 (C_q-1"), 151.8 (C_q-6), 155.4 (C_q-4), 163.2 (C_q-2), 202.6 (CO). MS (+ESI, *m/z* (%)): 503 ([M+Na]$^+$, 67), 505 ([M+Na+2]$^+$, 100), 507 ([M+Na+4]$^+$, 46). HRMS (+ESI, *m/z*): 502.8994 [M+Na]$^+$, 504.8970 [M+Na+2]$^+$, 506.8943 [M+Na+4]$^+$, (calcd. for $C_{19}H_{11}BrCl_2N_2O_2SNa$, 502.8994).

2-(2-Oxopropoxy)-4-(3-bromo-4-methoxyphenyl)-6-(2, 5-dichlorothiophen-3-yl)pyridine-3-carbonitrile (**2j**): Color: White solid. Yield: 82%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.22 (s, 3H, C*H*$_3$COCH$_2$O), 3.91 (s, 3H, OCH$_3$-4"), 4.98 (s, 2H, CH$_3$CO C*H*$_2$O), 6.99 (d, *J* = 8.9 Hz, 1H, H-5"), 7.11 (s, 1H, H-4'), 7.50 (s, 1H, H-5), 7.60 (dd, *J* = 8.6, 2.3 Hz, 1H, H-6"), 7.74 (d, *J* = 2.3 Hz, 1H, H-2"). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.3 (*C*H$_3$CO CH$_2$O), 56.5 (OCH$_3$-4"), 71.2 (CH$_3$CO*C*H$_2$O), 93.4 (C_q-3), 112.0 (CH-5"), 112.5 (C_q-3"), 114.7 (CN-3), 116.3 (CH-5), 127.0 (CH-4'), 129.0 (CH-6"), 129.1 (C_q-1"), 133.2 (CH-2"), 137.2 (C_q-3'), 151.5 (C_q-6), 155.2 (C_q-4), 157.6 (C_q-4"), 163.3 (C_q-2), 202.7 (CO). MS (+ESI, *m/z* (%)): 533 ([M+Na]$^+$, 67), 535 ([M+Na+2]$^+$, 100), 537 ([M+Na+24]$^+$, 47). HRMS (+ESI, *m/z*): 532.9100 [M+Na]$^+$, 534.9075 [M+Na+2]$^+$, 536.9048 [M+Na+4]$^+$, (calcd. for $C_{20}H_{13}BrCl_2N_2O_3SNa$, 532.9100).

2-(2-Oxopropoxy)-6-(2, 5-dichlorothiophen-3-yl)-4-(naphthalen-1-yl)pyridine-3-carbonitrile (**2k**): Color: White solid. Yield: 84%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.33 (s, 3H, C*H*$_3$COCH$_2$O), 5.11 (s, 2H, CH$_3$COC*H*$_2$O), 7.14 (s, 1H, H-4'), 7.55-7.98 (m, 8H, H-5, 2", 3", 4", 6", 7", 8", 9"). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.4 (*C*H$_3$COCH$_2$O), 71.2 (CH$_3$CO*C*H$_2$O), 96.2 (C-3), 114.1 (CN-3), 118.5 (C-5), 126.4, 127.1, 128.1, 133.4, 133.8, 135.0 (C_q-2', 3', 5', 1", 5'', 10''), 124.6, 125.2, 126.6, 130.4, 127.1, 127.2, 127.3, 128.8 (CH-4', 2'', 3'', 4'', 6'', 7'', 8'', 9''''), 151.0 (C_q-6), 156.8 (C_q-4), 163.0 (C_q-2), 202.8 (CO). MS (+ESI, *m/z* (%)): 453 ([M+H]$^+$, 20), 455 ([M+H+2]$^+$, 13), 475([M+Na]$^+$, 100), 477 ([M+Na+2]$^+$, 64), 479 ([M+Na+24]$^+$, 14), 927 ([2M+Na]$^+$, 32), 929 ([2M+Na+2]$^+$, 45), 931 ([2M+Na+4]$^+$, 26). HRMS (+ESI, *m/z*): 475.0047 [M+Na]$^+$, 477.0017 [M+Na+2]$^+$, (calcd. for $C_{23}H_{14}Cl_2N_2O_2SNa$, 475.0045).

2-(2-Oxopropoxy)-6-(2, 5-dichlorothiophen-3-yl)-4-(naphthalen-6-yl)pyridine-3-carbonitrile (**2l**): Color: White solid. Yield: 76%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.25 (s, 3H, C*H*$_3$COCH$_2$O), 5.01 (s, 2H, CH$_3$COC*H*$_2$O), 7.15 (s, 1H, H-4'), 7.49-7.96 (m, 7H, H- 5, 4", 5", 6", 7", 9", 10"), 8.09 (d, *J* =1.5 Hz, 1H, H-2"). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.4 (*C*H$_3$COCH$_2$O), 71.2 (CH$_3$CO*C*H$_2$O), 93.9 (C-3), 114.8 (CN-3), 117.0 (C-5), 123.9, 125.2, 133.0, 133.9, 135.1(C_q-2', 3', 5', 1", 3'', 8''), 127.0, 127.1, 127.7, 127.8, 128.6, 128.8, 129.1(CH-4', 2'', 4'', 5'', 6'', 7'', 9'', 10''), 151.4 (C_q-6), 157.1 (C_q-4), 163.3 (C_q-2), 202.9 (CO). MS (+ESI, *m/z* (%)): 453 ([M+H]$^+$, 18), 455 ([M+H+2]$^+$, 11), 475 ([M+Na]$^+$, 100), 477 ([M+Na+2]$^+$, 72), 479 ([M+Na+24]$^+$, 14), 927 ([2M+Na]$^+$, 26), 929 ([2M+Na+2]$^+$, 40), 931 ([2M+Na+4]$^+$, 25). HRMS (+ESI, *m/z*): 475.0047 [M+Na]$^+$, 477.0017 [M+Na+2]$^+$, (calcd. for $C_{23}H_{14}Cl_2N_2O_2SNa$, 475.0045).

2.3.2. General procedure for the synthesis of furo[2, 3-b] pyridines (3a-l)

The nicotinonitrile (**2**) (1.0 mmol), was dissolved in methanolic solution of a sodium methoxide, the resulting solution was heated under reflux for 3 h. After being cooled to

room temperature, the resulting solid was filtered off, washed with cold methanol and dried. The product was subjected to PTLC chromatography using the chloroform as mobile phase to afford the corresponding furo[2, 3-*b*]pyridine compound **3** (Scheme 1).

1-(3-Amino-6-(2, 5-dichlorothiophen-3-yl)-4-phenylfuro[2, 3-b]pyridin-2-yl)ethanone (**3a**): Color: Yellow solid. Yield: 79%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.59 (s, 3H, CH_3CO CH_2O), 5.21 (bs, 2H, NH_2), 7.52 (s, 1H, H-4'), 7.62 (m, 5H, H-2", 3", 4", 5", 6"), 7.81 (s, 1H, H-5). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.2 (*C*H_3CO), 109.7 (C_q-3a), 118.9 (CH-5), 127.9 (CH-4'), 128.4 (CH-2", 6"), 125.0, 126.8, 137.6 (C_q-3, 2', 5',), 129.1 (CH-3", 5"), 129.8 (CH-4"), 133.9 (C_q-2), 136.2 (C_q-1"), 136.5 (C_q-3'), 148.2 (C_q-4), 150.9 (C_q-6), 159.9 (C_q-7a), 190.1 (CO). MS (+ESI, *m/z* (%)): 403 ([M+H]$^+$, 100), 405 ([M+H+2]$^+$, 68), 407 ([M+H+4]$^+$, 58), 425 ([M+Na]$^+$, 23), 427 ([M+Na+2]$^+$, 16), 429 ([M+Na+4]$^+$, 11). HRMS (+ESI, *m/z*): 403.0071 [M+H]$^+$, 405.0041 [M+H+2]$^+$, 407.0021 [M+H+4]$^+$, (calcd. for $C_{19}H_{13}Cl_2N_2O_2S$, 403.0069).

1-(3-Amino-6-(2, 5-dichlorothiophen-3-yl)-4-o-tolylfuro[2, 3-b]pyridin-2-yl)ethanone (**3b**): Color: Yellow solid. Yield: 81%. ^{1}H NMR (300 MHz, $CDCl_3$, δ, ppm): 2.55 (s, 6H, CH_3CO, CH_3-2"), 5.20 (bs, 2H, NH_2), 7.43 (d, *J* = 7.1 Hz, 1H, H-3''), 7.50 (m, 3H, H-4', 4'', 5''), 7.75 (s, 1H, H-5), 7.78 (d, *J* = 7.8 Hz, 1H, H-6''). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.2 (*C*H_3CO, *C*H_3-2"), 110.6 (C_q-3a), 119.1 (CH-5), 128.0, 130.3, 131.1, 133.6 (CH-4', 3'', 4'', 5'', 6"), 122.2, 125.2, 126.7, 134.1, 136.5, 136.4 (C_q-2, 3, , 2', 5', 1'', 2''), 137.0 (C_q-3'), 146.1 (C_q-4), 150.8 (C_q-6), 159.5 (C_q-7a), 190.2 (CO).

1-(4-(4-Tert-butylphenyl)-3-amino-6-(2,5-dichlorothiophen -3-yl)furo[2, 3-b]pyridin-2-yl)ethanone (**3c**): Color: Yellow solid. Yield: 86%. ^{1}H NMR (500 MHz, DMSO-d_6, δ, ppm): 1.37 (s, 9H, C(CH_3)$_3$), 2.46 (s, 3H, COCH_3), 5.87 (bs, 2H, NH_2), 7.65 (s, 4H, H-2", 3", 5", 6"), 7.74 (s, 1H, H-4'), 7.75 (s, 1H, H-5). ^{13}C NMR (125 MHz, DMSO-d_6, δ, ppm): 26.7 (CO*C*H_3), 31.5 (C (*C*H_3)$_3$), 35.1 (*C*(CH$_3$)$_3$), 109.8 (C_q-3a), 119.6 (CH-5), 125.1, 125.9 (C_q-2', 5'), 126.7 (CH-3", 5"), 128.8 (CH-4', 2", 6"), 133.2 (C_q-1''), 133.4 (C_q-2), 136.7 (C_q-3'), 148.7 (C_q4), 150.4 (C_q-6), 152.8 (C-4''), 159.8 (C_q-7a), 188.8 (CO). MS (+ESI, *m/z* (%)): 464 ([M+H+4]$^+$, 100), 941 ([2M+Na]$^+$, 16).

1-(3-Amino-6-(2, 5-dichlorothiophen-3-yl)-4-(4-methoxy phenyl)furo[2, 3-b]pyridin-2-yl)ethanone (**3d**): Color: Yellow solid. Yield: 88%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.55 (s, 3H, CH_3CO), 3.91 (s, 3H, OCH_3-4"), 5.59 (bs, 2H, NH_2), 7.10 (d, *J* = 8.6Hz, 2H, H-3", 5"), 7.48 (s, 1H, H-4'), 7.54 (d, *J* = 8.6 Hz, 2H, H-2", 6"), 7.74 (s, 1H, H-5). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.2 (*C*H_3CO), 55.5 (OCH_3-4"), 109.7 (C_q-3a), 114.8 (CH-3", 5"), 118.8 (CH-5), 128.0 (CH-4'), 129.8 (CH-2", 6"), 124.9, 126.7, 128.3, 133.8, 136.6 (C_q-2, 3, 2', 5', 1''), 137.6 (C_q-3'), 148.0 C_q-4), 150.8 (C_q-6), 160.0 (C_q-4"), 160.9 (C_q-7a), 190.1 (CO). MS (+ESI, *m/z* (%)): 455 ([M+Na]$^+$, 100), 457 ([M+Na+2]$^+$, 67), 459 ([M+Na+24]$^+$, 23), 887 ([2M+Na]$^+$, 46), 889 ([2M+Na+2]$^+$, 48), 891 ([2M+Na+4]$^+$, 37). HRMS (+ESI, *m/z*): 454.9995 [M+Na]$^+$, 456.9966 [M+Na+2]$^+$, 458.9941 [M+Na+4]$^+$, (calcd. for $C_{20}H_{14}Cl_2N_2O_3SNa$, 454.9994).

1-(3-Amino-6-(2, 5-dichlorothiophen-3-yl)-4-(3-methoxy phenyl)furo[2, 3-b]pyridin-2-yl)ethanone (**3e**): Color: Yellow solid. Yield: 90%. ^{1}H NMR (300 MHz, $CDCl_3$, δ, ppm): 2.55 (s, 3H, CH_3CO), 3.89 (s, 3H, OCH_3-3"), 5.60 (bs, 2H, NH_2), 7.08 (d, *J* = 7.6 Hz, 2H, H-2", 4''), 7.09 (s, 1H, H-4'), 7.15 (d, *J* = 7.6, 1.8, 1.1 Hz, 1H, H-6"), 7.49 (t, *J* = 8.47 Hz, 1H, H-5"), 7.87 (s, 1H, H-5). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.2 (*C*H_3CO), 55.5 (OCH_3-3"), 109.7 (C_q-3a), 114.0 (CH-2"), 115.3 (CH-4"), 118.8 (CH-5), 120.5 (CH-6"), 127.9 (CH-4'), 130.5 (CH-5"), 126.8, 125.0, 136.5, 137.4 (C_q-2, 3, 2', 5', 1''), 137.5 (C_q-3'), 147.7 (C_q-4), 150.8 (C_q-6), 159.5 (C_q-3"), 160.2 (C_q-7a), 190.1 (CO). MS (+ESI, *m/z* (%)): 433 ([M+H]$^+$, 51), 435 ([M+H+2]$^+$, 34), 437 ([M+H+4]$^+$, 11), 455 ([M+Na]$^+$, 100), 457 ([M+Na+2]$^+$, 67), 459 ([M+Na+24]$^+$, 15), 887 ([2M+Na]$^+$, 37), 889 ([2M+Na+2]$^+$, 48), 891 ([2M+Na+4]$^+$, 30). HRMS (+ESI, *m/z*): 454.9996 [M+Na]$^+$, 456.9966 [M+Na+2]$^+$, (calcd. for $C_{20}H_{14}Cl_2N_2O_3SNa$, 454.9994).

1-(3-Amino-6-(2, 5-dichlorothiophen-3-yl)-4-(4-fluoro phenyl)furo[2, 3-b]pyridin-2-yl)ethanone (**3f**): Color: Yellow solid. Yield: 71%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.45 (s, 3H, COCH_3), 5.91 (bs , 2H, NH_2), 7.46 (t, *J* = 8.9 Hz, 2H, H-3", 5"), 7.74 (s, 2H, H-4', 5), 7.75 (dd, *J* = 8.9, 5.4 Hz, 2H, H-2", 6"). ^{13}C NMR (125 MHz, DMSO-d_6, δ, ppm): 26.6 (CO*C*H_3), 110.0 (C_q-3a), 116.7, 116.1 (CH-3", 5"), 119.7 (CH-5), 125.2, 126.0, 133.5, (C_q-2, 2', 5'), 128.9 (CH-4'), 131.5 (CH-2", 6"), 136.8 (C_q-1''), 137.9 (C_q-3'), 147.73 (C_q-4), 150.5 (C_q-6), 159.7 (C_q-7a), 162.5, 162.9, (C_q-4''), 189.0 (CO). MS (+ESI, *m/z* (%)): 421 ([M+H]$^+$, 100), 423 ([M+H+2]$^+$, 70), 425 ([M+H+4]$^+$, 25), 443 ([M+Na]$^+$, 82), 445 ([M+Na+2]$^+$, 58), 863 ([2M+Na]$^+$, 30), 865 ([2M+Na+2]$^+$, 50), 867 ([M+Na+4]$^+$, 30). HRMS (+ESI, *m/z*): 442.9802 [M+Na]$^+$, 444.9773 [M+Na+2]$^+$, (calcd. for $C_{19}H_{11}Cl_2FN_2O_2SNa$, 442.9795).

1-(3-Amino-4-(4-chlorophenyl)-6-(2, 5-dichlorothiophen-3-yl)furo[2, 3-b]pyridin-2-yl)ethanone (**3g**): Color: Yellow solid. Yield: 75%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.59 (s, 3H, CH_3CO), 5.47 (bs, 2H, NH_2), 7.52 (s, 1H, H-4'), 7.57 (d, *J* = 8.7 Hz, 2H, H-3", 5"), 7.60 (d, *J* = 8.7Hz, 2H, H-2", 6"), 7.78 (s, 1H, H-5). ^{13}C NMR (125 MHz, $CDCl_3$, δ, ppm): 26.3 (*C*H_3CO), 109.7 (C_q-3a), 118.8 (CH-5), 129.7 (CH-3", 5"), 127.9 (CH-4'), 129.8 (CH-2", 6"), 134.1, 134.6, 134.6, 136.3, 136.4 (C_q-2, 3, 2', 5', 1'', 4''), 137.0 (C_q-3'), 146.7 (C_q-4), 152.8 (C_q-6), 159.9 (C_q-7a), 190.3 (CO). MS (+ESI, *m/z* (%)): 437 ([M+H]$^+$, 33), 439 ([M+H+2]$^+$, 36), 441 ([M+H+24]$^+$, 82).

1-(3-Amino-4-(2-bromophenyl)-6-(2, 5-dichlorothiophen-3-yl)furo[2, 3-b]pyridin-2-yl)ethanone (**3h**): Color: Yellow solid. Yield: 73%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.56 (s, 3H, CH_3CO), 5.21 (bs, 2H, NH_2), 7.45 (m, 2H, H-3", 4"), 7.53 (m, 2H, H-4', 5"), 7.77 (s, 1H, H-5), 7.82 (dd, *J* = 8.8 Hz, 1H, H-6''). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.3 (*C*H_3CO), 110.6 (C_q-3a), 119.2 (CH-5), 122.2 (C_q-2"), 128.0, 130.4, 131.1, 133.6 (CH-4', 3", 4", 5", 6"), 126.8, 134.1, 136.4, 136.5, (C_q-2, 3, 2', 5', 1"), 137.0 (C_q-3'), 146.1 (C_q-4), 150.8 (C_q-6), 159.5 (C_q-7a), 190.2 (CO). MS (+ESI, *m/z* (%)): 481 ([M+H]$^+$, 44), 483 ([M+H+2]$^+$, 65), 485 ([M+H+24]$^+$, 36), 503 ([M+Na]$^+$, 66), 505 ([M+Na+2]$^+$, 100), 507 ([M+Na+4]$^+$, 52). HRMS (+ESI, *m/z*): 502.8995 [M+Na]$^+$, 504.8971 [M+Na+2]$^+$, 506.8943 [M+Na+4]$^+$, (calcd. for $C_{19}H_{11}BrCl_2N_2O_2SNa$, 502.8994).

1-(3-Amino-4-(3-bromophenyl)-6-(2, 5-dichlorothiophen-3-yl)furo[2, 3-b]pyridin-2-yl)ethanone (**3i**): Color: Yellow solid. Yield: 68%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.59 (s, 3H, CH_3CO), 5.50 (bs, 2H, NH_2), 7.49 (t, *J* = 7.8 Hz, 1H, H-5"), 7.51 (s, 1H, H-4'), 7.56 (ddd, *J* = 7.6, 1.6, 1.2 Hz, 1H, H-6"), 7.73 (ddd, *J* = 8.0, 1.9, 1.1 Hz, 1H, H-4"), 7.77 (t, *J* = 1.7 Hz, 1H, H-2"), 7.78 (s, 1H, H-5). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.3 (*C*H_3CO), 109.7 (C_q-3a), 118.8 (CH-5), 123.5 (C_q-3"), 127.0 (CH-6"), 127.9 (CH-4'), 125.2, 126.9, 137.0 (C_q-3, 2', 5',), 130.8 (CH-5"), 131.4 (CH-2"), 132.8 (CH-4"), 137.0 (C_q-3'), 138.1 (C_q-1''), 146.3 (C_q-4), 151.0 (C_q-6), 159.8 (C_q-7a), 190.3 (CO). MS (+ESI, *m/z* (%)): 481 ([M+H]$^+$, 36), 483 ([M+H+2]$^+$, 58), 503 ([M+Na]$^+$, 60), 505 ([M+Na+2]$^+$, 100), 507 ([M+Na+4]$^+$, 66), 985 ([2M+Na]$^+$, 47), 987 ([2M+Na+2]$^+$, 63), 989 ([2M+Na+4]$^+$, 53).

1-(3-Amino-4-(3-bromo-4-methoxyphenyl)-6-(2, 5-dichloro thiophen-3-yl)furo[2, 3-b]pyridin-2-yl)ethanone (**3j**): Color: Yellow solid. Yield: 66%. ^{1}H NMR (300 MHz, $CDCl_3$, δ, ppm): 2.22 (s, 3H, CH_3CO), 3.92 (s, 3H, OCH_3-4"), 4.98 (bs, 2H, NH_2), 6.99 (d, *J* = 8.6 Hz, 1H, H-5"), 7.11 (s, 1H, H-4'), 7.50 (s, 1H, H-5), 7.60 (dd, *J* = 8.6, 2.3 Hz, 1H, H-6"), 7.74 (d, *J* = 2.28 Hz, 1H, H-2"). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.3 (*C*H_3CO), 56.5 (OCH_3-4"), 109.6 (C_q-3a), 112.3 (CH-5"), 112.8 (C_q-3"), 118.8 (CH-5), 127.9 (CH-4'), 128.7 (CH-6"), 133.3 (CH-2"), 126.8, 129.6, 134.0, 136.4 (C_q-2, 3, 2', 5', 1''), 137.2 (C_q-3'), 146.3 (C_q-4), 150.9 (C_q-6), 157.2 (C_q-4"), 159.9 (C_q-7a), 190.2 (CO). MS (+ESI, *m/z* (%)): 533 ([M+Na]$^+$, 70), 535 ([M+Na+2]$^+$, 100),

537 ([M+Na+24]$^+$, 65). HRMS (+ESI, *m/z*): 532.9101 [M+Na]$^+$, 534.9077 [M+Na+2]$^+$, 536.9050 [M+Na+4]$^+$, 538.9026 [M+Na+6]$^+$, (calcd. for $C_{20}H_{13}BrCl_2N_2O_3SNa$, 532.9100).

1-(3-Amino-6-(2, 5-dichlorothiophen-3-yl)-4-(naphthalen-1-yl)furo[2, 3-b]pyridin-2-yl)ethanone (**3k**): Color: Yellow solid. Yield: 63%. ^{1}H NMR (500 MHz, $CDCl_3$, δ, ppm): 2.55 (s, 3H, C*H*$_3$CO), 5.03 (s, 2H, NH_2), 7.54 (s, 1H, H-4'), 7.59-8.02 (m, 8H, H-5, 2", 3", 4", 6", 7", 8", 9"). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.2 (*C*H$_3$CO), 111.4 (C_q-3a), 120.0 (CH-5), 125.1, 125.3, 126.9, 127.0, 127.5, 128.0, 128.7, 130.0 (CH-4', 2'', 3'', 4'', 6'', 7'', 8'', 9'''), 126.8, 126.8, 130.7, 133.2, 133.7, 134.0, 136.5, 137.3 (C_q-2, 3, 2', 3', 5', 1'', 3'', 8''), 146.3 (C_q-4), 150.8 (C_q-6), 159.7 (C_q-7a), 189.7 (CO). MS (+ESI, *m/z* (%)): 453 ([M+H]$^+$, 62), 455 ([M+H+2]$^+$, 44), 457 ([M+H+4]$^+$, 18), 475 ([M+Na]$^+$, 100), 477 ([M+Na+2]$^+$, 77), 479 ([M+Na+24]$^+$, 18), 927 ([2M+Na]$^+$, 44), 929 ([2M+Na+2]$^+$, 55), 931 ([2M+Na+4]$^+$, 41). HRMS (+ESI, *m/z*): 475.0048 [M+Na]$^+$, 477.0018 [M+Na+2]$^+$, (calcd. for $C_{23}H_{14}Cl_2N_2O_2SNa$, 475.0045).

1-(3-Amino-6-(2, 5-dichlorothiophen-3-yl)-4-(naphthalen-3-yl)furo[2, 3-b]pyridin-2-yl)ethanone (**3l**): Color: Yellow solid. Yield: 67%. ^{1}H NMR (300 MHz, $CDCl_3$, δ, ppm): 2.57 (s, 3H, C*H*$_3$CO), 5.55 (bs, 2H, NH_2), 7.51 (s, 1H, H-4'), 7.65-8.01 (m, 8H, H-5, 2", 4", 5", 6", 7", 9", 10"). ^{13}C NMR (75 MHz, $CDCl_3$, δ, ppm): 26.3 (*C*H$_3$CO), 109.7 (C-3a), 119.1 (C-5), 125.5, 127.4, 127.5, 127.9, 128.0, 128.1, 128.4, 129.4 (CH-4', 2'', 4'', 5'', 6'', 7'', 9'', 10''), 125.1, 126.8, 133.1, 133.4, 133.5, 133.9, 136.5, 137.4 (C_q-2, 3, 2', 3', 5', 1'', 3'', 8''), 148.2 (C_q-4), 150.9 (C_q-6), 159.9 (C_q-7a), 190.1 (CO).

2.4. Biological screening test

2.4.1. Antimicrobial activity

Antimicrobial activity of the new nicotinonitriles (**2a-l**) and furo[2,3-*b*]pyridines (**3a-l**) was determined by the wells diffusion method, for these assays, cultures of the following microorganisms were used: two Gram-positive bacteria (*Staphylococcus aureus* and *Bacillus subtilis*), one Gram-negative bacteria (*Escherichia coli*), and fungal strains (*Aspergillus niger* and *Penicillium sp.*) [13].

2.4.2. Antioxidant activity by DPPH method

The antioxidant activity of methanolic solutions of compounds **2a-l** and **3a-l** was determined using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging assay determined by a modified method described by Ayoola *et al.* [22]. Solutions of concentration 4.0 mg/mL were prepared for each solution of compounds **2a-l** and **3a-l** dissolved in methanol. Freshly prepared methanolic solution of DPPH (1.0 mL, 0.25 μM) was mixed with 2.0 mL of different samples of each solution of compounds **2a-l** and **3a-l**. The mixture was shaken by vortex and the absorbance was determined after 0.5, 24 and 48 hours by spectrophotometer at 517 nm. Ascorbic acid of same concentration (4.0) mg/mL was used as standard reference. Lower absorbance of the reaction mixture indicates higher free radical scavenging activity. The scavenging activity of the samples was calculated using the following equation:

$$\text{Percentage of radical scavenging activity } (AA\%) = \frac{(AB - AS)}{AB} \times 100 \quad (1)$$

3. Results and discussion

3.1. Synthesis and characterization of nicotinonitriles and furo[2,3-b]pyridines

The nicotinonitrile derivatives **2a-l** are readily obtained in very good to excellent yields by treatment of the 4-aryl-3-cyano-2(*H*)-pyridones (**1a-l**) with potassium carbonate and chloroacetone in DMF as a solvent at room temperature, Scheme 1. The synthesized derivatives and different substituents are listed in Scheme 1. The reflux of nicotinenitriles (**2a-l**) in methanolic solution of sodium methoxide leads to the target furo[2,3-*b*]pyridines (**3a-l**) in good to high yields (Scheme 1).

The structures of the new synthesized heterocycles were proposed using standard spectroscopic techniques including IR, 1D-NMR, 2D-NMR, and mass spectrometry. The exact chemical shifts of all protons and carbons were determined by a full analysis of 2D-NMR spectra. The complete data for all protons and carbons are listed in the experimental section. The ESI-MS and HR-ESI-MS data analyses revealed the correct molecular ion peaks for all compounds as suggested by their molecular formulas.

The ^{1}H NMR data of nicotinonitriles (**2a-l**) showed two aromatic resonances at δ 7.50-7.78, 7.11-7.77 ppm attributed to H-5 and H-4', respectively, as well as two resonance signals in the up field region due to the oxygenated methylene protons –CH_2– at δ 4.97-5.11 ppm while the methyl singlet of an acetate moiety in the region δ 2.18-2.34 ppm. Furo[2,3-*b*]pyridines (**3a-l**) showed similar ^{1}H NMR signals to nicotinenitriles (**2a-l**) except for the presence of an exchangeable protons at δ 4.98-5.91 ppm (NH_2) instead of the oxygenated methylene. In the ^{13}C NMR spectra, the –CH_2– and CN carbons in the newly synthesized nicotinonitriles (**2a-l**) appeared at δ 71.2-71.5 ppm and δ 113.8-115.3 ppm, respectively. The absence of the –CH_2– and CN carbon signals in these region confirms the formation of the furo[2,3-*b*]pyridine compounds **3a-l**. The ESI/MS and HR-ESI/MS mass spectra confirmed the proposed molecular formulas of compounds **2a-l** and **3a-l**.

3.2. Biological screening

3.2.1. Antimicrobial activity

The nicotinonitriles (**2a-l**) and furo[2,3-*b*]pyridine compounds **3a-l** had no effect on any of bacterial nor fungal strains used in antimicrobial activity test.

3.2.2. Antioxidant activity

An antioxidant is molecule that inhibit or quench free radical reactions and delay or inhibit the cellular damage [11,23,24]. A broader definition of antioxidant was suggested by Halliwell *et al.* 1995 as "any substance that when present at low concentrations, compared to those of an oxidizable substrate significantly delays or prevents oxidation of that substrate" [25]. Several compounds with the antioxidant activity have been used to slow down the radical associated oxidative reactions [11].

In the present study radical scavenging activity were evaluated to clarify the antioxidant properties of the nicotinenitriles (**2a-l**) and furo[2,3-*b*]pyridine (**3a-l**) at various time intervals (0.5, 24 and 48 hours). The results of radical scavenging activity usually were determined based on a fixed endpoint [17]. Some investigators have proposed kinetic parameters that can provide more complete information about antioxidant behavior [26,27] and suggested that the kinetics could be more important than the total antioxidant capacities determined at a fixed point [11,28]. The DPPH reagent reacted very slowly with chemical samples, approaching, but not reaching, steady state after 8 h [16]. Some researchers reported similar slow reaction of most antioxidants which were tested when DPPH were used [16,29]. The scavenging percentage of ascorbic acid was higher than all other compounds.

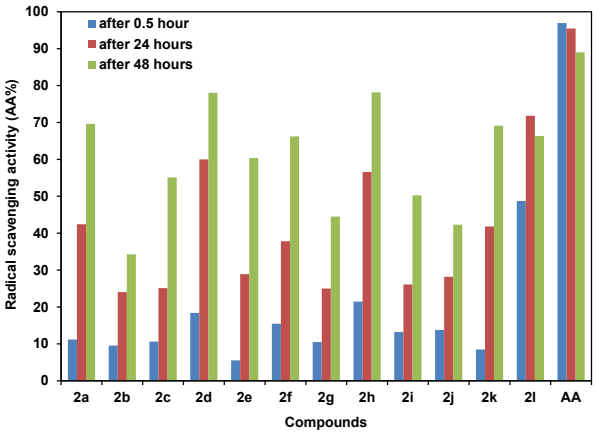


Figure 1. The DPPH radical scavenging activity of nicotinonitriles (**2a-l**) in methanol at 4.0 mg/mL concentration after 0.5, 24 and 48 hours.

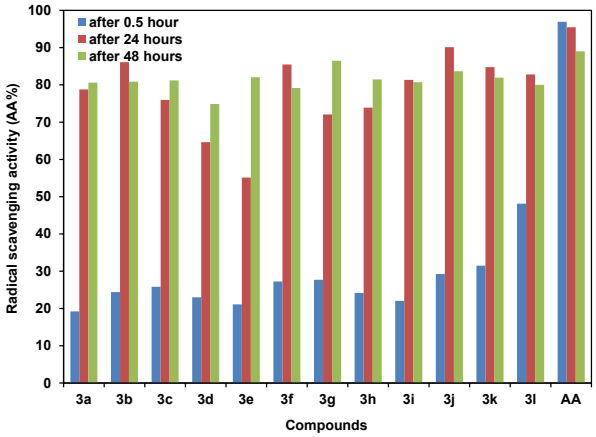


Figure 2. The DPPH radical scavenging activity of furo[2,3-*b*]pyridine (**3a-l**) in methanol at 4.0 mg/mL concentration after 0.5, 24 and 48 hours.

Antioxidant activities of nicotinonitriles (**2a-l**) in methanol were measured using DPPH free radical scavenging assay monitored at various time intervals (0.5, 24 and 48 hours), and the results of DPPH scavenging effect are shown in Figure 1. Nicotinonitriles (**2a-l**) exhibited a significant antioxidant activity after 24 and 48 hours. The highest percentage (%) was shown by methanolic solvent of compounds **2d**, **2h** and **2l** at 4.0 mg/mL concentration.

The DPPH radical scavenging activity of furo[2,3-*b*]pyridine (**3a-l**) in methanol at 4.0 mg/mL concentration, monitored at various time intervals (0.5, 24 and 48 hours) are shown in Figure 2. Furo[2,3-*b*]pyridine (**3a-l**) exhibited a significant radical scavenging activity after 24 and 48 hours compared with 0.5 hour, all chemical compounds exhibited high radical scavenging activity after 24 and 48 hours compared with 0.5 hour. The highest radical scavenging activity percentage (%) was shown by compounds **3b**, **3f** and **3j** after 24 hours with 86.1, 85.5 and 90.1%, respectively.

4. Conclusion

New nicotinonitrile and furo[2,3-*b*]pyridine derivatives bearing aromatic substituents have been synthesized and fully characterized. The new heterocyclic compounds were studied for their antioxidant activity. Notably, both nicotinonitriles and furo[2,3-*b*]pyridine derivatives possess antioxidant activity after 24 and 48 hours compared with 0.5 hour.

Acknowledgements

We are grateful to Al Al-Bayt University (Mafraq, Jordan), and the Deutsche Forschungemeinschaft (DFG, Germany) for financial support.

Disclosure statement

Conflict of interests: The authors declare that they have no conflict of interest.
Author contributions: All authors contributed equally to this work.
Ethical approval: All ethical guidelines have been adhered.
Sample availability: Samples of the compounds are available from the author.

ORCID

Mahmoud Al-Refai
http://orcid.org/0000-0001-7154-7888
Mohammad Ibrahim
http://orcid.org/0000-0003-0720-9889
Abdullah Al-Fawwaz
http://orcid.org/0000-0003-0795-7537
Armin Geyer
http://orcid.org/0000-0001-8096-2266

References

[1]. Kumar, N. R.; Poornachandra, Y.; Nagender, P.; Mallareddy, G.; Kumar, R. N.; Ranjithreddy, P.; Kumar, G. C.; Narsaiah, B. *Eur. J. Med. Chem.* **2016**, *108*, 68-78.
[2]. Kawakami, K.; Takahashi, H.; Ohki, H.; Kimura, K.; Miyauchi, S.; Miyauchi, R.; Takemura, M. *Chem. Pharm. Bull.* **2000**, *48*, 1667-1672.
[3]. Ledoussal, B.; Bouzard, D.; Coroneos, E. *J. Med. Chem.* **1992**, *35*,198-200.
[4]. Dorsey, B. D.; McDonough, C.; McDaniel, S. L.; Levin, R. B.; Newton, C. L.; Hoffman, J. M.; Darke, P. L.; Zugay-Murphy, J. A.; Emini, E. A.; Schleif, W. A.; Olsen, D. B.; Stahlhut, M. W.; Rutkowski, C. A.; Kuo, L. C.; Lin, J. H.; Chen, I. W.; Michelson, S. R.;Holloway, M. K.; Huff, J. R.; Vacca, J. P. *J. Med. Chem.* **2000**, *43(18)*, 3386-3399.
[5]. Bathula, C.; Mamidala, R.; Thulluri, C.; Agarwal, R.; Jha, K. K.; Munshi, P.; Adepally, U.; Singh, M. T.; Chary, M. T.; Sen, S. *RSC Adv.* **2015**, *5*, 90374-90385.
[6]. Fokialakis, N.; Magiatis, P.; Aligiannis, N.; Mitaku, S.; Tillequin, F.; Sevenet, T. *Phytochemistry* **2001**, *57*, 593-596.
[7]. Sirakanyan, S. N.; Hovakimyan, A. A.; Noravyan, A. S. *Russ. Chem. Rev.* **2015**, *84(4)*, 441-454.
[8]. Eren, G.; Unlu, S.; Nunez, M. T.; Labeaga, L.; Ledo, F.; Entrena, A.; Banoglu, E.; Costantino, G.; Sahin, M. F. *Bioorg. Med. Chem.* **2010**, *18*, 6367-6376.
[9]. Lucera, A.; Costa, C.; Conte, A.; Del Nobile, M. A. *Front Microbiol.* **2012**, *3(287)*, 1-13.
[10]. Hintz, T.; Matthews, K. K.; Di, R. *Biomed. Res. Int.* **2015**, *2015*, 1-12.
[11]. Nimse, S. B.; Pal, D.; Mazumder, A.; Mazumder, R. *J. Chem.* **2015**, *2015*, 1-5.
[12]. Avsar, C.; Ozler, H.; Berber, I.; Civek, S. *Int. Food Res. J.* **2016**, *23(4)*, 1711-1716.
[13]. Venkatesh, T.; Bodke, Y. D.; Kenchappa, R.; Telkar, S. *Med. Chem. (Los Angeles)* **2016**, *6*, 440-448.
[14]. Valko, M.; Leibfritz, D.; Moncol, J.; Cronin, M. T. D.; Mazur, M.; Telser, J. *Int. J. Biochem. Cell Biol.* **2007**, *39*, 44-84.
[15]. Dildar, A.; Muhammad, M.; Ramsha, S. *Antioxidants* **2015**, *4*, 394-409.
[16]. Shalaby, E.; Sh. Sanaa, M. *Indian J. Mar. Sci.* **2013**, *42(5)*, 556-564.
[17]. Terpinc, P.; Bezjak, M.; Abramovic, H. *Food Chem.* **2009**, *115(2)*, 740-744.
[18]. Ibrahim, M. M.; Al-Refai, M.; Al-Fawwaz, A.; Ali, B. F.; Geyer, A.; Harms, K.; Marsch, M.; Krugerd, M.; Osmane, H.; Azmi, M. N. *J. Fluoresc.* **2018**, *28(2)*, 655-662.
[19]. Ibrahim, M. M.; Al-Refai, M.; Azmi, M. N.; Osman, H.; Abu Bakar , M. H.; Geyer, A. *J. Iran. Chem. Soc.* **2018**, https://doi.org/10.1007/s13738-018-1549-y
[20]. Al-Refai, M. *Asian J. Chem.* **2015**, *27*, 725-728.
[21]. Sahauneh, Z., M.Sc. thesis, Synthesis of some new 6-(2, 5-dichloro thiene-3-yl)-4-aromatic-3-carbonitrile-2-oxo-1,2-dihydropyridine derivatives, Al Al-Bayt University, Jordan, 2011.
[22]. Ayoola, G. A.; Folawewo, A. D.; Adesegun, S. A.; Abioro, O. O.; Adepoju-Bello, A. A.; Coker, H. A. B. *Afr. J. Plant Sci.* **2008**, *2(10)*, 124-128.
[23]. Young, I. S.; Woodside, J. V. *J. Clin. Pathol.* **2001**, *54(3)*, 176-186.
[24]. Pal, D. K.; Nimse, S. B. *Asian J. Chem.* **2006**, *18(4)*, 3004-3008.
[25]. Halliwell, B. Murica, M. A.; Chirico, S.; Aruoma, O. L. *Crit. Rev. Food Sci. Nutr.* **1995**, *35(1-2)*, 7-20.
[26]. Perez-Jimenez, J.; Saura-Calixto, F. *Int. J. Food Sci. Technol.* **2008**, *43(1)*, 185-191.
[27]. Sanchez-Moreno, C.; Larrauri, J. A.; Saura-Calixto, F. *J. Sci. Food Agric.* **1998**, *76(2)*, 270-276.
[28]. Goupy, P.; Dufour, C.; Loonis, M.; Dangles, O. *J. Agric. Food Chem.* **2003**, *51*, 615-622.
[29]. Bondet, V.; Brand-Williams, W.; Berset, C. *Lebensm. Wiss. Technol.* **1997**, *30*, 609-615.