Indian Journal of Chemistry
Vol. 57B, January 2018, pp. 98-107

Synthesis and biological activities of nicotinaldehyde based azlactones

Ch Dayakar[a], L Mounika[a], K Rajkumar[a], A Zehra[b], T R Murthy[c],
Shasi V Kalivendi[c], A K Tiwari[b] & B China Raju*[a]

[a] Natural Products Chemistry Division, [b] Medicinal Chemistry & Biotechnology Division and
[c] Chemical Biology Division, CSIR-Indian Institute of Chemical Technology,
Hyderabad 500 007, India

E-mail: chinaraju@iict.res.in

Received 8 June 2017; accepted (revised) 16 November 2017

A series of nicotinaldehyde based azlactones **3a-g**, **6a-f**, **11a-d**, **16b-c**, and **16e-f** have been prepared and screened for their free radicals scavenging, α-glucosidase inhibitory and anti-proliferative activities on cell lines, namely lung adenocarcinoma (A549), human breast cancer (MCF7) and human epithelial cervical cancer (HeLa). Compound **3g** is the most potent α-glucosidase inhibitor followed by compounds **6b** and **6a**. Compound **11b** is the better DPPH and $ABTS^{+}$ radical scavenger. Compounds **11c-d** and **16f** show anti-proliferative activity on all the tested cell lines. However, compounds **16c** and **16e** display anti-proliferative activity on MCF7 and HeLa cell lines.

Keywords: Nicotinaldehydes, azlactones, DPPH, $ABTS^{+}$ radical scavenging activity, α-glucosidase inhibition, anti-proliferative activity

Neonicotinoids are used for controlling the insects due to their broad spectrum insecticidal with low mammalian toxicity[1,2]. Imidacloprid was the first chloronicotinyl non-natural insecticide[3]. It has a novel mode of action by binding to the nicotinergic acetylcholine receptor in the postsynaptic region of the insect nerve. Chloronicotinaldehydes are important heterocyclic compounds and have potential applications in the areas of pharmaceuticals and agrochemicals[4-6]. These are the precursors for annulations of heterocyclic ring system for the preparation of arachidonic acid metabolites[7,8].

Azlactones are an important class of heterocyclic compounds and exhibit various biological activities[9]. Azlactones are used as intermediates in the preparation of amino acids, peptides and photosensitive devices for proteins. However, there is no report available on 2-chloronicotinaldehyde based azlactones and their biological activities.

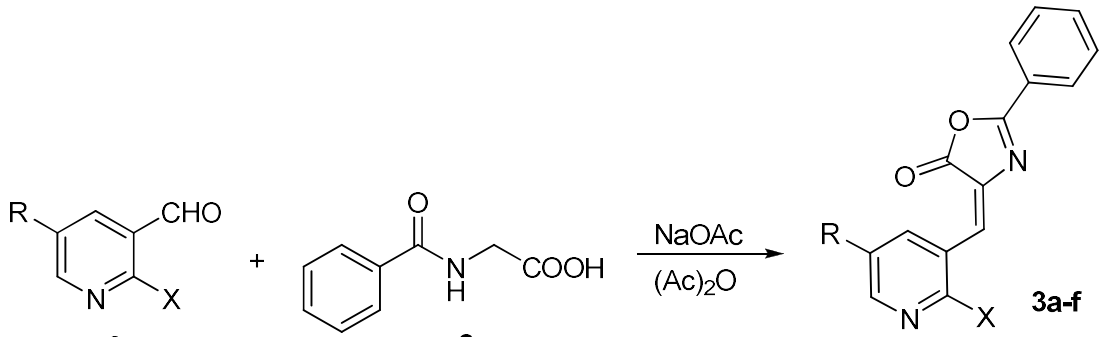
As part of our ongoing research on 2-chloronicotinaldehydes, we prepared biologically important 2-chloronicotinaldehyde based heterocyclic compounds[10-16]. Recently, we have reported pyridinyl-1*H*-1,2,3-triazoles, triazolylisoxazoles, triazolyldihydroisoxazoles and triazolylbenzohydrazides from 2-chloronicotinaldehydes[17,18]. The present manuscript describes the preparation of nicotinaldehyde based azlactones and their biological activities.

Results and Discussion

The preparation of azlactones **3a-g**, **6a-f**, **11a-d**, **16b-c** and **16e-f** are presented in Schemes I-V. The Scheme I represents preparation of 3-pyridylazlactone **3a** and 2-chloro-5-substituted pyridylazlactones **3b-f**. The 2-chloro-5-substituted carboxaldehydes **1c-f** are prepared as per our earlier reported method[19]. Condensation of nicotinaldehydes **1a-f** with hippuric acid **2** in the presence of sodium acetate in acetic anhydride provided the corresponding azlactones **3a-f**.

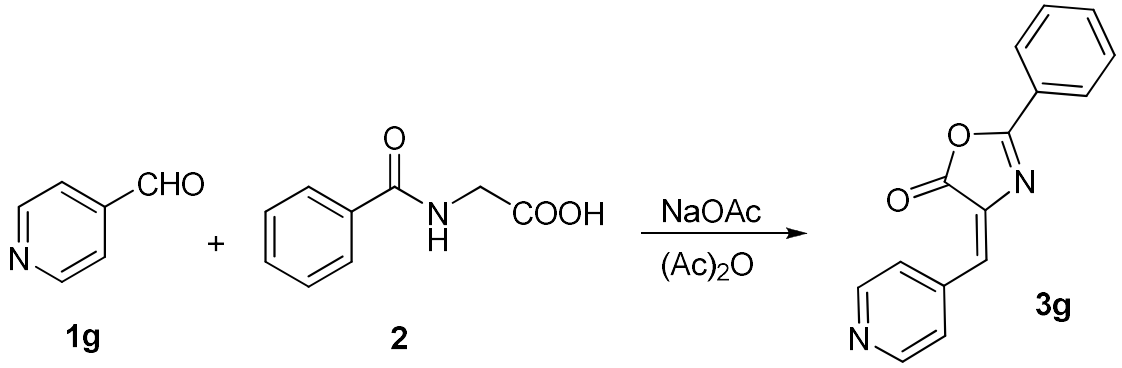
Scheme II describes the preparation of 4-pyridylazlactone **3g** by the condensation of 4-pyridinecarboxaldehyde with hippuric acid **2** in the presence of sodium acetate in acetic anhydride.

Scheme III illustrates the preparation of 2-morpholine/thio-morpholine pyridylazlactones **6a-f**. 2-Chloro-5-substituted carboxaldehydes **1c-e** were reacted with morpholine **4a** and thio-morpholine **4b** in the presence of K_2CO_3 in dry DMF at 110°C to furnish the corresponding compounds **5a-f**. Condensation of **5a-f** with hippuric acid **2** in the presence of sodium acetate in acetic anhydride provided the corresponding azlactones **6a-f**.

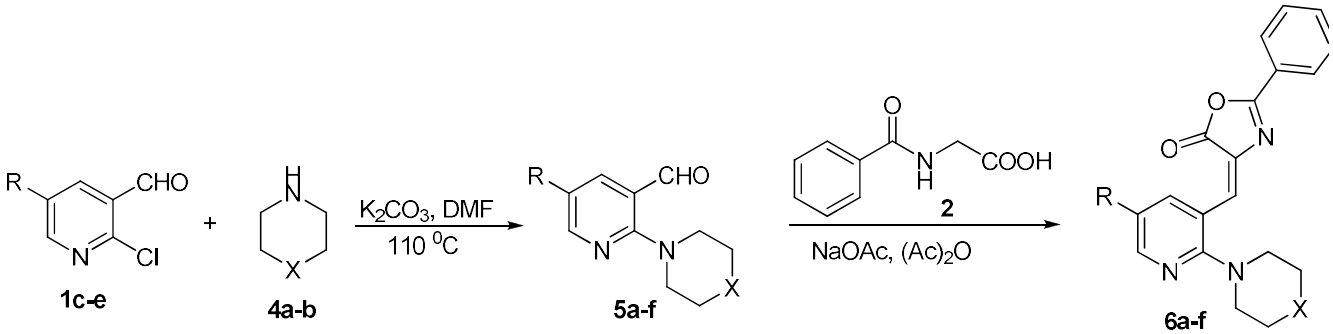


3a R = H, X = H
3b R = H, X = Cl
3c R = CH_3, X = Cl
3d R = Ph, X = Cl
3e R = *p*-Me-Ph, X = Cl
3f R = *p*-F-Ph, X = Cl

Scheme I



Scheme II

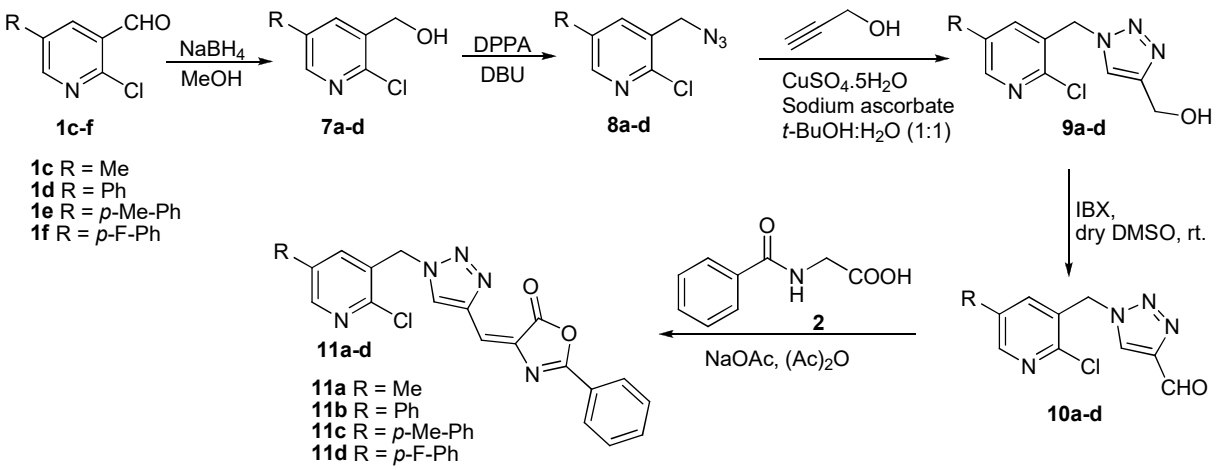


1c R = CH_3
1d R = Ph
1e R = *p*-Me-Ph

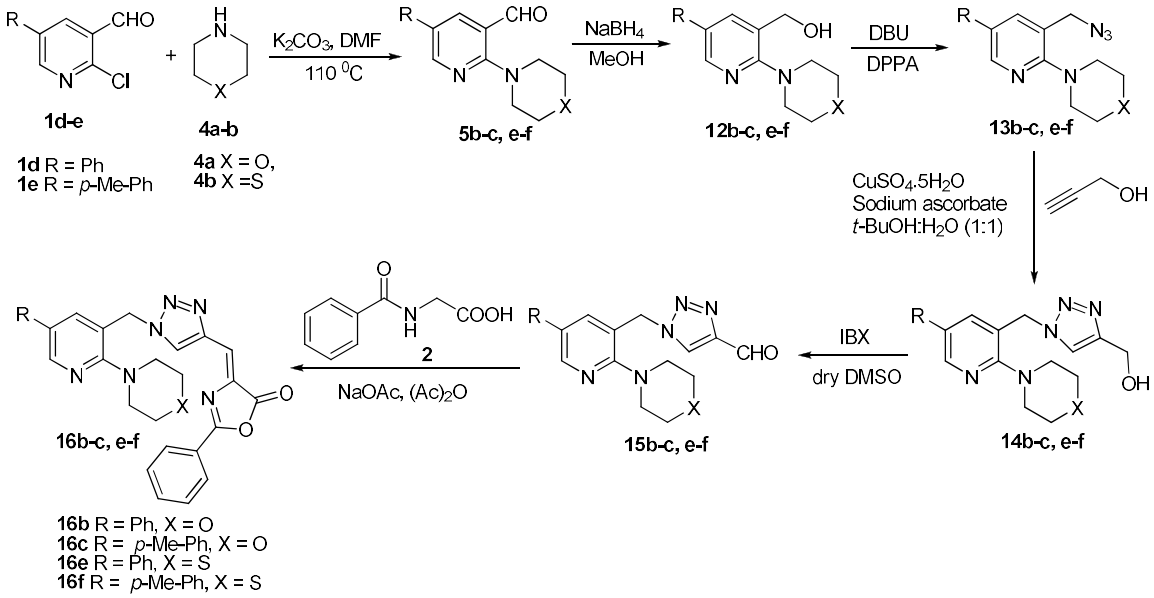
4a X = O,
4b X = S

6a R = CH_3, X = O
6b R = Ph, X = O
6c R = *p*-Me-Ph, X = O
6d R = CH_3, X = S
6e R = Ph, X = S
6f R = *p*-Me-Ph, X = S

Scheme III



Scheme IV



Scheme V

Scheme IV describes the preparation of pyridyl triazolylazlactones **11a-d**. Reduction of 2-chloronicotinaldehydes **1c-f** with $NaBH_4$ in methanol at 0°C furnished the corresponding alcohols **7a-d**. Azidation of alcohols **7a-d** with diphenylphosphoryl azide (DPPA) in the presence of DBU at RT afforded the corresponding azides[20] **8a-d**. Cycloaddition of azides **8a-d** with propargyl alcohol in the presence of $CuSO_4$/sodium ascorbate at RT provided the corresponding triazolylalcohols[21,22] **9a-d**. Oxidation of alcohols **9a-d** under IBX conditions furnished the 1*H*-1,2,3-triazole-4-carboxaldehydes[18] **10a-d**. Condensation of **10a-d** with hippuric acid **2** in the presence of sodium acetate in acetic anhydride provided the corresponding azlactones **11a-d**.

Scheme V illustrates the preparation of 2-morpholine/thio-morpholinepyridyl- triazolylazlactones. 2-Chloro-5-substituted carboxaldehydes **1d-e** were reacted with morpholine **4a** and thio-morpholine **4b** in the presence of K_2CO_3 in dry DMF at 110°C and furnished the corresponding compounds **5b-c** and **5e-f**. Sequential reactions such as reduction, azidation and click reaction of **5b-c** and **5e-f** provided the corresponding triazolylalcohols **14b-c** and **14e-f**. Oxidation of alcohols **14b-c**, **14e-f** under IBX conditions furnished the corresponding pyridinyl-1*H*-1,2,3-triazole-4-carbaldehydes **15b-c** and **15e-f**. Condensation of **15b-c**, **15e-f** with hippuric acid **2** in the presence of sodium acetate in acetic anhydride provided the corresponding azlactones **16b-c** and

16e-f. Thus synthesized azlactones **3a-g**, **6a-f**, **11a-d**, **16b-c** and **16e-f** are new compounds and well characterised by IR, ^{1}H ^{13}C NMR and mass spectroscopy.

Azlactones **3a-g**, **6a-f**, **11a-d**, **16b-c** and **16e-f** were evaluated for their DPPH, $ABTS^{+\cdot}$ free radical scavenging and α-glucosidase inhibitory activities[23]. These compounds were also tested for their anti-proliferative activity on three cancer cell lines namely lung adenocarcinoma (A549), human breast cancer (MCF7) and human epithelial cervical cancer (HeLa) by MTT assay and compared with the standard drug doxorubicin[18].

DPPH and $ABTS^{+\cdot}$ free radical scavenging activities of azlactones

The DPPH and $ABTS^{+\cdot}$ radical scavenging activities of azlactones **3a-g**, **6a-f**, **11a-d**, **16b-c** and **16e-f** are presented in Table I. Pyridylazlactones **3a-g** and 2-morpholine/thio-morpholine pyridylazlactones **6a-f** could not scavenge DPPH and $ABTS^{+\cdot}$radicals. The pyridyltriazolyl- azlactone **11b** displayed the DPPH (SC_{50}, 46.70 μg/mL) and $ABTS^{+\cdot}$ ($SC_{50,}$ 39.31 μg/mL) scavenging activity.

Table I — DPPH, $ABTS^{+\cdot}$radical scavenging and α-glucosidase inhibitory activities of azlactones **3a-g**, **6a-f**, **11a-d** and **16b-f**

Compd	DPPH% Inhibition at 25 μg/mL (SC_{50} μg/mL)	ABTS% Inhibition at 20 μg/mL (SC_{50} μg/mL)	AGI% Inhibition at 20 μg/mL (IC_{50} μg/mL)
3a	25.81	37.39	ND
3b	42.32	57.38	ND
3c	ND	ND	ND
3d	0.63	ND	ND
3e	2.09	ND	ND
3f	ND	ND	39.00
3g	9.20	9.29	92.61 (15.05)
6a	9.53	10.66	61.55 (37.05)
6b	1.71	ND	78.93 (20.22)
6c	5.50	11.35	ND
6d	ND	ND	59.33
6e	7.46	8.73	42.69
6f	ND	ND	ND
11a	ND	ND	ND
11b	55.62 (46.70)	67.01 (39.31)	ND
11c	0.48	ND	ND
11d	ND	ND	30.13
16b	16.91	23.24	49.35
16c	25.26	38.12	3.88
16e	16.86	24.01	ND
16f	28.93	40.09	ND
Ascorbic acid	72.90 (28.08)	–	–
Trolox	–	80.20 (18.42)	–
Acarbose	–	–	98.99 (12.57)

ND-Not detected

α-Glucosidase inhibitory activity of azlactones

The intestinal α-glucosidase enzyme plays an important role in digestion of dietary carbohydrate. Inhibition of the enzyme has become an important target in mitigating excursion in postprandial hyperglycemia (PPHG) in diabetic individuals. The percentage inhibition and IC_{50} of α-glucosidase inhibitory activity values for the azlactones **3a-g**, **6a-f**, **11a-d**, **16b-c** and **16e-f** are presented in Table I. The 3-pyridylazlactone **3a**, 2-chloropyridylazlactone **3b**, 2-chloropyridyl-5-substitutedazlactones **3c-f** did not inhibit α-glucosidase. However, 4-pyridylazlactone **3g** (IC_{50}, 15.05 μg/mL) displayed potent α-glucosidase inhibitory activity. 2-Morpholinepyridylazlactones having methyl **6a** (IC_{50}, 37.05 μg/mL) and phenyl **6b** (IC_{50}, 20.22 μg/mL) substitutions displayed α-glucosidase inhibitory activity when compared to thio-morpholine substitution (**6d-e**). Further, 2-chloro-5-substituted triazolylpyridylazlactones **11a-d** and morpholine/thiomorpholine-5-substituted triazolylpyridylazlactones **16b-c**, **16e-f** did not display the activity.

Anti-proliferative activity of azlactones

The IC_{50} values of azlactones **3a-g**, **6a-f**, **11a-d**, **16b-c** and **16e-f** along with the standard drug doxorubicin are presented in Table II. 3-Pyridyl, 2-chloropyridyl, and 4-pyridylazlactones (**3a-b**, **3g** and **11a**) did not show anti-proliferative activity. Methyl, phenyl, *p*-methylphenyl, *p*-fluorophenyl-azlactones (**3c-f**, IC_{50} between 20.39 to 50.05 μM) and morpholine/thio-morpholine substituted pyridylazlactones (**6a-f**, IC_{50} between 19.43 to 33.88 μM) displayed anti-proliferative activity on all tested cell lines. The phenyl substituted 2-chloro-triazolylpyridylazlactone (**11b**) showed better activity selectively on HeLa cell lines when compared to methyl substitution (**11a**). *p*-Methylphenyl and *p*-fluorophenyl tri-azolylpyridylazlactones (**11c-d**) displayed anti-proliferative activity on all the tested cell lines. The phenyl substituted 2-morpholine triazolylpyridylazlactone (**16b**) showed moderate activity on all the tested cell lines. *p*-Methylphenyl morpholine (**16c**) and phenyl thio-morpholine (**16e**) triazolylpyridylazlactones displayed selectively on MCF7 and HeLa cell lines. The

Table II — Anti-proliferative activity of azlactones **3a-g**, **6a-f**, **11a-d** and **16b-f**

Compd[a,b]	MCF7	A549	HeLa
3a	ND	ND	ND
3b	ND	ND	ND
3c	23.12 ± 0.19	50.05 ± 0.15	28.45 ± 0.16
3d	23.94 ± 0.09	32.90 ± 0.10	27.34 ± 0.08
3e	20.85 ± 0.26	27 ± 0.09	20.46 ± 0.08
3f	25 ± 0.33	37.84 ± 0.09	20.39 ± 0.10
3g	ND	ND	ND
6a	21.60 ± 0.05	32.16 ± 0.09	27.31 ± 0.07
6b	26.07 ± 0.12	31.55 ± 0.18	19.48 ± 0.17
6c	25.59 ± 0.21	28.08 ± 0.11	22.30 ± 0.05
6d	21.55 ± 0.08	33.88 ± 0.10	33.36 ± 0.07
6e	26.54 ± 0.13	27.53 ± 0.10	24.29 ± 0.12
6f	25.83 ± 0.33	28.91 ± 0.09	19.43 ± 0.07
11a	ND	ND	ND
11b	20.31 ± 0.14	36.83 ± 0.08	17.74 ± 0.09
11c	17.23 ± 0.11	18.70 ± 0.03	17.73 ± 0.06
11d	14.66 ± 0.10	16.8 ± 0.08	15.53 ± 0.04
16b	22.29 ± 0.12	31.61 ± 0.11	24.55 ± 0.09
16c	18.21 ± 0.20	23.75 ± 0.04	16.59 ± 0.04
16e	18.58 ± 0.04	24.01 ± 0.02	17.63 ± 0.04
16f	14.08 ± 0.16	17.84 ± 0.03	14.84 ± 0.01
Doxorubicin	4.05 ± 1.2	2.89 ± 1.1	3.03 ± 1.1

[a]Cell lines were treated with different concentrations of compounds for 48 h. Cell viability was measured employing MTT assay. [b] IC_{50} (μM) values are indicated as the mean ± SD of three independent experiments. ND-Not detected

p-methylphenyl thio-morpholine triazolylpyridylazlactone (**16f**) showed anti-proliferative activity on all the tested cell lines.

Experimental Section

All the chemicals and reagents were purchased from Aldrich (Sigma-Aldrich, USA), AVRA Chemicals Pvt. Ltd (Hyderabad, India) and were used without further purification. Reactions were monitored by thin layer chromatography (TLC) on pre-coated silica gel 60 F_{254} (mesh) and spots were visualized under UV light. Column chromatographic separations were carried out on silica gel (60-120 mesh). Melting points were determined on a Mettler-Temp apparatus and are uncorrected. IR spectrum was recorded with a Thermo Nicolet Nexus 670 spectrometer. ^{1}H and ^{13}C NMR spectra were recorded on Bruker Avance 300 MHz spectrometers. Chemical shifts (δ) are quoted in parts per million and are referenced to tetramethylsilane (TMS) as internal standard. ESI-MS were obtained on 7070H spectrometer operating at 70 eV using a direct inlet system.

General procedure for the preparation of pyridylazlactones (3a-g, 6a-f, 11a-d, 16b-c, 16e-f)

Sodium acetate (1 mmol) was added to a stirred solution of nicotinaldehyde **1a** (1 mmol) and hippuric acid **2** (1 mmol) in acetic anhydride (3 mmol) at RT. The contents were stirred at 90°C for 15 min. After completion of the reaction (TLC), the reaction temperature was brought to RT and ethanol was added. The contents were left standing overnight. The obtained solid was filtered and washed with hot water to provide the pure azlactone **3a**.

2-Phenyl-4-(pyridin-3-ylmethylene)oxazol-5(4*H*)-one, 3a: Brown solid. m.p.138-140°C. Yield 62%. IR (KBr): 2924 (C-H), 1708 (C=O), 1461 (C=C), 1288 cm^{-1} (C=N); ^{1}H NMR (500 MHz, $CDCl_3$): δ 9.22 (d, *J* = 1.8 Hz, 1H, Ar-H), 8.74 (dt, *J* = 8.0, 1.8 Hz, 1H, Ar-H), 8.68 (dd, *J* = 4.8, 1.5 Hz, 1H, Ar-H), 8.22 (dd, *J* = 5.2, 3.4 Hz, 2H, Ar-H), 7.70-7.65 (m, 1H, Ar-H), 7.58 (dd, *J* = 10.6, 4.7 Hz, 2H, Ar-H), 7.47 (dd, *J* = 8.0, 4.8 Hz, 1H, Ar-H), 7.25 (s, 1H, CH); ^{13}C NMR (75 MHz, $CDCl_3$): δ 166.79, 164.57, 152.92, 151.01, 138.59, 135.20, 133.82, 129.72, 129.03, 128.58, 127.37, 125.17, 123.84; ESI-MS: *m/z* 251 $[M+H]^+$.

4-((2-Chloropyridin-3-yl)methylene)-2-phenyloxazol-5(4*H*)-one, 3b: Yellow solid. m.p.142-144°C. Yield 52%. IR (KBr): 3067 (C-H), 1796 (C=O), 1651 (C=C), 1449 (C=C), 1294 (C=N), 700 cm^{-1} (C-Cl); ^{1}H NMR (300 MHz, $CDCl_3$): δ 9.24 (d, *J* = 6.4 Hz, 1H, Ar-H), 8.42 (d, *J* = 2.7 Hz, 1H, Ar-H), 8.19 (d, *J* = 7.2 Hz, 2H, Ar-H), 7.72-7.61 (m, 2H, Ar-H), 7.56 (t, *J* = 7.5 Hz, 2H, Ar-H), 7.42 (dd, *J* = 7.6, 4.7 Hz, 1H, Ar-H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 166.41, 165.38, 152.36, 150.55, 141.35, 136.22, 134.11, 129.09, 128.70, 128.46, 124.96, 124.39, 122.85.

4-((2-Chloro-5-methylpyridin-3-yl)methylene)-2-phenyloxazol-5(4*H*)-one, 3c: Yellow solid. m.p. 182-184°C. Yield 32%. IR (KBr): 2925 (CH_3), 1792 (C=O), 1651 (C=C), 1562 (C=C), 1436 (C=N), 1152 (C-O), 696 cm^{-1} (C-Cl); ^{1}H NMR (500 MHz, $CDCl_3$): δ 9.02 (d, *J* = 2.3 Hz, 1H, Ar-H), 8.25 (d, *J* = 1.9 Hz, 1H, Ar-H), 8.19 (d, *J* = 1.0 Hz, 1H, Ar-H), 8.17 (t, *J* = 1.6 Hz, 1H, Ar-H), 7.69-7.65 (m, 1H, Ar-H), 7.62 (s, 1H, CH), 7.59-7.54 (m, 2H, Ar-H), 2.46 (s, 3H, CH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 166.46, 165.23, 151.01, 149.99, 141.71, 135.92, 134.05, 132.71, 129.09, 128.66, 127.63, 125.03, 124.75, 17.98; ESI-MS: *m/z* 299 $[M+H]^+$.

4-((2-Chloro-5-phenylpyridin-3-yl)methylene)-2-phenyloxazol-5(4*H*)-one, 3d: Yellow solid. m.p. 184-186°C. Yield 25%. IR (KBr): 3054 (C-H), 1802

(C=O), 1648 (C=C), 1553 (C=C), 1420 (C=N), 1157 (C-O), 696 cm^{-1} (C-Cl); ^1H NMR (500 MHz, $CDCl_3$): δ 9.47 (d, *J* = 2.5 Hz, 1H, Ar-H), 8.63 (d, *J* = 2.5 Hz, 1H, Ar-H), 8.17 (s, 1H, Ar-H), 8.15 (d, *J* = 1.3 Hz, 1H, Ar-H), 7.71-7.67 (m, 3H, Ar-H), 7.66 (d, *J* = 7.4 Hz, 1H, Ar-H), 7.60-7.53 (m, 4H, Ar-H), 7.50 (t, *J* = 7.4 Hz, 1H, Ar-H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 166.36, 165.46, 151.26, 148.72, 139.47, 136.39, 136.22, 136.14, 134.13, 129.38, 129.14, 128.72, 128.69, 128.17, 127.09, 124.96, 124.42; ESI-MS: *m/z* 361 $[M+H]^+$.

4-((2-Chloro-5-(*p*-tolyl)pyridin-3-yl)methylene)-2-phenyloxazol-5(4*H*)-one, 3e: Yellow solid. m.p. 166-168°C. Yield 47%. IR (KBr): 2929 (CH_3), 1802 (C=O), 1651 (C=C), 1326 (C=N), 1154 (C-O), 701 cm^{-1} (C-Cl); ^1H NMR (500 MHz, $CDCl_3$): δ 9.45 (d, *J* = 2.4 Hz, 1H, Ar-H), 8.62 (d, *J* = 2.5 Hz, 1H, Ar-H), 8.16 (d, *J* = 7.9 Hz, 2H, Ar-H), 7.68 (s, 1H, CH), 7.66 (d, *J* = 7.6 Hz, 1H, Ar-H), 7.57 (dd, *J* = 17.0, 8.0 Hz, 4H, Ar-H), 7.37 (d, *J* = 8.1 Hz, 2H, Ar-H), 2.46 (s, 3H, CH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 166.33, 165.34, 150.89, 148.51, 139.13, 138.79, 136.24, 135.98, 134.07, 133.18, 130.06, 129.10, 128.62, 128.03, 126.83, 124.93, 124.44, 21.20; ESI-MS: *m/z* 375 $[M+H]^+$.

4-((2-Chloro-5-(4-fluorophenyl)pyridin-3-yl) methylene)-2-phenyloxazol-5(4*H*)-one, 3f: Yellow solid. m.p.240-242°C. Yield 60%. IR (KBr): 2924 (C-H), 1803 (C=O), 1651 (C=C), 1555 (C=C), 1417 (C=N), 1157 (C-O), 830 (C-F), 700 cm^{-1} (C-Cl); ^1H NMR (300 MHz, $CDCl_3$): δ 9.41 (d, *J* = 2.4 Hz, 1H, Ar-H), 8.59 (d, *J* = 2.4 Hz, 1H, Ar-H), 8.15 (d, *J* = 7.3 Hz, 2H, Ar-H), 7.71-7.61 (m, 4H, Ar-H), 7.56 (t, *J* = 7.5 Hz, 2H, Ar-H), 7.29 (s, 1H, Ar-H), 7.23 (s, 1H, CH); ^{13}C NMR (75 MHz, $CDCl_3$): δ 166.29, 165.59, 151.28, 148.47, 139.31, 136.49, 135.24, 134.21, 132.39, 129.17, 128.88, 128.81, 128.68, 128.22, 124.94, 124.26 , 116.53, 116.36; ESI-MS: *m/z* 379 $[M+H]^+$.

2-Phenyl-4-(pyridin-4-ylmethylene)oxazol-5(4*H*)-one, 3g: Brown solid. m.p.108-110°C. Yield 68%. IR (KBr): 2923 (C-H), 1720 (C=O), 1646 (C=C), 1467 (C=C), 1264 cm^{-1} (C=N); ^1H NMR (500 MHz, $CDCl_3$): δ 8.76 (d, *J* = 6.0 Hz, 2H, Ar-H), 8.27-8.17 (m, 2H, Ar-H), 8.03 (d, *J* = 6.0 Hz, 2H, Ar-H), 7.67 (dd, *J* = 10.6, 4.3 Hz, 1H, Ar-H), 7.57 (t, *J* = 7.7 Hz, 2H, Ar-H), 7.14 (s, 1H, CH); ^{13}C NMR (75 MHz, $CDCl_3$): δ 166.24, 164.76, 150.36, 149.87, 140.02, 137.09, 134.26, 129.40, 128.49, 128.40, 127.73, 126.83, 124.97, 123.47; ESI-MS: *m/z* 251 $[M+H]^+$.

4-((5-Methyl-2-morpholinopyridin-3-yl)methylene)-2-phenyloxazol-5(4*H*)-one, 6a: Orange red solid. m.p.166-168°C. Yield 30%. IR (KBr): 2901 (CH_3), 2845 (CH_2), 1788 (C=O), 1647 (C=C), 1446 (C=C), 1370 (C=N), 1220 (C-O), 1115 (C-N), 1056 cm^{-1} (C-O); ^1H NMR (300 MHz, $CDCl_3$): δ 8.70 (d, *J* = 1.8 Hz, 1H, Ar-H), 8.20 (s, 2H, Ar-H), 8.18 (s, 1H, Ar-H), 7.64 (d, *J* = 7.3 Hz, 1H, Ar-H), 7.57 (t, *J* = 7.4 Hz, 2H, Ar-H), 7.46 (s, 1H, CH), 3.94-3.89 (m, 4H, $2CH_2$), 3.30-3.25 (m, 4H, $2CH_2$), 2.39 (s, 3H, CH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 167.24, 163.58, 160.86, 150.19, 141.52, 133.46, 133.39, 128.99, 128.33, 127.39, 127.33, 125.26, 119.33, 66.95, 52.01, 17.81; ESI-MS: *m/z* 350 $[M+H]^+$.

4-((2-Morpholino-5-phenylpyridin-3-yl)methylene)-2-phenyloxazol-5(4*H*)-one, 6b: Orange red solid. m.p.206-208°C. Yield 44%. IR (KBr): 2925 (CH_2), 1789 (C=O), 1650 (C=C), 1448 (C=C), 1322 (C=N), 1228 (C-O), 1114 cm^{-1} (C-N); ^1H NMR (500 MHz, $CDCl_3$): δ 9.08 (dd, *J* = 21.9, 2.4 Hz, 1H, Ar-H), 8.55 (dd, *J* = 23.1, 2.4 Hz, 1H, Ar-H), 8.15 (dd, *J* = 7.0, 5.9 Hz, 2H, Ar-H), 7.66-7.60 (m, 2H, Ar-H), 7.55 (ddd, *J* = 10.9, 8.0, 4.4 Hz, 4H, Ar-H), 7.48 (dd, *J* = 17.4, 8.9 Hz, 2H, Ar-H), 7.43 (d, *J* = 4.5 Hz, 1H, Ar-H), 3.94-3.90 (m, 4H, $2CH_2$), 3.41-3.36 (m, 4H, $2CH_2$); ^{13}C NMR (75 MHz, $CDCl_3$): δ 167.07, 163.94, 161.60, 148.08, 147.79, 139.33, 136.72, 133.81, 133.64, 129.32, 129.13, 129.03, 128.36, 127.88, 126.84, 126.70, 119.06, 66.90, 51.77; ESI-MS: *m/z* 412 $[M+H]^+$.

4-((2-Morpholino-5-(*p*-tolyl)pyridin-3-yl)methylene)-2-phenyloxazol-5(4*H*)-one, 6c: Orange red solid. m.p.220-222°C. Yield 51%. IR (KBr): 2922 (CH_2), 1803 (C=O), 1664 (C=C), 1449 (C=C), 1382 (C=N), 1239 (C-O), 1159 cm^{-1} (C-N); ^1H NMR (300 MHz, $CDCl_3$): δ 9.11 (d, *J* = 2.3 Hz, 1H, Ar-H), 8.57 (d, *J* = 2.4 Hz, 1H, Ar-H), 8.17 (d, *J* = 7.2 Hz, 2H, Ar-H), 7.64 (d, *J* = 7.3 Hz, 1H, Ar-H), 7.56 (dd, *J* = 7.8, 2.6 Hz, 4H, Ar-H), 7.47 (s, 1H, CH), 7.35 (d, *J* = 7.9 Hz, 2H, Ar-H), 3.98-3.91 (m, 4H, $2CH_2$), 3.42-3.35 m, 4H, $2CH_2$), 2.45 (s, 3H, CH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 167.19, 163.72, 161.35, 147.94, 139.29, 137.56, 134.56, 133.65, 133.50, 130.98, 129.84, 129.01, 128.35, 127.17, 126.53, 125.38, 119.17, 66.93, 51.85, 21.15; ESI-MS: *m/z* 426 $[M+H]^+$.

4-((5-Methyl-2-thiomorpholinopyridin-3-yl) methylene)-2-phenyloxazol-5(4*H*)-one, 6d: Orange red solid. m.p.186-188°C. Yield 55%. IR (KBr): 2917 (CH_2), 1790 (C=O), 1651 (C=C), 1441 (C=C), 1376 (C=N), 1227 (C-O), 1175 (C-N), 704 cm^{-1} (C-S); ^1H NMR (300 MHz, $CDCl_3$) δ 8.69 (s, 1H, Ar-H),

8.19 (d, J = 7.1 Hz, 3H, Ar-H), 7.59 (dt, J = 14.7, 7.2 Hz, 3H, Ar-H), 7.40 (s, 1H, CH), 3.62-3.50 (m, 4H, $2CH_2$), 2.95-2.77 (m, 4H, $2CH_2$), 2.39 (s, 3H, CH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 167.27, 163.53, 161.64, 150.13, 141.46, 133.44, 133.29, 128.98, 128.31, 127.38, 127.25, 125.44, 119.17, 54.02, 27.79, 17.80; ESI-MS: *m/z* 366 $[M+H]^+$.

2-Phenyl-4-((5-phenyl-2-thiomorpholinopyridin-3-yl)methylene)oxazol-5(4*H*)-one, 6e: Orange red solid. m.p.158-160°C. Yield 41%. IR (KBr): 3027 (C-H), 2919 (CH_2), 1793 (C=O), 1649 (C=C), 1447 (C=C), 1368 (C=N), 1224 (C-O), 1189 (C-N), 757 cm^{-1} (C-S); ^{1}H NMR (300 MHz, $CDCl_3$): δ 9.08 (dd, J = 13.2, 2.3 Hz, 1H, Ar-H), 8.54 (dd, J = 13.9, 2.4 Hz, 1H, Ar-H), 8.15 (d, J = 7.2 Hz, 2H, Ar-H), 7.63 (t, J = 6.9 Hz, 3H, Ar-H), 7.59-7.46 (m, 5H, Ar-H), 7.40 (dd, J = 15.7, 8.0 Hz, 2H, Ar-H), 3.71-3.62 (m, 4H, $2CH_2$), 2.90-2.83 (m, 4H, $2CH_2$); ^{13}C NMR (75 MHz, $CDCl_3$): δ 167.14, 163.69, 162.16, 147.96, 139.42, 133.56, 133.47, 129.09, 129.02, 128.99, 128.31, 127.81, 127.61, 127.26, 127.10, 126.63, 119.15, 53.81, 27.70; ESI-MS: *m/z* 428 $[M+H]^+$.

2-Phenyl-4-((2-thiomorpholino-5-(*p*-tolyl)pyridin-3-yl)methylene)oxazol-5(4*H*)-one, 6f: Orange red solid. m.p.246-248°C. Yield 36%. IR (KBr): 2910 (CH_2), 1792 (C=O), 1648 (C=C), 1444 (C=C), 1366 (C=N), 1224 (C-O), 1185 (C-N), 700 cm^{-1} (C-S); ^{1}H NMR (500 MHz, $CDCl_3$): δ 9.08 (d, J = 2.4 Hz, 1H, Ar-H), 8.54 (d, J = 2.4 Hz, 1H, Ar-H), 8.15 (d, J = 7.3 Hz, 2H, Ar-H), 7.63 (t, J = 7.4 Hz, 1H, Ar-H), 7.54 (dt, J = 7.3, 3.5 Hz, 4H, Ar-H), 7.39 (s, 1H, CH), 7.32 (d, J = 7.9 Hz, 2H, Ar-H), 3.66-3.63 (m, 4H, $2CH_2$), 2.89-2.85 (m, 4H, $2CH_2$), 2.43 (s, 3H, CH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 167.26, 163.69, 162.09, 147.91, 139.28, 137.56, 133.57, 133.51, 133.50, 130.86, 129.85, 129.03, 128.35, 127.27, 126.54, 125.42, 119.27, 53.88, 27.76, 21.26; ESI-MS: *m/z* 442 $[M+H]^+$.

4-((1-((2-Chloro-5-methylpyridin-3-yl)methyl)-1*H*-1,2,3-triazol-4-yl)methylene)-2-phenyloxazol-5(4*H*)-one, 11a: Colorless solid. m.p.154-156°C. Yield 22%. IR (KBr): 2962 (C-H), 2854 (CH_3), 1801 (C=O), 1665 (C=C), 1450 (C=C), 1276 (C=N), 1149 (C-N), 697 cm^{-1} (C-Cl); ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.76 (s, 1H, Ar-H), 8.24 (d, J = 1.9 Hz, 1H, Ar-H), 8.13 (dt, J = 7.9, 3.7 Hz, 2H, Ar-H), 7.67-7.61 (m, 1H, Ar-H), 7.57-7.51 (m, 3H, Ar-H), 7.31 (dd, J = 9.3, 1.9 Hz, 1H, Ar-H), 5.74 (d, J = 12.4 Hz, 2H, CH_2), 2.31 (s, 3H, CH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 165.96, 163.79, 150.30, 147.08, 142.85, 139.20, 135.62, 133.84, 133.75, 133.53, 129.04, 128.41, 127.97, 125.13, 120.87, 51.00, 17.63; ESI-MS: *m/z* 380 $[M+H]^+$.

4-((1-((2-Chloro-5-phenylpyridin-3-yl)methyl)-1*H*-1,2,3-triazol-4-yl)methylene)-2-phenyloxazol-5(4*H*)-one, 11b: Colorless solid. m.p.174-176°C. Yield 41%. IR (KBr): 3059 (C-H), 2923 (CH_2), 1797 (C=O), 1670 (C=C), 1432 (C=C), 1279 (C=N), 1157 (C-N), 765 cm^{-1} (C-Cl); ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.80 (s, 1H, Ar-H), 8.63 (d, J = 2.4 Hz, 1H, Ar-H), 8.16-8.08 (m, 2H, Ar-H), 7.71 (d, J = 2.4 Hz, 1H, Ar-H), 7.62 (d, J = 7.4 Hz, 1H, Ar-H), 7.55-7.51 (m, 3H, Ar-H), 7.49-7.44 (m, 5H, Ar-H), 5.85 (s, 2H, CH_2); ^{13}C NMR (75 MHz, $CDCl_3$): δ 165.91, 163.80, 148.58, 148.17, 142.84, 137.04, 136.91, 135.39, 133.89, 133.79, 129.29, 129.02, 128.92, 128.70, 128.39, 128.00, 127.05, 125.08, 120.75, 51.10; ESI-MS: *m/z* 442 $[M+H]^+$.

4-((1-((2-Chloro-5-(*p*-tolyl)pyridin-3-yl)methyl)-1*H*-1,2,3-triazol-4-yl)methylene)-2-phenyloxazol-5(4*H*)-one, 11c: Colorless solid. m.p.232-234°C. Yield 46%. IR (KBr): 3012 (C-H), 2919 (CH_3), 1803 (C=O), 1673 (C=C), 1440 (C=C), 1282 (C=N), 1158 (C-N), 975 cm^{-1} (C-Cl); ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.80 (s, 1H, Ar-H), 8.61 (d, J = 1.9 Hz, 1H, Ar-H), 8.12 (d, J = 7.5 Hz, 2H, Ar-H), 7.68 (t, J = 4.1 Hz, 1H, Ar-H), 7.62 (d, J = 7.3 Hz, 1H, Ar-H), 7.53 (t, J = 7.5 Hz, 3H, Ar-H), 7.39 (d, J = 8.0 Hz, 2H, Ar-H), 7.27 (d, J = 1.6 Hz, 1H, Ar-H), 7.25 (s, 1H, CH), 5.84 (s, 2H, CH_2), 2.39 (s, 3H, CH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 165.96, 163.78, 148.22, 148.03, 142.86, 139.07, 136.85, 136.75, 133.86, 133.74, 132.48, 130.01, 129.03, 128.82, 128.40, 127.99, 126.87, 125.09, 120.83, 51.13, 21.14; ESI-MS: *m/z* 456 $[M+H]^+$.

4-((1-((2-Chloro-5-(4-fluorophenyl)pyridin-3-yl)methyl)-1*H*-1,2,3-triazol-4-yl)methylene)-2-phenyloxazol-5(4*H*)-one, 11d: Colorless solid. m.p.176-178°C. Yield 49%. IR (KBr): 2925 (C-H), 2851 (CH_2), 1805 (C=O), 1668 (C=C), 1439 (C=C), 1233 (C=N), 1159 (C-N), 837 (C-F), 699 cm^{-1} (C-Cl); ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.80 (s, 1H, Ar-H), 8.59 (d, J = 2.4 Hz, 1H, Ar-H), 8.15-8.10 (m, 2H, Ar-H), 7.66 (dd, J = 4.7, 1.8 Hz, 1H, Ar-H), 7.63 (dd, J = 5.0, 3.7 Hz, 1H, Ar-H), 7.56-7.52 (m, 3H, Ar-H), 7.48-7.44 (m, 2H, Ar-H), 7.16 (ddd, J = 10.4, 6.0, 2.6 Hz, 2H, Ar-H), 5.84 (s, 2H, CH_2); ^{13}C NMR (75 MHz, $CDCl_3$): δ 165.92, 163.84, 148.62, 148.01, 142.88, 136.86, 135.95, 134.23, 133.78, 130.01, 129.04, 128.91, 128.84, 128.40, 125.07, 120.74, 116.48, 116.31, 51.05; ESI-MS: *m/z* 460 $[M+H]^+$.

4-((1-((2-Morpholino-5-phenylpyridin-3-yl)methyl)-1*H*-1,2,3-triazol-4-yl)methylene)-2-phenyloxazol-5(4*H*)-one, 16b: Colorless solid. m.p.209-211°C. Yield 53%. IR (KBr): 2966 (C-H), 2850 (CH_2), 1802 (C=O), 1666 (C=C), 1465 (C=C), 1326 (C=N), 1238 (C-O), 1111 cm^{-1} (C-N); ^1H NMR (300 MHz, $CDCl_3$): δ 8.65 (s, 1H, CH, Ar-H), 8.60 (d, J = 2.3 Hz, 1H, Ar-H), 8.08 (d, J = 7.3 Hz, 2H, Ar-H), 7.62 (t, J = 7.4 Hz, 1H, Ar-H), 7.57 (d, J = 2.2 Hz, 1H, Ar-H), 7.54 (s, 1H, Ar-H), 7.51-7.46 (m, 4H, Ar-H), 7.43 (t, J = 7.5 Hz, 2H, Ar-H), 7.36 (t, J = 7.2 Hz, 1H, Ar-H), 5.78 (s, 2H, CH_2), 3.96-3.93 (m, 4H, $2CH_2$), 3.23-3.20 (m, 4H, $2CH_2$); ^{13}C NMR (75 MHz, $CDCl_3$): δ 165.80, 162.70, 159.56, 145.44, 141.02, 136.45, 136.03, 133.82, 133.10, 130.90, 129.72, 129.21, 129.09, 128.05, 127.73, 126.31, 124.91, 123.44, 120.30, 66.14, 50.66, 49.53.

4-((1-((2-Morpholino-5-(*p*-tolyl)pyridin-3-yl)methyl)-1*H*-1,2,3-triazol-4-yl)methylene)-2-phenyloxazol-5(4*H*)-one, 16c: Colorless solid. m.p.212-214°C. Yield 68%. IR (KBr): 2922 (C-H), 2851 (CH_2), 1800 (C=O), 1665 (C=C), 1462 (C=C), 1237 (C=N), 1156 (C-N), 1111 (C-N), 1043 cm^{-1} (C-O); ^1H NMR (300 MHz, $CDCl_3$): δ 8.65 (s, 1H, CH, Ar-H), 8.59 (d, J = 2.3 Hz, 1H, Ar-H), 8.11-8.05 (m, 2H, Ar-H), 7.62 (t, J = 7.5 Hz, 1H, Ar-H), 7.56-7.53 (m, 2H, Ar-H), 7.50 (t, J = 7.8 Hz, 2H, Ar-H), 7.36 (d, J = 8.1 Hz, 2H, Ar-H), 7.23 (d, J = 7.9 Hz, 2H, Ar-H), 5.77 (s, 2H, CH_2), 3.96-3.93 (m, 4H, $2CH_2$), 3.22-3.18 (m, 4H, $2CH_2$), 2.37 (s, 3H, CH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 166.00, 163.71, 159.69, 146.64, 142.71, 138.01, 136.12, 133.86, 133.69, 133.62, 132.88, 129.83, 129.00, 128.34, 127.79, 126.61, 125.18, 122.66, 121.17, 67.03, 51.38, 50.01, 21.10; ESI-MS: *m/z* 507 $[M+H]^+$.

2-Phenyl-4-((1-((5-phenyl-2-thiomorpholinopyridin-3-yl)methyl)-1*H*-1,2,3-triazol-4-yl)methylene)oxazol-5(4*H*)-one, 16e: Colorless solid. m.p.206-208°C. Yield 51%. IR (KBr): 2908 (C-H), 2840 (CH_2), 1787 (C=O), 1664 (C=C), 1448 (C=C), 1277 (C=N), 1152 (C-N), 858 cm^{-1} (C-S); ^1H NMR (300 MHz, $CDCl_3$): δ 8.66 (s, 1H, CH, Ar-H), 8.59 (d, J = 2.4 Hz, 1H, Ar-H), 8.13-8.07 (m, 2H, Ar-H), 7.65-7.58 (m, 2H, Ar-H), 7.54 (s, 3H, Ar-H), 7.51 (t, J = 7.8 Hz, 2H, Ar-H), 7.48-7.45 (m, 2H, Ar-H), 7.43 (t, J = 7.5 Hz, 2H, Ar-H), 7.37 (dt, J = 9.3, 4.2 Hz, 1H, Ar-H), 5.73 (s, 2H, CH_2), 3.48-3.45 (m, 4H, $2CH_2$), 2.93-2.89 (m, 4H, $2CH_2$); ^{13}C NMR (75 MHz, $CDCl_3$): δ 146.80, 136.75, 136.49, 133.73, 132.95, 129.13, 129.04, 128.39, 128.05, 127.76, 126.79, 122.87, 121.14, 53.43, 50.02, 28.01; ESI-MS: *m/z* 509 $[M+H]^+$.

2-Phenyl-4-((1-((2-thiomorpholino-5-(*p*-tolyl)pyridin-3-yl)methyl)-1*H*-1,2,3-triazol-4-yl)methylene)oxazol-5(4*H*)-one, 16f: Colorless solid. m.p.214-216°C. Yield 51%. IR (KBr): 2910 (CH_2), 1802 (C=O), 1663 (C=C), 1450 (C=C), 1231 (C=N), 1159 (C-N), 863 cm^{-1} (C-S); ^1H NMR (300 MHz, $CDCl_3$): δ 8.66 (s, 1H, CH, Ar-H), 8.57 (d, J = 2.2 Hz, 1H, Ar-H), 8.09 (d, J = 7.3 Hz, 2H, Ar-H), 7.62 (t, J = 7.4 Hz, 1H, Ar-H), 7.57 (d, J = 2.1 Hz, 1H, Ar-H), 7.54-7.48 (m, 3H, Ar-H), 7.36 (d, J = 8.0 Hz, 2H, Ar-H), 7.23 (d, J = 7.9 Hz, 2H, Ar-H), 5.72 (s, 2H, CH_2), 3.48-3.43 (m, 3H, $2CH_2$), 2.93-2.88 (m, 3H, $2CH_2$), 2.37 (s, 3H, CH_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 166.01, 163.70, 160.68, 146.63, 142.65, 138.01, 136.27, 133.81, 133.69, 133.59, 132.95, 129.82, 129.02, 128.37, 127.75, 126.61, 125.14, 122.87, 121.15, 53.45, 50.03, 28.03, 21.10; ESI-MS: *m/z* 523 $[M+H]^+$.

DPPH free radical scavenging activity

Assay for the scavenging of stable free radical 1,1-diphenyl-2-picrylhydrazyl (DPPH) was done as reported earlier[23]. Briefly, in a 96-well micro plate, to 25 µL of test sample dissolved in DMSO (1 mg/mL), 125 µL of 0.1 M tris–HCl buffer (*p*H 7.4) and 125 µL of 0.5 mM DPPH solution dissolved in absolute ethyl alcohol were added. The reaction mixture was shaken well and incubated in the dark for 30 min and read at 517 nm spectrophotometrically (Spectra Max plus 384, Molecular Devices Corporation, Sunnyvale, CA, USA). Percentage of DPPH scavenging was calculated as $(1-B/A) \times 100$ where A represents absorbance of control without test samples, and B represents absorbance in presence of test samples.

$ABTS^{+\cdot}$ Free radical scavenging assay

Scavenging of the $ABTS^{+\cdot}$[2,2′-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid)] cation was performed with suitable modifications[24]. Briefly, 100 mL stock solution of $ABTS^{+\cdot}$ (0.5 mM) was prepared by addition of 1 mL potassium persulfate (6.89 mM PBS, *p*H 8.0). The mixture was stored in the dark for 16 h. Test compounds were dissolved in DMSO (5 mg/mL). Primary screening was done by mixing 10 µL of test compounds in 100 µL of methanol followed by 190 µL of $ABTS^{+\cdot}$ in a 96-well microplate. Absorbance of decolorized $ABTS^{+\cdot}$ was measured at 734 nm after 15 min incubation in the dark on a BioTek synergy multi-mode microplate reader. For each test sample a separate blank sample (devoid of $ABTS^{+\cdot}$) was used for background subtraction. The percentage of $ABTS^{+\cdot}$

scavenging was calculated applying the following formula:% $ABTS^{+\cdot}$ scavenging = [($Absorbance_{control}$ − $Absorbance_{test}$)/ $Absorbance_{control}$ × 100]. Various serial dilutions of active compounds were prepared and tested for determination of SC_{50} values. Suitable regression analysis was applied for calculation of SC_{50}.

α-Glucosidase inhibitory assay

α-Glucosidase inhibitory activities were determined as per earlier reported methods[23]. Rat intestinal acetone powder in normal saline (100:1; w/v) was sonicated properly and the supernatant was used as a source of crude intestinal α-glucosidase after centrifugation. In brief, 20 µL of test samples (5 mg/mL DMSO solution) were reconstituted in 100 µL of 100 mM-phosphate buffer (*p*H 6.8) in 96-well microplate and incubated with 50 µL of crude intestinal α-glucosidase for 5 min before 50 µL substrate (5 mM, *p*-nitrophenyl-α-D-glucopyranoside prepared in same buffer) was added. Release of *p*-nitrophenol was measured at 405 nm spectrophotometrically (Spectra Max plus 384, Molecular Devices Corporation, Sunnyvale, CA, USA) 5 min after incubation with substrate. Individual blanks for test samples were prepared to correct background absorbance where substrate was replaced with 50 µL of buffer. Control sample contained 10 µL DMSO in place of test samples. Percentage of enzyme inhibition was calculated as (1-B/A) × 100 where [A] represents absorbance of control without test samples, and [B] represents absorbance in presence of test samples. For calculation of 50% enzyme inhibitory activity ($IC50_{\%}$) more than five dilutions of primary screening concentration (5 mg/mL DMSO solution) of test compounds were prepared. The IC_{50} values were calculated applying logarithmic regression analysis.

Anti-proliferative activity assay

All the cell lines used in this study were purchased from the American Type Culture Collection (ATCC, United States). A549, HeLa and MCF7 were grown in Dulbecco's modified Eagle's medium (containing 10% FBS in a humidified atmosphere of 5% CO_2 at 37°C). DU145 cells were cultured in Eagle's minimal essential medium (MEM) containing non-essential amino acids, 1 mM sodium pyruvate, 10 mg/mL bovine insulin, and 10% FBS. Cells were trypsinized when sub-confluent from T25 flasks/60 mm dishes and seeded in 96-well plates. The synthesized compounds were evaluated for their *in vitro* anti-proliferative activity in the above mentioned cell lines. A protocol of 48 h continuous drug exposure was used, and a MTT cell proliferation assay was employed to estimate cell viability[25]. The cell lines were grown in their respective media containing 10% fetal bovine serum and were seeded into 96-well microtiter plates in 200 µL aliquots at plating densities depending on the doubling time of individual cell lines. The microtiter plates were incubated at 37°C, 5% CO_2, 95% air, and 100% relative humidity for 24 h prior to addition of experimental drugs. Aliquots of 2 µL of the test compounds were added to the wells already containing 198 µL of cells, resulting in the required final concentrations of test compounds. For each compound, four concentrations (1, 10, 25 and 100 µM) were evaluated, and each was done in triplicate wells. Plates were incubated further for 48 h, and the experiment was terminated by the addition of 10 µL of 5% MTT and incubated further for 60 min at 37°C. Later, the plates were washed and air-dried and bound stain was subsequently dissolved using 100 µL of DMSO. The absorbance was monitored on a multimode plate reader (Tecan M 200) at a wavelength of 560 nm. Percent growth was calculated on a plate by plate basis for test wells relative to control wells. The above determinations were repeated thrice. The growth inhibitory effects of the compounds were analyzed by generating dose response curves as a plot of the percentage surviving cells *versus* compound concentration. The sensitivity of the cells to the test compound was expressed in terms of IC_{50}, the concentration of compound that produced 50% reduction as compared to the control absorbance. IC_{50} values are indicated as means ± SD of three independent experiments.

Conclusion

In conclusion, series of azlactones **3a-g**, **6a-f**, **11a-d**, **16b-c**, and **16e-f** were prepared and evaluated for their DPPH, $ABTS^{+\cdot}$ radical scavenging, α-glucosidase inhibitory and anti-proliferative activities. Compound **3g** was the most potent α-glucosidase inhibitor in present series of the compounds. Compounds **11c-d** and **16f** show anti-proliferative activity on the all the three tested cell lines. However, compounds **16c** and **16e** displayed selective anti-proliferative activity on MCF7 and HeLa cell lines.

Acknowledgements

The authors thank Dr. S. Chandrasekhar, Director, CSIR-IICT for constant encouragement. CD thanks

CSIR, New Delhi, for his research fellowship. BCR acknowledges CSIR, New Delhi, for financial support through the programme ORIGIN of XII five year plan (CSC0108).

References

1 Samaritoni J G, Demeter D A, Gifford J M, Watson G B, Kempe M S & Bruce T J, *J Agri Food Chem*, 51 (2003) 3035.

2 Tian Z, Jiang Z, Li Z, Song G & Huang Q, *J Agri Food Chem*, 55 (2007) 143.

3 Moriya K, Shibuya K, Hattori Y, Tsuboi S, Shiokawa K & Kagabu S, *Biosci Biotechnol Biochem*, 56 (1992) 364.

4 Hanket T, Brunne R M, Miller H & Reichel F, *Angew Chem Int Ed*, 38 (1999) 643.

5 Kumar R & Chandra R, *Adv Heterocycl Chem*, 78 (2001) 269.

6 Gavin D H, *Tetrahedron*, 60 (2004) 6043.

7 Gigou A, Beaucourt J P, Lellouche J P & Gree R, *Tetrahedron Lett*, 32 (1991) 635.

8 Caijo F, Mosset P, Gree R, Bouchez V A, Boutin J, Renard P, Caignard D H & Dacquet C, *Bioorg Med Chem Lett*, 15 (2005) 4421.

9 Conway P A, Devine K & Paradisi F, *Tetrahedron*, 65 (2009) 2935.

10 Gangadasu B, Raju B C & Rao V J, *J Heterocycl Chem*, 46 (2009) 1213.

11 Gangadasu B, Reddy M J R, Ravinder M, Kumar S B, Raju B C, Kumar K P, Murthy U S N & Rao V J, *Eur J Med Chem*, 44 (2009) 4661.

12 Narender P, Ravinder M, Sadhu P S, Raju B C, Ramesh C & Rao V J, *Helv Chim Acta*, 92 (2009) 959.

13 Narender P, Srinivas U, Gangadasu B, Biswas S & Rao V J, *Bioorg Med Chem Lett*, 15 (2005) 5378.

14 Narender P, Gangadasu B, Ravinder M, Srinivas U, Swamy G Y S K, Ravi Kumar K & Rao V J, *Tetrahedron*, 62 (2006) 954.

15 Raju B C & Suman P, *Chem Eur J*, 16 (2010) 11840.

16 Suman P, Rao R N, Raju B C, Sriram D & Koushik P V, *Med Chem Res*, 23 (2014) 199.

17 Suman P, Dayakar Ch, Rajkumar K, Yashwanth B, Yogeeswari P, Sriram D, Rao J V & Raju B C, *Bioorg Med Chem Lett*, 25 (2015) 2390.

18 Suman P, Murthy T R, Rajkumar K, Srikanth D, Dayakar Ch, Kishor Ch, Addlagatta A, Kalivendi S V & Raju B C, *Eur J Med Chem*, 90 (2015) 603.

19 Gangadasu B, Narender P, Kumar S B, Ravinder M, Rao B A, Ramesh Ch, Raju B C & Rao V J, *Tetrahedron*, 62 (2006) 8398.

20 Odlo K, Chabert J F D, Ducki S, Gani O A B S M, Sylte I & Hansen T V, *Bioorg Med Chem*, 18 (2010) 6874.

21 Meldal M & Tornoe C W, *Chem Rev*, 108 (2008) 2952.

22 Thirumurugan P, Matosiuk D & Jozwiak K, *Chem Rev*, 113 (2013) 4905.

23 Raju B C, Tiwari A K, Kumar J A, Ali Z A, Agawane S B, Saidachary G & Madhusudana K, *Bioorg Med Chem*, 18 (2010) 358.

24 Kumar J A, Tiwari A K, Saidachary G, Kishor Ch, Kumar D A, Zehra A, Sridhar B, Addlagatta A & Raju B C, *Med Chem Res*, 23 (2014) 2821.

25 Kumar A S, Reddy M A, Jain N, Kishor C, Murthy T R, Ramesh D, Supriya B, Addlagatta A, Kalivendi S V & Sreedhar B, *Eur J Med Chem*, 60 (2013) 305.