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A facile and efficient protocol for the synthesis of polyfunctionalized 4*H*-pyran derivatives and its antimicrobial evaluation

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ABSTRACT

A series of novel 4*H*-pyran derivatives containing thiophene were designed and synthesized via Domino Knoevenagel condensation–Michael addition–intramolecular *O*-cyclization sequence in a clean, efficient and straight forward manner. Characterizations of synthesized compounds were done by various spectroscopic techniques like IR, NMR and Mass spectrometry. All synthesized compounds have been screened against four anti-bacterial and two anti-fungal species. Among synthesized compound, no one is relatively active against antimicrobial.

***Keywords*:** 4*H*-Pyran, Thiophene, NMSM, Antimicrobial

1. INTRODUCTION

Pyran based heterocyclic compounds are having an important place due to their distinct structures and great potential for further transformations. Active 4*H*-Pyrans are particularity privilege pharmacological scaffolds among the heterocyclic family members[1]. Polyfunctionalized 4*H*-pyran derivatives are very well known compounds and their reactivity has been largely explored[2,3]. Polyfunctionalized 4*H*-pyrans also constitute a structural unit of

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many natural products and biologically interesting compounds[7–9]. which possess various pharmacological activities, such as anti-allergic[5,6], antitumor[7–9], antibacterial[10–13]. 4*H*-Pyran derivatives are also potential calcium channel antagonists, which are structurally similar to biologically active 1,4- dihydropyridines[14]. Cyanopyran derivatives represent one of the modest classes of the compounds possessing a wide range of therapeutic activities such as antibacterial[15,16], antifungal[17–19], anticancer[20,21], antiviral[22–24], anticonvulsant[25,26] etc.

4*H*-pyran derivatives represent an important class of compounds. They are often used in cosmetics and pigments and utilized as potentially biodegradable agrochemicals[27]. Figure 2 represents a glimpse of some of the naturally occurring bioactive pyran-annulated heterocyclic compounds exhibiting diverse kind of pharmaceutical potentials.

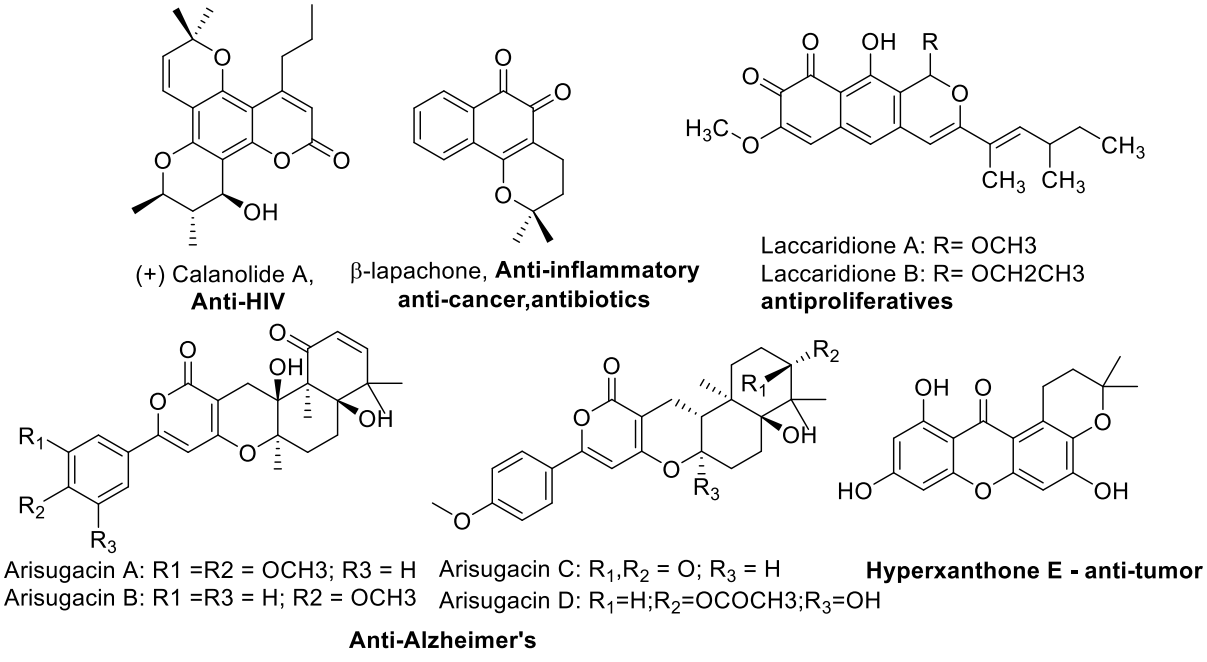


Figure 1 Some of the naturally occurring bioactive compounds bearing pyran-annulated scaffolds

Keeping in mind, various biomedical applications and with a view to further assess the pharmacological profile of new polyfunctionalized pyran heterocycles combine with various biological active/moderately active nucleus have been synthesized in the framework of this article. We have synthesised the polyfunctionalized 4*H*-pyran ring system and evaluated them against five bacterial stain and two fungi to check their potency.

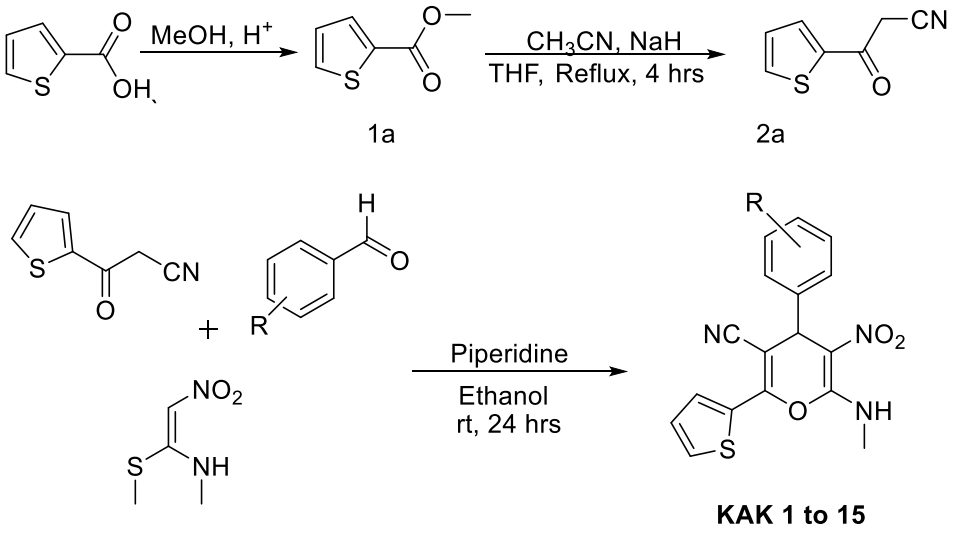
2. EXPERIMENTAL

2. 1. General information

Chemicals and solvents were purchased from the Sigma Aldrich Chemical Co., Merck chemical, Finar and Spectrochem Ltd. The entire chemicals were used without further purification. Precoated plates of silica gel G60 F254 (0.2 mm, Mfg. by Merck) were used for

thin-layer chromatography. Visualization was made under UV light (254 and 365nm) or with an iodine vapour. Spectral analysis of the synthesized compounds was done with the help of FTIR-8400 (Shimadzu); AVANCE-II (400 and 600 MHz) NMR spectrophotometer and GC-MS QP-2010 mass spectrophotometer. NMR (^{1}H & ^{13}C) done by using $CDCl_3$ and DMSO-d_6 as a solvent, TMS as an internal standard and represented in δ ppm. Mass analysis was done by direct probe method. BUCHI rotary evaporator was used for the isolation of products and recovery of solvents. Melting points were measured with help of digital melting point apparatus (Make: Labronics).

2. 2. Synthesis part



Scheme 1

Table 1. Optimization of reaction conditions by selecting the proper solvent, base and appropriate reaction time for the completion of the reaction

Entry	Base (eq.)	Solvent	Time(h)	Yield[a]
1	--	EtOH	24	--
2	Pyridine (1.0)	EtOH	16	33
3	Pyrolidone (1.0)	EtOH	14	25
4	L-Proline (1.0)	EtOH	25	35
5	DABCO (1.0)	EtOH	26	40
6	DBU (1.0)	EtOH	18	36

7	DMAP (1.0)	EtOH	18	42
8	Piperidine (1.0)	EtOH	16	93
9	K_2CO_3 (1.0)	EtOH	24	trace
10	NaOH (1.0)	EtOH	24	trace
11	**Piperidine (0.5)**	**EtOH**	**16**	**93**
12	Piperidine (2)	EtOH	16	93
13	Piperidine (0.5)	MeOH	18	80
14	Piperidine (1.0)	MeOH	16	83
15	Piperidine (0.5)	n-Propanol	24	53
16	Piperidine (0.5)	IPA	22	56
17	Piperidine (0.5)	THF	30	40
18	Piperidine (0.5)	CH_3CN	30	60

*a = Isolated yield

General synthesis of methyl thiophene-2-carboxylate (1a)

In a 100 ml round-bottomed flask, a mixture of 12.8 g of Thiophene-2-carboxylic acid (0.1 mole), 30 ml of methyl alcohol, and 3 ml of concentrated sulfuric acid are placed. The reaction mixture was reflux at 70 ºC for 4 hrs and the reaction was monitored on TLC (30% EtOAc: Hexane). After completion of the reaction, the mixture was poured into ice-cold water and neutralized by $NaHCO_3$, extracted with CH_2Cl_2 (3*50 mL). The extract was washed with brine solution, dried over anhydrous Na_2SO_4 and concentrated in vaccuo to obtain 1a. The product was used without further purification.

General synthesis of 3-oxo-3-(thiophene-2-yl)propanenitrile (2a)

Compound 1a (0.05 mol, 7 g) and NaH (60% in paraffin) (0.15 mol, 3.6 g) in tetrahydrofuran (30 mL) were treated with a solution of acetonitrile (0.05 mol, 2.0 g) in tetrahydrofuran (4 mL) dropwise, and the mixture was then refluxed for 4 h. After completion of the reaction, kept the reaction at room temperature and diluted with diethyl ether. The chemical reaction mixture kept at room temperature for 48 h. The sodium salt precipitate was filtered and washed with diethyl ether. The dry compound was dissolved in water (5 mL) and acidified with HCl up to pH 2. Then extract it by toluene, the collected extracts were dried with Na_2SO_4 and evaporate the solvent in vaccuo to obtain compound 2a. Compound 2a used the next step without further purification.

General procedure for the synthesis of 4-(Substituted phenyl)-6-(methylamino)-5-nitro-2-(thiophen-2-yl)-4*H*-pyran-3-carbonitrile (KAK-1 to KAK-8)

A mixture of 3-Oxo-3-(thiophen-2-yl)propanenitrile (0.151 gm, 1.0 mmol, 1 eq.) and substituted aromatic aldehyde (1.0 mmol, 1 eq.) was dissolved in absolute EtOH (10 mL) in a 25 ml round bottom flask. Then piperidine (0.05 mmol, 0.5 eq.) was added slowly and the reaction mixture was stirred at room temperature. After 15-30 min, the precipitation of imine was formed out from the solution. After formation imine, (E)-N-methyl-1-(methylthio)-2-nitroethenamine (NMSM) (0.148 gm, 1.0 mmol) was added into the reaction mixture, followed stirred for 14-18 hours at room temperature. The consumption of starting material was monitored by TLC, Isolate the product and washed with cold EtOH (2 * 5mL) and dried it.

2. 3. Methodology for biological screening

Sample preparation: Samples were provided by dry powder and stored frozen at -20 °C. Samples were prepared in DMSO and water to a final testing concentration of 32 µg/mL or 20 µM (unless otherwise indicated in the datasheet), in 384-well, non-binding surface plate (NBS) for each bacterial/fungal strain, and in duplicate (n = 2), and keeping the final DMSO concentration to a maximum of 1% DMSO. All the sample preparation were done using liquid handling robots.

2. 3. 1. Antimicrobial Assay

Procedure: All bacteria were cultured in Cation-adjusted Mueller Hinton broth (CAMHB) at 37 °C overnight. A sample of each culture was then diluted 40-fold in fresh broth and incubated at 37 °C for 1.5-3 h. The resultant mid-log phase cultures were diluted (CFU/mL measured by OD600), then added to each well of the compound containing plates, giving a cell density of 5×105 CFU/mL and a total volume of 50 µL. All the plates were covered and incubated at 37 °C for 18 h without shaking.

Analysis: Inhibition of bacterial growth was determined to measure absorbance at 600 nm (OD600), using a Tecan M1000 Pro monochromator plate reader. The percentage of growth inhibition was calculated for each well, using the negative control (media only) and positive control (bacteria without inhibitors) on the same plate as references. The significance of the inhibition values was determined by modified Z-scores, calculated using the median and MAD of the samples (no controls) on the same plate. Samples with inhibition value above 80% and Z-Score above 2.5 for either replicate (n = 2 on different plates) were classed as actives. Samples with inhibition values between 50 - 80% and Z-Score above 2.5 for either replicate (n = 2 on different plates) were classed as partial actives. Samples with inhibition values between 50 - 80% and Z-Score above 2.5 for either replicate (n = 2 on different plates) were classed as partial actives.

2. 3. 2. Antifungal Assay

Procedure: Fungi strains were cultured for 3 days on Yeast Extract-Peptone Dextrose (YPD) agar at 30 °C. A yeast suspension of 1 × 106 to 5 × 106 CFU/mL (as determined by OD530) was prepared from five colonies. The suspension was subsequently diluted and added to each well of the compound-containing plates giving a final cell density of fungi suspension of 2.5

×103 CFU/mL and a total volume of 50 µL. All plates were covered and incubated at 35 °C for 24 h without shaking.

Analysis: Growth inhibition of C. albicans was determined measuring absorbance at 530 nm (OD530), while the growth inhibition of C. neoformans was determined measuring the difference in absorbance between 600 and 570 nm (OD600-570), after the addition of resazurin (0.001% final concentration) and incubation at 35 °C for additional 2 h. The absorbance was measured using a Biotek Synergy HTX plate reader. The percentage of growth inhibition was calculated for each well, using the negative control (media only) and positive control (fungi without inhibitors) on the same plate. The significance of the inhibition values was determined by modified Z-scores, calculated using the median and MAD of the samples (no controls) on the same plate. Samples with inhibition value above 80% and Z-Score above 2.5 for either replicate (n = 2 on different plates) were classed as actives. Samples with inhibition values between 50 - 80% and Z-Score above 2.5 for either replicate (n = 2 on different plates) were classed as partial actives.3

2. 3. 3. Antibiotic Standards Preparation and Quality Control

Colistin and vancomycin were used as positive bacterial inhibitor standards for Gram-negative and Gram-positive bacteria, respectively. Fluconazole was used as a positive fungal inhibitor standard both for C. albicans and C. neoformans. The antibiotics were provided in four concentrations, with two above and two below its MIC value, and plated into the first 8 wells of column 23 of the 384-well NBS plates. The quality control (QC) of the assays was determined by the antimicrobial controls and the Z'-factor (using positive and negative controls). Each plate was deemed to fulfil the quality criteria (pass QC), if the Z'-factor was above 0.4, and the antimicrobial standards showed the full range of activity, with full growth inhibition at their highest concentration, and no growth inhibition at their lowest concentration.

2. 3. 4. Materials

Both the compound preparation plate and the assay plates were purchased from Corning (Corning, NY, USA). CAMHB from Bacto Laboratories (Mount Pritchard, Australia) was used as growth media for bacteria; culture agar and growth media for fungi were purchased from Becton Dickinson (Franklin Lakes, NJ, USA). Resazurin was provided by Sigma Aldrich (Sydney, Australia).

2. 3. 5. Microbial Strains

Table 2. Microbial stains

Escherichia coli	ATCC 25922 (FDA control strain)
Klebsiella pneumoniae	ATCC 700603 (MDR)
Acinetobacter baumannii	ATCC 19606 (type strain)
Pseudomonas aeruginosa	ATCC 27853 (Quality control strain)

Staphylococcus aureus	ATCC 43300 (MRSA)
Candida albicans	ATCC 90028 (CLSI reference)
Candida albicans	ATCC 90028 (CLSI reference)
Cryptococcus neoformans	ATCC 208821 (H99—Type strain)

3. RESULT AND DISCUSSION

3. 1. Synthetic and spectroscopic

The synthetic path of targeted pyran molecule was described in Scheme 1. The targeted molecules KAK-1 to KAK-8 were obtained by reaction between 3-oxo-3-(thiophen-2-yl)propanenitril, aromatic aldehyde and *N*-methyl-1-(methylthio)-2-nitroethenamine in presence of an organic base as a catalyst and ethanol as a solvent.

The structure of the target 4*H*-pyran compounds were characterised by ^{1}H NMR, ^{13}C NMR, IR and Mass spectrometry technique. The two clear peaks were shown in the IR spectra of the compounds at 1560-1515 cm^{-1} and 1385-1345 cm^{-1}, which are confirmed NO_2 group present in compounds. Another one sharp peak is shown at 2200 cm^{-1}, which is indicated for –CN group. In ^{1}H NMR spectra, one sharp signal of one proton shown at ~5.5 δ ppm; which are due to the presence of chiral proton in the synthesized molecule. One broad singlet of one proton signal shown at ~10.5 δ ppm due to the presence of -NH group and signal of three proton of $–CH_3$ was appeared at near about ~3.3 δ ppm. In ^{13}C NMR spectrum has shown the confirmation of carbon atom in the assigned molecular structures of the synthesized compounds.

4-(4-Methoxyphenyl)-6-(methylamino)-5-nitro-2-(thiophen-2-yl)-4*H*-pyran-3-carbonitrile (KAK-1): Isolated yield: 93 % m.p.: 223 ºC; IR (KBr, cm^{-1}): 3209 (N-H, stretching), 2206 (-CN, stretching), 1512 & 1385 (-NO_2, bending), 1257 (C-N, stretching), 1049 (C-O(ether) stretching), 725 (oop, p-substitution); ^{1}H NMR (600 MHz, DMSO-d_6) δ = 3.186 (s, 3H), 3.734 (s, 3H), 4.862 (s, 1H), 6.871 – 6.947 (d, J=8.70, 2H), 7.222 – 7.278 (d, J=8.70, 2H), 7.274 – 7.306 (dd, J=5.01, 3.85, 1H), 7.882 – 7.911 (dd, J=3.85, 1.20, 1H), 7.967 – 7.997 (dd, J=5.02, 1.18, 1H), 10.129 – 10.365 (s, 1H); ^{13}C NMR (151 MHz, DMSO-d_6) δ 14.02, 28.55, 40.05, 55.05, 89.60, 105.84, 114.02, 116.86, 128.55, 128.91, 130.67, 130.95, 131.93, 132.73, 149.90, 156.47, 158.70; MS: *m/z* 369 (M^+).

6-(Methylamino)-5-nitro-2-(thiophen-2-yl)-4-(*p*-tolyl)-4*H*-pyran-3-carbonitrile (KAK-2): Isolated yield: 93 % m.p.: 217 ºC; IR (KBr, cm^{-1}): 3225 (N-H, stretching), 2207 (-CN, stretching), 1512 & 1357 (-NO_2, bending), 1257 (C-N, stretching), 1049 (C-O(ether) stretching), 732 (oop, p-substitution); ^{1}H NMR (400 MHz, $CDCl_3$) δ = 2.339 (s, 3H), 2.858 (s, 3H), 5.063 (s, 1H), 6.947 – 7.084 (m, 2H), 7.113 – 7.197 (dd, J=7.87, 5.68, 1H), 7.303 – 7.385 (m, 2H), 7.686 – 7.759 (dd, J=7.78, 1.56, 1H), 7.877 – 7.945 (dd, J=5.67, 1.47, 1H), 10.185 (s, 1H); ^{13}C NMR (101 MHz, CDCl3) δ 18.97, 27.39, 45.82, 84.26, 114.87, 123.52, 127.92, 128.20, 128.60, 129.63, 137.57, 138.20, 141.49, 144.02, 152.82, 169.43; MS: *m/z* 369 (M^+).

6-(Methylamino)-5-nitro-2-(thiophen-2-yl)-4-(3,4,5-trimethoxyphenyl)-4*H*-pyran-3-carbonitrile (KAK-3): Isolated yield: 95 % m.p.: 257 ºC; IR (KBr, cm-1): 3244 (N-H,

stretching), 2212 (-CN, stretching), 1516 & 1363 (-NO_2, bending), 1240 (C-N, stretching), 1020 (C-O(ether) stretching),850,740 (oop, disubstitution); ^{1}H NMR (400 MHz, $CDCl_3$) δ = 3.299 (s, 3H), 3.816 (s, 3H), 3.846 (s, 6H), 4.940 (s, 1H), 6.519 (s, 2H), 7.108 – 7.237 (m, 1H), 7.539 – 7.712 (d, J=5.09, 1H), 7.938 – 8.100 (d, J=3.96, 1H), 10.041 (s, 1H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 28.52, 41.41, 56.36, 60.82, 90.26, 105.02, 106.58, 116.61, 128.78, 130.62, 130.84, 131.20, 135.09, 138.10, 150.59, 153.61, 157.28; MS: *m/z* 369 (M^+).

4-(3,4-Dimethoxyphenyl)-6-(methylamino)-5-nitro-2-(thiophen-2-yl)-4*H*-pyran-3-carbonitrile (KAK-4): Isolated yield: 94 % m.p.: 251 ºC; IR (KBr, cm-1): 3091 (N-H, stretching), 2212 (-CN, stretching), 1516 & 1396 (-NO_2, bending), 1259 (C-N, stretching), 1145 (C-O(ether) stretching), 740 (oop, 1,2-disubstitution); ^{13}C NMR (101 MHz, $CDCl_3$) δ 27.39, 45.85, 56.04, 84.26, 111.16, 112.81, 114.87, 122.62, 123.52, 127.92, 128.60, 135.62, 137.57, 144.02, 148.64, 148.68, 152.82, 169.43; MS: *m/z* 399 (M^+).

4-(4-Bromophenyl)-6-(methylamino)-5-nitro-2-(thiophen-2-yl)-4*H*-pyran-3-carbonitrile (KAK-5): Isolated yield: 93 % m.p.: 215 ºC; IR (KBr, cm^{-1}): 3217 (N-H, stretching), 2206 (-CN, stretching), 1481 & 1380 (-NO_2, bending), 1257 (C-N, stretching), 1049 (C-O(ether) stretching), 817 (oop, *p*-disubstitution); ^{1}H NMR (600 MHz, $CDCl_3$) δ = 3.789 (s, 3H), 4.945 (s, 1H), 6.845 – 6.895 (m, 2H), 7.171 – 7.201 (dd, J=5.09, 3.95, 1H), 7.228 – 7.260 (d, J=2.08, 2H), 7.586 – 7.616 (dd, J=4.94, 1.11, 1H), 7.967 – 8.031 (dd, J=4.00, 1.11, 1H), 10.027 (s, 1H); MS: *m/z* 417 (M^+).

4-(3-Bromophenyl)-6-(methylamino)-5-nitro-2-(thiophen-2-yl)-4*H*-pyran-3-carbonitrile (KAK-6): Isolated yield: 93 % m.p.: 237 ºC; IR (KBr, cm^{-1}): 3209 (N-H, stretching), 2214 (-CN, stretching), 1556 & 1365 (-NO_2, bending), 1257 (C-N, stretching), 1064 (C-O(ether) stretching), 864 (oop, *m*-disubstitution); ^{13}C NMR (100 MHz,$CDCl_3$) δ 27.39, 45.85, 84.26, 114.87, 121.02, 123.52, 127.20, 127.92, 128.60, 129.33, 129.59, 129.89, 137.57, 144.02, 145.40, 152.82, 169.43; MS: *m/z* 417 (M^+).

4-(2-Chloro-6-methoxyquinolin-3-yl)-6-(methylamino)-5-nitro-2-(thiophen-2-yl)-4*H*-pyran-3-carbonitrile (KAK-7): Isolated yield: 90 % m.p.: 260 ºC; IR (KBr, cm^{-1}): 3495 (N-H, stretching), 2207 (-CN, stretching), 1512 & 1396 (-NO_2, bending), 1257 (C-N, stretching), 1064 (C-O(ether) stretching), 786 (oop, disubstitution); ^{1}H NMR (400 MHz, $CDCl_3$) δ = 3.324 (s, 3H), 3.926 (s, 3H), 5.329 (s, 1H), 7.031 – 7.140 (d, *J*=2.79, 1H), 7.164 – 7.238 (m, 1H), 7.331 – 7.437 (dd, *J*=9.27, 2.78, 1H), 7.586 – 7.702 (d, *J*=5.05, 1H), 7.827 – 7.943 (d, *J*=9.17, 1H), 7.965 – 8.094 (d, *J*=3.95, 1H), 8.192 (s, 1H), 10.200 (s, 1H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 28.56, 41.27, 55.68, 86.95, 104.84, 105.03, 116.16, 124.01, 128.31, 128.81, 129.50, 130.93, 131.00, 131.19, 140.28, 143.38, 145.93, 151.75, 157.57, 158.46; MS: *m/z* 417 (M^+).

4-(2,7-Dichloroquinolin-3-yl)-6-(methylamino)-5-nitro-2-(thiophen-2-yl)-4*H*-pyran-3-carbonitrile (KAK-8): Isolated yield: 93 % m.p.: 265 ºC; IR(KBr, cm^{-1}): 3217 (N-H, stretching), 2206 (-CN, stretching), 1558 & 1366 (-NO_2, bending), 1242 (C-N, stretching), 1064 (C-O(ether) stretching), 850 (oop, substitution); MS: *m/z* 458 (M^+).

3. 2. Biological result

The antimicrobial activity for all the synthesized compounds was performed by CO-ADD (The Community for Antimicrobial Drug Discovery), funded by the Wellcome Trust (UK) and The University of Queensland (Australia)[28]. All twelve newly synthesized target compounds were evaluated for their in *vitro* antibacterial activity against five bacteria (*Escherichia coli*,

Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa and *Staphylococcus aureus*) and two fungi (*Candida albicans* and *Cryptococcus neoformans).* The initial screening results of *in vitro* antimicrobial activity are presented in Table 3.

Table 3. Anti-microbial data of tested compounds.

Name	*% Growth inhibition*							
	Bacteria					Fungi		Conc. (µg/mL)
	Sa	***Ec***	***Kp***	***Pa***	***Ab***	***Ca***	***Cn***	
	G+Ve	G-Ve				Yeast		
KAK-1	20.37	23.73	16.74	1.07	11.4	11.03	-13.09	32
KAK-2	10.88	20.52	18.03	-3.39	16.38	5.89	-24.22	32
KAK-3	13.05	23.32	10.87	-1.32	8.31	7.13	-0.49	32
KAK-4	9.4	20.33	2.51	4.92	-18.84	9.32	24.81	32
KAK-5	18.67	17.24	5.06	3.59	-8.57	4	-7.54	32
KAK-6	16.12	18.14	7.02	4.47	-22.98	7.98	-9	32
KAK-7	18.09	19.34	5.02	6.81	10.29	59.34	-0.43	32
KAK-8	18.64	17.86	2	5.3	12.12	12.16	-32.07	32
Colistin - sulfate	--	0.125	0.25	0.25	0.25	--	--	
Vancomycin - HCl	1	--	--	--	--	--	--	
Fluconazole	--	--	--	--	--	0.125	8	

**Sa = Staphylococcus aureus, Ec = Escherichia coli, Kp = Klebsiella pneumonia, Ab = Acinetobacter baumannii, Pa = Pseudomonas aeruginosa, Ca = Candida albicans, Cn = Cryptococcus neoformans var. Grubii*

Samples with inhibition value above 80% were classed as actives. Samples with inhibition values between 50 and 80% were classed as partial actives while the samples with inhibition values less than 50% were classed as inactive. Among the samples tested, no one has been selected for hit confirmation. The tested compounds displayed more activity towards Gram-negative bacteria than Gram-positive bacteria. Overall, most of the tested compounds exhibit antibacterial activity against all strains and especially against the strain *E. coli*. From these results, we can state that compound **KAK-7** exhibited the highest activity against *Candida albicans.*

4. CONCLUSIONS

In conclusion, we have successfully developed a library of 4H- pyran derivatives. We have synthesized 4*H*-pyran derivatives in excellent yields via Domino one-pot three-component reactions. This reaction presumably occurs via Domino Knoevenagel condensation–Michael addition–intramolecular *O*-cyclization sequence of reactions. The significant advantages of this reaction include one-pot process, simple work-up procedure, excellent yields and no column chromatographic purification. All newly synthesized compounds were evaluated for their antimicrobial activities against a different set of bacteria and fungi. In the present results demonstrated no activity of synthesized pyran as antibacterial as well as antifungal agents.

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References

[1] W. Chen, Y. Cai, X. Fu, X. Liu, L. Lin, X. Feng, *Org. Lett.* 2011, *13*, 4910–4913.

[2] G. Brahmachari, B. Banerjee, *ACS Sustain. Chem. Eng.* 2013, *2*, 411–422.

[3] M. Bihani, P. P. Bora, G. Bez, *J. Chem.* 2012, *2013*.

[4] E. M. Samir, *Open Access Libr. J.* 2016, *3*, 1.

[5] P. Coudert, J. Couquelet, J. Bastide, Y. Marion, J. Fialip, *Ann Pharm Fr.* 1988; 46(2): 91-6.

[6] W. Winter, W.-G. Friebe, A. Roesch, O.-H. Wilhelms, 1983.

[7] P. S. Morahan, J. A. Munson, L. G. Baird, A. M. Kaplan, W. Regelson, *Cancer Res.* 1974, *34*, 506–511.

[8] A. M. Kaplan, P. S. Morahan, W. Regelson, *J. Natl. Cancer Inst.* 1974, *52*, 1919–1923.

[9] S. J. Mohr, M. A. Chirigos, F. S. Fuhrman, J. W. Pryor, *Cancer Res.* 1975, *35*, 3750–3754.

[10] D. Kumar, V. B. Reddy, S. Sharad, U. Dube, S. Kapur, *Eur. J. Med. Chem.* 2009, *44*, 3805–3809.

[11] M. Ghorab, A. Hassan, *Phosphorus Sulfur Silicon Relat. Elem.* 1998, *141*, 251–261.

[12] H. M. Faidallah, K. A. Khan, A. M. Asiri, *Eur. J. Chem.* 2011, *2*, 243–250.

[13] A. H. Bedair, H. A. Emam, N. A. El-Hady, K. A. Ahmed, A. M. El-Agrody, *Il Farm.* 2001, *56*, 965–973.

[14] S. S. Mansoor, K. Aswin, K. Logaiya, S. Sudhan, H. Ramadoss, *J. Saudi Chem. Soc.* 2016, *20*, S393–S400.

[15] P. K. Paliwal, S. R. Jetti, S. Jain, *Med. Chem. Res.* 2013, *22*, 2984–2990.

[16] S. Hanessian, J. Szychowski, J. P. Maianti, *Org. Lett.* 2008, *11*, 429–432.

[17] A. Evidente, A. Cabras, L. Maddau, S. Serra, A. Andolfi, A. Motta, *J. Agric. Food Chem.* 2003, *51*, 6957–6960.

[18] M. B. Rubio, R. Hermosa, J. L. Reino, I. G. Collado, E. Monte, *Fungal Genet. Biol.* 2009, *46*, 17–27.

[19] B. Das, K. Laxminarayana, M. Krishnaiah, D. N. Kumar, *Bioorg. Med. Chem. Lett.* 2009, *19*, 6396–6398.

[20] Y. Dong, K. Nakagawa-Goto, C.-Y. Lai, S. L. Morris-Natschke, K. F. Bastow, K.-H. Lee, *Bioorg. Med. Chem. Lett.* 2011, *21*, 2341–2344.

[21] A. Nakhi, R. Adepu, D. Rambabu, R. Kishore, G. Vanaja, A. M. Kalle, M. Pal, *Bioorg. Med. Chem. Lett.* 2012, *22*, 4418–4427.

[22] X. Fan, D. Feng, Y. Qu, X. Zhang, J. Wang, P. M. Loiseau, G. Andrei, R. Snoeck, E. De Clercq, *Bioorg. Med. Chem. Lett.* 2010, *20*, 809–813.

[23] C. B. Bridges, K. Fukuda, T. M. Uyeki, N. J. Cox, J. A. Singleton, *MMWR Recomm. Rep. Morb. Mortal. Wkly. Rep. Recomm. Rep.* 2002, *51*, 1–31.

[24] R. S. McCord, M. K. Breinig, P. S. Morahan, *Antimicrob. Agents Chemother.* 1976, *10*, 28–33.

[25] M. D. Aytemir, Ü. Çaliş, M. Oezalp, *Arch. Pharm. Int. J. Pharm. Med. Chem.* 2004, *337*, 281–288.

[26] W. N. Chan, J. M. Evans, M. S. Hadley, H. J. Herdon, J. C. Jerman, H. K. Morgan, T. O. Stean, M. Thompson, N. Upton, A. K. Vong, *J. Med. Chem.* 1996, *39*, 4537–4539.

[27] E. A. A. Hafez, M. H. Elnagdi, A. A. Elagamey, F. M. A. A. El-Taweel, *ChemInform* 1987, *18*, no-no.

[28] M. A. T. Blaskovich, J. Zuegg, A. G. Elliott, M. A. Cooper, *ACS Infect. Dis.* 2016, *1*, 285–287.