European Journal of Chemistry 6 (2) (2015) 157-162

European Journal of Chemistry

Journal webpage: www.eurjchem.com

Design, synthesis, antimicrobial activity and anticancer screening of some new 1,3-thiazolidin-4-ones derivatives

Wesam Saber Shehab [1,*] and Samar Mohamed Mouneir [2]

[1] *Department of Chemistry, Faculty of Science, Zagazig University, Zagazig, 44511, Egypt*
[2] *Department of Pharmacology, Faculty of Veterinary Medicine, Cairo University, Cairo, 12211, Egypt*

** Corresponding author at: Department of Chemistry, Faculty of Science, Zagazig University, Zagazig, 44511, Egypt.*
Tel.: +2.01023599259. Fax: +2.055.2285374. E-mail address: dr_wesam123@yahoo.com (W.S. Shehab).

ARTICLE INFORMATION

DOI: 10.5155/eurjchem.6.2.157-162.1219

Received: 17 November 2014
Received in revised form: 11 January 2015
Accepted: 01 February 2015
Published online: 30 June 2015
Printed: 30 June 2015

KEYWORDS

Anticancer
Schiff's bases
3-Acetylindol
Antimicrobial
1,3-Thiazolidin-4-ones
Thiazolopyridine derivatives

ABSTRACT

A series of new thiazolidin-4-ones have been synthesized by the reaction of 3-acetylindole with thiourea to yield 2-amino-arylthiazole (1) which, reacted with 2-chloroacetyl chloride to produce 2-chloroacetamido-4-arylthiazoles (2). The later was treated with potassium thiocyanate to afford the related 2-amino-3-(4-arylthiazol-2-yl) thiazolidin-4-ones (3). Condensation of compounds 1 and 3 with different aromatic aldehydes give Schiff's bases (4a-c) and (5a-c) reaction of compound 5a-c with thioglycollic acids furnishes the target thiazolidin-4-one molecules (6a-c). Further, condensation of compound 6a with benzaldehyde affords benzylidenethiazolo derivative (7) which on refluxing with malononitrile, acetylacetone afforded thiazolopyridine derivatives (8,9). Structure elucidation of the products has been accomplished on the basis of elemental analysis, IR, ^{1}H NMR data. Compound 3exhibited the most potent antibacterial and anticancer activity.

Cite this: *Eur. J. Chem.* **2015**, *6(2)*, 157-162

1. Introduction

Thiazolidinones are derivatives of thiazolidine and they also constitute an important group of heterocyclic compounds [1]. Thiazolidin-4-one derivatives exhibit various biological activities such as anti-microbial [2], anti-inflammatory [3,4], antihistaminic [5], anti-hypertensive, analgesic [6] and anti-bacterial activities [7-9].

Thiazolidinones, with a carbonyl group in position 2, 4- or just 4-, have been extensively studied [10,11] and literature surveys showed that thiazolidin-4-ones are important compounds due to their broad range of biological activities [12-18]. 2-Substituted 4-thiazolidinones derivatives exhibit unusually high activity against mycobacterium tuberculosis when tested in vitro [19]. Overviews of their synthesis, properties, reactions and applications have been published [10,11].

Indoles have been reported to possess a wide variety of biological activities like anti-inflammatory [20], anticancer [21], antifungal [22] and were used in the treatment of gastrointestinal, cardiovascular and central nervous system (CNS) disorders, HIV-1 integrase inhibitors for antitumor activity, inhibitors of hepatitis, as antibacterial and as anti-malarial agents [23-28]. Therefore, the aim of the present work was to prepare thiazolidin-4-one-derivatives using 3-acetylindol in order to find out new biologically active compounds.

2. Experimental

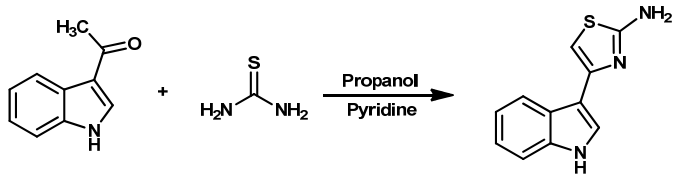
2.1. Instrumentation

All melting points are uncorrected and were determined on GallenKamp electric melting point apparatus. IR spectra (KBr discs) were recorded on of FT/IR-400 spectrophotometer (Perkin Elmer). ^{1}HNMR spectra were recorded on ovarian 300 MHz (DMSO-d_6) solutions. Chemical shifts were reported as δ values relative to tetramethylsilan (TMS) as internal reference. The analyses were carried out at Micro-Analytical Center, Cairo University.

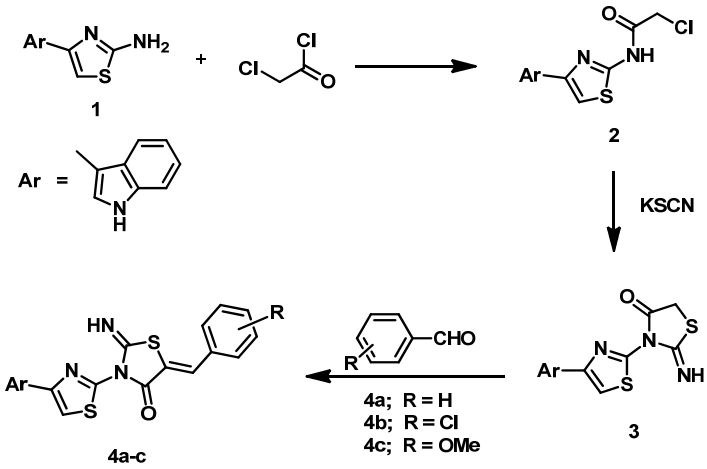
2.2. Synthesis of 4-(1H-indol-3-yl) thiazol-2-amine (1)

3-Acetylindole (0.05 mole) and thiourea (0.05 mole) were taken in round bottom flask and dissolved in propanol (35 mL) and refluxed for 2 h. The solid obtained was triturated with ethanol to remove unreacted acetylindole. To this pyridine (5 mL) was added continued refluxed for 5 h.

ISSN 2153-2249 (Print) / ISSN 2153-2257 (Online)
http://dx.doi.org/10.5155/eurjchem.6.2.157-162.1219



Scheme 1



Scheme 2

The reaction completion was monitored by TLC. The solid separated is filtered recrystallized from ethanol, **1** (Scheme 1). Color: Pale yellow. Yield: 69%. M.p.: 158-160 °C. FT-IR (KBr, ν, cm^{-1}): 3340 (N-H), 3012 (Ar-CH), 1582 (C=N), 1457 (C=C). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 4.32 (s, 2H, NH_2), 7.08 (s, 1H, thiazole-H), 7.10-7.60 (m, 5H, Ar-H), 14.01 (s, 1H, NH). Anal. calcd. for $C_{11}H_9N_3S$: C, 61.37; H, 4.21; N, 19.52. Found: C, 61.31; H, 4.20; N, 19.49%.

2.3. Synthesis of N-(4-(1H-indol-3-yl)thiazol-2-yl)-2-chloro acetamide (2)

In conical flask, 0.01 mole of 2-amino-4-aryl thiazole (**1**) in 25 mL benzene was stirred for 30 min in ice-bath till the temperature becomes below 0-5 °C then add 0.01 mole chloroacetyle chloride was add drop by drop in conical flask within 2h. After complete addition the reaction mixture was refluxed it for2h in water bath then the solvent was evaporates. The product that separated was recrystallization from ethanol (Scheme 2). Color: Pale yellow. Yield: 82%. M.p.: 145-147 °C. FT-IR (KBr, ν, cm^{-1}): 3160 (NH), 3012 (Ar-H),1633 (C=O). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 4.22 (s, 2H, $COCH_2$), 7.14-7.55 (m, 6H, Ar-H), 14.00 (s, 1H, NH). Anal. calcd. for $C_{13}H_{10}ClN_3OS$: C, 53.52; H, 3.45; N, 14.40. Found: C, 53.50; H, 3.41; N, 14.39%.

2.4. Synthesis of 3-(4-(1H-indol-3-yl) thiazol-2-yl)-2-imino thiazolidin-4-one (3)

A mixture of 2-chloro acetamido-4-aryl thiazole (**2**) (0.03 mole), KSCN (0.06 mole) in dry acetone (100 mL) was refluxed for 3 h. The reaction mixture was kept for evaporated under vacuum to obtain crude product. The residue was stirred with water. The solid product was filtered, washed with water, dried and recrystallized with ethanol (Scheme 2). Color: Pale brown. Yield: 93%. M.p.: 162-164 °C. FT-IR (KBr, ν, cm^{-1}): 3497 (=NH), 3197 (Ar-H), 1671 (C=O), 1602 (C=N), 705 (C-S). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 3.80 (s, 1H, =NH), 4.10 (s, 2H, CH_2), 7.12-8.40 (m, 6H, Ar-H), 11.9 (bs, 1H, NH). Anal. calcd. for $C_{14}H_{10}N_4OS_2$: C, 53.49; H, 3.21; N, 17.82. Found: C, 53.45; H, 3.19; N, 17.80%.

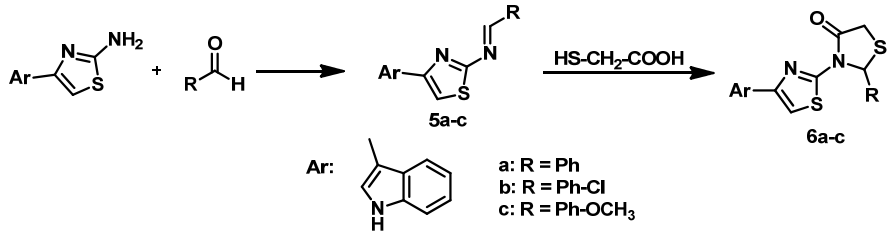
2.5. General procedure for preparation 3-(4-(1-indol-3-yl) thiazol-2-yl)-5-benzylidene-2-iminothiazolidine-4-one (4a-c)

A mixture iminothiazolidin-4-one (**3**) (1 mmol), aldehyde and sodium acetate (1.5 mmol) in glacial acetic acid was refluxed for 2 to 4 h. Till completion of the reaction (TLC check), the reaction mixture was poured onto ice-cold water the solid thus separated was filtered and the crude product was recrystallized using absolute ethanol to get compounds **4a-c** (Scheme 2).

3-(4-(1H-Indol-3-yl) thiazol-2-yl)-5-benzylidene-2-iminothia zolidin-4-one (**4a**): Color: Brown. Yield: 94%. M.p.: 220-223 °C. FT-IR (KBr, ν, cm^{-1}): 3488 (NH), 1700 (C=O), 1640 (C=CH-), 1558 (C=N). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 3.9 (s, 1H, =NH), 7.24-7.37 (d, 1H, -C=CH), 7.38-8.19 (m, 11H, Ar-H), 10.16 (br, 1H, exchangeable with D_2O). Anal. calcd. for $C_{21}H_{14}N_4OS_2$: C, 62.67; H, 3.51; N, 13.92. Found: C, 62.64; H, 3.42; N, 13.80%.

3-(4-(1H-Indol-3-yl)thiazol-2-yl)-2-imino-5-(4-methoxy benzylidene)thiazolidin-4-one (**4b**): Color: Brown. Yield: 96%. M.p.: 210-212 °C. FT-IR (KBr, ν, cm^{-1}): 3497 (NH), 1670 (C=O), 1660 (-C=CH-), 1570 (C=N). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 3.8 (s, 3H, $-CH_3$), 7.24-7.27 (d, 1H, -C=CH-), 7.38-8.41 (m, 10H, Ar-H), 13.90 (s, 1H, NH).

3-(4-(1H-indol-3-yl)thiazol-2-yl)-5-(4-chlorobenzylidene)-2-iminothiazolidin-4-one (**4c**): Color: Pale yellow. Yield: 94%. M.p.: 200-204 °C. FT-IR (KBr, ν, cm^{-1}): 3460 (NH), 1680 (C=O), 1560 (-C=CH-). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 3.9 (s, 1H, =NH), 7.24-7.37 (d, 1H, -C=CH-), 7.38-8.41 (m, 10H, Ar-H), 14.02 (s, 1H, NH).



Scheme 3

2.6. General procedure for the preparation of N-(substitute benzylidene)-4-(1H-indol-3-yl) thiazol-2-amine (5a-c)

To an equimolar methanolic solution of 2-amino-4-aryl thiazole (0.1 mol) and substituted benzaldehyde (0.1 mol), a few drops of glacial acetic acid were added. The mixture was then refluxed on water bath 5-6 h. It was then allowed to cool, poured into crushed ice and recrystallized from methanol (Scheme 3).

N-(Benzylidene)-4-(1H-indol-3-yl) thiazol-2-amine (**5a**): Color: Yellow. Yield: 82%. M.p.: 166-168 °C. FT-IR (KBr, ν, cm^{-1}): 3400 (N-H), 1620 (-N=CH-). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 7.0-7.6 (m, 11H, Ar-H), 8.1 (s, 1H, -N=CH), 10.1 (s, 1H, NH). Anal. calcd. for $C_{18}H_{13}N_3S$ (303.08): C, 71.26; H, 4.32; N, 13.85. Found: C, 71.22; H, 4.31; N, 13.82%.

N-(4-Chlorobenzylidene)-4-(1H-indol-3-yl)thiazol-2-amine (**5b**): Color: Pale yellow. Yield: 85%. M.p.: 160-165 °C. FT-IR (KBr, ν, cm^{-1}): 3310 (N-H), 1600 (-N=H). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 7.12-7.68 (m, 10H, Ar-H), 8.20 (s, 1H, -N=CH), 10.10 (s, 1H, NH). Anal. calcd. for $C_{18}H_{12}ClN_3S$: C, 64.00; H, 3.58; N,12.44. Found: C, 64.12; H, 3.52; N, 12.39%.

N-(4-Methoxybenzylidene)-4-(1H-indol-3-yl)thiazol-2-amin (**5c**): Color: Yellow. Yield: 84%. M.p.: 170-172 °C. FT-IR (KBr, ν, cm^{-1}): 3390 (NH), 2920 (Ar-CH), 2840 (Aliph-CH), 1600 (-N=CH). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 3.53 (s, 3H, $-OCH_3$), 6.87-7.61 (m, 10H, Ar-H), 8.01 (s, 1H, N=CH), 10.10 (s, 1H, NH). Anal. calcd. for $C_{19}H_{15}N_3OS$ (333.41): C, 68.45; H, 4.53; N, 12.60. Found: C, 64.42; H, 4.52; N, 12.59%.

2.7. General procedure for the preparation of the 3-(4-1H-indol-3-yl)thiazol-2-yl)-2-(4-substituted phenyl)thiazolidin-4-one (6a-c)

A mixture of Schiff's base (**5a-c**) (0.01 mol) and thioglycolic acid (0.01 mol) was refluxed in dimethyl formamide (15 mL) for 6 h. The reaction mixture was cooled and poured into crushed ice. The solid obtained was filtered and recrystallized from ethanol to give compounds **6a-c** (Scheme 3).

3-(4-(1H-Indol-3-yl)thiazol-2-yl)-2-phenyl thiazolidin-4-one (**6a**): Color: Pale brown. Yield: 82%. M.p.: 200-204 °C. FT-IR (KBr, ν, cm^{-1}): 3225 (NH), 1700 (C=O), 1631(C=N), 1583 (C=C). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 4.00 (s, 2H, $COCH_2S$), 5.9 (s, 1H, -NCHS), 6.6-7.6 (m, 11H, Ar-H), 10.1 (s, 1H, NH). Anal. calcd. for $C_{20}H_{15}N_3OS_2$: C, 63.64; H, 4.01; N, 11.13. Found: C, 63.62; H, 4.00; N, 11.10%.

3-(4-(1H-Indol-3-yl)thiazol-2-yl)-2-(4-chlorophenyl)thiazolidin-4-one (**6b**): Color: Pale brown. Yield: 84%. M.p.: 210-212 °C. FT-IR (KBr, ν, cm^{-1}): 3300 (NH), 1700 (C=O), 1600 (-N=C-). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 3.83 (s, 2H, $COCH_2S$), 5.8 (s, 1H, -NCHS), 7.1-7.8 (m, 10H, Ar-H), 10.1 (s, 1H, NH). Anal. calcd. for $C_{20}H_{14}ClN_3OS_2$: C, 58.31; H, 3.43; N, 10.20. Found: C, 58.29; H, 3.40; N, 10.19%.

3-(4-(1H-Indol-3-yl)thiazol-2-yl)-2-(4-methoxyphenyl)thiazolidin-4-one (**6c**): Color: Brown. Yield: 84%. M.p.: 205-210 °C. FT-IR (KBr, ν, cm^{-1}): 3290 (NH), 2920 (Ar-CH), 2840 (Aliph-CH), 1690 (C=O), 1600 (-N=C). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 3.4 (s, 2H, $COCH_2S$), 3.70 (s, 3H, $-OCH_3$), 5.90 (s, 1H, NCHS), 6.60-7.90 (m, 10H, Ar-H), 10.10 (s, 1H, NH). Anal. calcd. for $C_{21}H_{17}N_3O_2S_2$: C, 61.89; H, 4.20; N, 10.31. Found: C, 61.88; H, 4.19; N, 10.30%.

2.8. Synthesis of 3-(4-(1H-Indol-3-yl)thiazol-2yl)-5-benzylidiene-2-phenylthiazolidin-4-one (7)

A mixture of compound **6a** (0.01 mole) and benzaldehyde (0.01 mole) was refluxed in absolute ethanol (30 mL) and catalyzed with few drops of TEA for 5 h. After cooling the obtained solid was filtered, washed, dried and recrystallized from ethanol (Scheme 4). Color: Brown. Yield: 89%. M.p.: 220-222 °C. FT-IR (KBr, ν, cm^{-1}): 3420 (NH), 3040 (Ar-CH), 1700 (C=O), 1600 (C=C). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 5.90 (s, 1H, NCHS), 6.10 (s, 1H, -CH-), 7.10-7.90 (m, 16H, Ar-H), 10.40 (s, 1H, NH). Anal. calcd. for $C_{27}H_{19}N_3OS_2$: C, 69.65; H, 4.11; N, 9.03. Found: C, 69.64; H, 4.10; N, 9.04 %.

2.9. Synthesis of 3-(4-(1H-indol-3-yl)thiazol-2-yl)-5-amino-2,3-dihydro-2,7-diphenylthiazolo[4,5-b]pyridine-6-carbonitrile (8)

A mixture of compound **7** (0.01 mole), malononitrile (0.01 mole) and ammonium acetate (1 g) in 30 mL acetic acid was refluxed for 3 h. The solid formed upon cooling was collected by filtration, washed with water and recrystallized from ethanol (Scheme 4). Color: Pale orange. Yield: 72%. M.p.: 256-258 °C. FT-IR (KBr, ν, cm^{-1}): 3410, 3285 (NH, NH_2), 3040 (Ar-CH),2200 (CN), 1600 (C=C). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 4.00 (s, 2H, NH_2), 4.90 (s, 1H, -NCHS), 7.00-7.90 (m, 16H, Ar-H), 10.01 (s, 1H, NH). Anal. calcd. for $C_{30}H_{20}N_6S_2$: C, 68.16; H, 3.81; N, 15.90. Found: C, 68.14; H, 3.80; N, 15.89%.

2.10. Synthesis of 1-(3-(4-(1H-indol-3-yl)thiazol-2-yl)-2,3-dihydro-5-methyl-2,7-Diphenylthiazolo[4,5-b]pyridine-6-yl)ethanone (9)

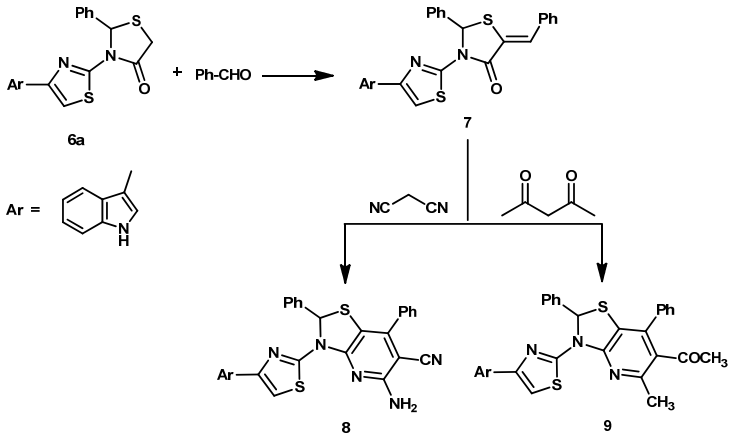
A mixture of compound **7** (0.01 mole), acetylacetone (0.01 mole) and ammonium acetate (1 g) in 30 mL acetic acid was refluxed for 3 h. The solid formed upon cooling was collected by filtration, washed with water and recrystallized from ethanol (Scheme 4). Color: Pale brown. Yield: 64%. M.p.: 200-204 °C. FT-IR (KBr, ν, cm^{-1}): 3410 (NH), 3040 (Ar-CH), 2840 (Aliph-CH),1660 (CO), 1595 (C=C). ^{1}H NMR (300 MHz, DMSO-d_6, δ, ppm): 3.20 (s, 3H, $COCH_3$), 3.80 (s, 3H, -N=C-CH_3), 4.95 (s, 1H, -NCHS), 6.90-7.90 (m, 16H, Ar-H), 10.20 (s, 1H, NH). Anal. calcd. for $C_{32}H_{24}N_4OS_2$: C, 70.56; H, 4.44; N, 10.29. Found: C, 70.50; H, 4.41; N, 10.27%.

2.11. Antimicrobial activity

Antimicrobial activity of the tested compounds was determined using a modified Kirby-Bauer disc diffusion method [29]. Briefly, 100 µL of the test bacteria/fungi were grown in 10 mL of fresh media until they reached a count of approximately108 cells/mL for bacteria 105 cells/mL for fungi [30].

Table 1. *In-vitro* antibacterial and antifungal screening of the newly synthesized compounds.

Sample	Inhibition zone diameter (mm/mg sample)			
	Escherichia coli (G^-)	*Staphylococcus aureus* (G^+)	*Aspergillus flavus* (Fungus)	*Candida albicans* (Fungus)
1	10	9	0.0	0.0
2	9	0.0	0.0	0.0
3	10	11	0.0	0.0
6_a	10	9	0.0	0.0
6_b	12	16	0.0	0.0
6_c	10	10	0.0	0.0
DMSO	0.0	0.0	0.0	0.0
Ampicillin	22	18	0.0	0.0
Amphotericin B	0.0	0.0	17	19



Scheme 4

100 µL of microbial suspension was spread onto agar plates corresponding to the broth in which they were maintained. Isolated colonies of each organism that might be playing a pathogenic role should be selected from primary agar plates and tested for susceptibility by disc diffusion method [31-33]. Of the many media available, National Committee for Clinical Laboratory Standards (NCCLS) recommends Mueller-Hinton agar due to itsgood results in batch-to-batch reproducibility. Disc diffusion method for filamentous fungi tested by using approved standard method (M38-A) [34] for evaluating the susceptibilities of filamentous fungi to antifungal agents. Disc method for yeasts developed by using approved standard method (M44-P) [35]. Plates inoculated with filamentous fungi as *Aspergillus flavus* at 25 °C for 48 h; Gram (+) bacteria as *Staphylococcus aureus*; Gram (-) bacteria as *Escherichia coli*, they were incubated at 35-37 °C for 24-48 h and yeast as *Candida albicans* incubated at 3 °C for 24-48 h and, then the diameters of the inhibition zones were measured in millimeters [27]. Standard discs of Ampicillin (Antibacterial agent), Amphotericin B (Antifungal agent) served as positive controls for antimicrobial activity but filter discs impregnated with 10 µL of solvent (Distilled water, chloroform, DMSO) were used as a negative control. The agar used is Meuller-Hinton agar that is rigorously tested for composition and pH. Furthermore, the depth of the agar in the plate is a factor to be considered in the disc diffusion method. This method is well documented and standard. A zone of inhibition has been determined for susceptible and resistant values. Blank paper disks (Schleicher & Schuell, Spain) with a diameter of 8.0 mm were impregnated with 10 µL of tested concentration of the stock solutions. When a filter paper disc impregnated with a tested chemical is placed on agar the chemical will diffuse from the disc into the agar. This diffusion will place the chemical in the agar only around the disc. The solubility of the chemical and its molecular size will determine the size of the area of chemical infiltration around the disc. If an organism is placed on the agar it will not grow in the area around the disc if it is susceptible to the chemical. This area of no growth around the disc is known as a "*Zone of inhibition*" or" *Clear zone*". For the disc diffusion, the zone diameters were measured with slipping calipers of the National Committee for Clinical Laboratory Standards [31]. Agar-based methods such as test and disk diffusion can be good alternatives because they are simpler and faster than broth-based methods [35-37] (Table 1).

2.12. Anticancer activity

2.12.1. Cell culture

The cells were obtained from Egyptian Holding Company for Biological Products & Vaccines (VACSERA), Giza, Egypt and then maintained in the tissue culture unit .The cells were grown in RBMI-1640 medium, supplemented with 10% heat inactivated FBS, 50 units/mL of penicillin and 50 mg/mL of streptomycin and maintained at [37] in a humidified atmosphere containing 5% CO_2. The cells were maintained as monolayer culture by serial sub-culturing. Cell culture reagents were obtained from Lonza (Basel, Switzerland). The anticancer activity of the rested compounds was evaluated against MCF-7 cells (Breast cancer), HEPG-2 cells (Liver cancer) and HCT 116 (Colon cancer) (Table 2).

2.12.2. The sulforhodamine B (SRB) cytotoxicity assay

Cytotoxicity was determined using the sulforhodamine B (SRB) assay method as previously described by Skehan *et al.* [35]. Exponentially growing cells were collected using 0.25% Trypsin-EDTA and seeded in 96-well plates at 1000-2000 cells/well in RBMI-1640 supplemented medium. After 24 h, cells were incubated for 72 h with various concentrations of the tested compounds. Following 72 h treatments, the cells would be fixed with 10% trichloroacetic acid for 1 h at 4 °C.

Table 2. *In-vitro* anticancer screening of the newly synthesized compounds against different cell lines.

Compound	IC_{50} (µM)		
	MCF-7 (Breast cancer)	HEPG-2 (Liver cancer)	HCT 116 (Colon cancer)
1	20.35	16.54	14.91
2	73.87	50.60	45.48
3	19.23	12.11	12.42
6_a	26.74	14.15	16.44

Wells were stained for 10 minutes at room temperature with 0.4% SRBC (Sulphorhodamine B) dissolved in 1% acetic acid. The plates were air dried for 24 h and the dye was solubilized with Tris-HCl for 5 min on a shaker at 1600 rpm. The optical density (OD) of each well was measured spectrophotometrically at 564 nm with an ELISA microplate reader (ChroMate-4300, FL, USA). The IC_{50} values were calculated according to the equation for Boltzman sigmoidal concentration response curve using the nonlinear regression fitting models (Graph Pad, Prism Version 5)

3. Results and discussion

3.1. Synthesis

Our synthetic strategy for 1, 3-thiazolidin-4-one derivatives starts with refluxing of 3- acetyl indol and thiourea in propanol to afforded 4(1*H*-indol-3-yl) thiazol-2-amine (**1**) which formed by the attack of sulphur nucleophile on imine carbon followed by intermolecular cyclization on elimination of water, Scheme 1.

Chloroacetamide (**2**) was obtained by reacting of 2-amino thiazoles (**1**) with chloroacetyl chloride in presence of pyridine. The IR of compound **2** showed the presence of bands characteristic for C=N at 1631-1642 cm^{-1} and an amide function at 1661 (C=O) and 3160-3184 cm^{-1} (NH). The ^1H NMR of compound **2** revealed a broad singlet at δ 10.32 ppm characteristic for a secondary amine group, a multiplet at δ 7.14-7.55 ppm for aromatic protons and $COCH_2$ as a singlet at δ 4.22 ppm.

When chloroacetamide (**2**) was refluxed with potassium thiocyanate in dry acetone, 2-iminothiazolidine-4-one (**3**) was obtained in a moderate to good yields. The structures of the isolated compounds were determined by spectral methods. The IR of compound **3** revealed characteristic bands for C=N at 1602cm^{-1}, C=O at 1671cm^{-1}, primary and secondary amines at 3224-3278$cm^{-1}$and 3166-3178 cm^{-1}. The ^1H NMR spectra showed the presence of broad exchangeable singlets at δ 11.90 ppm NH protons, while O=C-CH_2 protons appears at δ 4.1, 4.3 ppm for C=NH and the aromatic protons of compound **3**appear as a multiplet at δ 7.12-8.57 ppm.

The iminothiazolidine-4-one (**3**) on Knoevenagel condensation with different substituted aryl aldehydes in presence of sodium acetate as a base in glacial acetic acid afforded the 3-(4-(1-indol-3-yl)thiazol-2-yl)- 5-benzylidene-2-iminothiazolidine-4-one (**4a-c**) which established on the basis of IR, ^1H NMR spectra did not only show the absence of CH_2 protons at δ 4.1 ppm, but also the presence of HC=C proton at δ 7.24-7.37 ppm, Scheme 2.

New compounds (**5a-c**)were prepared by refluxing 2-amino-4-aryl thiazole (**1**) and substituted benzaldehyde, a few drops of glacial acetic acid in methanol for 5-6 h. These Schiff's base (**5a-c**) and thioglycolic acid (0.01 mol) when refluxed in dimethyl formamide (15 mL) for 6 h afforded the 3-(4-1*H*-indol-3-yl)thiazol-2-yl)-2-(4-substituted phenyl)thiazolidin-4-one (**6a-c**) in good yield which determined by IR and ^1H NMR spectra. IR spectra showed the characteristic bands for C=N at 1630-1600 cm^{-1} and amide function at 1690-1700 (C=O) and 3255-3300 cm^{-1} (NH). The ^1H NMR of showed the presence of one broad exchangeable singlet at δ 10.1 ppm (NH), a multiplet at δ 7.1-7.8 ppm (Ar-H's), a singlet at δ 5.8 ppm (thiazolyl-C_5-H), and a singlet at δ 3.83 ppm (O=C-CH_2), Scheme 3.

Condensation of compound (**6a**) with benzaldehyde affords 5-benzylidenethiazolo derivatives (**7**), later, refluxing of compound **7** with malononitrile in the presence of ammonium acetate resulted in cycloaddition affording thiazolopyridine derivative (**8**) which showed IR spectra at 3410, 3285 (NH, NH_2) and 2200 cm^{-1} (CN). The ^1H NMR spectra showed the presence of broad exchangeable singlets at δ 10.01 ppm NH protons, while NCHS protons appears at δ 4.9 ppm and the aromatic protons appears as a multiplet at δ 7.00-7.90 ppm.

Also, thiazolopyridine derivative (**9**) was obtained by using acetylacetonein the same manner the structure was confirmed by ^1H NMR spectra showed signal at δ 3.20 ppm for OCH_3, δ 3.80 ppm for -N=C-CH_3, δ 4.95 ppm -NCHS and singlets at δ 10.20 ppm for NH. The structure of all newly isolated compounds was fully confirmed by spectral and elemental analyses methods, Scheme 4.

3.2. Antibacterial activity

All the tested compounds have antibacterial activity against Gram negative (*Escherichia coli*) and Gram positive (*Staphylococcus aureus*), bacteria except compound **2** which had antibacterial activity against Gram negative bacteria only. Compound **6b** showed the highest antibacterial activity. All the tested compounds had no antifungal activity against *Aspergillus flavus* and *Candida albicans*. The antibacterial activity of the newly synthesized compounds is due to the presence of thiazolidine ring [6,7].

All the synthesized compounds were tested for in vitro anticancer activity using SRB cytotoxicity assay method. The anticancer screening of the tested compounds revealed that all the synthesized compounds exhibited a significant cytotoxic activity against MCF-7 Breast cancer, HEPG-2 Liver cancer and HCT116 Colon cancer cell lines in variable degrees. The anticancer activity of these newly synthesized compounds may be due to the presence of indoles. Previous studies revealed that indoles possess a high anticancer activity against different cell lines [19].

From Table 2, we can notice that compound **3** exhibited the most potent anticancer activity against MCF-7 Breast cancer, HEPG-2 Liver cancer and HCT116 Colon cancer cell lines by IC_{50} 19.23, 12.11 and 12.42 µM, respectively, while compound **2** showed the least anticancer activity.

4. Conclusion

Our present investigation is centered on the studies of synthesis, reactions, spectral analysis and biological activities of 1,3-thiazolidin-4-ones derivatives. The procedure proved more beneficial than those previously reported in the literature. Compound **3** exhibited the most potent antibacterial and anticancer activity. All the compounds had no antifungal activity against *Aspergillus flavus* and *Candida albicans*.

References

[1]. Agarwal, O. P. Textbook of Organic Chemistry, 4th edition, Goel Publisher, p730, 2006.
[2]. Bama, K. G.; Rajani, K. B. *Indian J. Chem. B.* **1988**, *27*, 1157-1158.

[3]. Mahd, A. K.; Zaman, M. S. *Indian J. Chem. B*. **2004**, *43*, 2189-2194.
[4]. Verma, L.; Wakode, S. *Pharma Innov. J.* **2013**, *2,* 41-50.
[5]. Raghuram, R. A.; Rajan, K. S.; Devsingh, S.; Bhagavan, R. M. *Indian J. Chem. B* **2001**, *40*, 813-816 .
[6]. Reddy, P. S. N.; Manmahan, R.; Pradap, R. P. *Indian J. Chem. B* **2003**, *40,* 2119-2120.
[7]. Mulwad, W.; Choudhari, B. *Indian. J. Chem. B* **2005**, *42,* 1074-1078.
[8]. Mhogale, V. A. *Indian J. Chem. Sci.* **1990**, *102(4),* 535-540.
[9]. Sharshira, E. M.; Hamada, N. M. M. *Am. J. Org. Chem.* **2012**, *2(3)*, 69-73.
[10]. Brown, F. C. *Chem. Rev*. **1961**, *61,* 463-463.
[11]. Singh, S. P.; Parmar, S. S.; Raman, K.; Stenberg, V. I. *Chjem. Rev.* **1981**, *61,* 175-203.
[12]. Doran, W. J.; Shonle, H. A. *J. Org. Chem*. **1938**, *3,* 193-197.
[13]. Troutman, H. D.; Long, L. M. *J. Am. Chem. Soc.* **1948**, *70,* 3436-3439.
[14]. Rout, M. K.; Mahapatra; G. N. *J. Am. Chem. Soc.* **1955**, *77,* 2427-2428.
[15]. Gaikwad, N. J.; Trupude, R. N. *Indian Drugs* **1994**, *31,* 593-594.
[16]. El-Gendy, Z.; Abdel-Rahman, R. M.; Fawzy, M. M; Mahmoud, M. B. *J. Ind. Chem. Soc.* **1994**, *67,* 927-929.
[17]. Diumo, M. V.; Mazzoni, O.; Piscopo, E.; Calignano, A.; Giordano, F.; Bolognese, A. *J. Med. Chem.* **1992**, *35,* 2190-2912.
[18]. Shah, V.; Pant, C. K.; Joshi, P. C. *Asian J. Chem*. **1993**, *5*, 83-88.
[19]. Mukhatar, S.; Majeebar, R. V. P.; Ansari, W. H.; Lemiere, G.; De Groot, A.; Dommisse, R. *J. Ind. Chem. Soc*. **1999**, *4,* 232-237.
[20]. Radwan, M. A. A.; Ragab, E. A.; Nermien, M. *Bioorg. Med. Chem*. **2007**, *15,* 3832-3841.
[21]. Singh, P.; Mittal, A.; Kaur, S. *Bioorg. Med. Chem. Lett*. **2008**, *18,* 85-89.
[22]. Ryu, C. K. K.; Lee, J. Y.; Park, R.; Ma, N. *Bioorg. Med. Chem. Lett.* **2007**, *5,* 127-31.
[23]. Liu, K.; Sun, J.; Lou, H.; Dai, S. , Xu, H. *Phytochem.* **2008**, *69*, 1405-1410.
[24]. Abdel-Rahman, A. A. H.; Abdel-Megied, A. E. S.; Hawata, M. A. M.; Kasem, E. R.; Shaaban, M. T. *Monatsh. Chem*. **2007**, *138,* 889-897.
[25]. Geroge, S.; Waran, M. P.; Chakraborty, A.; Ravi, T. K. *Acta. Pharm*. **2008**, *58,* 119-129.
[26]. Metawally, M. A.; Shaaban, S.; Abdel-Wahab, B. F.; Gamal, A. Curr. Org. Chem. **2009**, *13,* 1475-1495.
[27]. Winter, C. A.; Risley, E. A.; Nuss, G. W. *Soc. Exp. Biol. Med.* **1962**, *111,* 544-547.
[28]. Loveleen, V.; Sharad, W. *Pharma Innov. J.* **2013**, *2(5)*, 41-50
[29]. Bauer, A. W.; Kirby, W. M.; Sherris, C.; Turck, M. *Am. J. Clin. Path.* **1966**, *45,* 493-496.
[30]. Pfaller, M. A.; Burmeister, L.; Bartlett, M. A.; Rinaldi, M. G. *J. Clin. Microbiol.* **1988**, *26*, 1437-1441.
[31]. National Committee for Clinical Laboratory Standards. Performance Vol. 41, Antimicrobial susceptibility of Flavobacteria, 1997.
[32]. National Committee for Clinical Laboratory Standards. Methods for dilutionantimicrobial susceptibility tests for bacteria that grow aerobically. Approved standard M7-A3. National Committee for Clinical Laboratory Standards, Villanova, PA, USA, 1997.
[33]. National Committee for Clinical Laboratory Standards, Reference Methodfor Broth Dilution Antifungal Susceptibility Testing of Conidium-Forming Filamentous Fungi: Proposed Standard M38-A, NCCLS, Wayne, PA, USA, 2002.
[34]. National Committee for Clinical Laboratory Standards, Method forAntifungal Disk Diffusion Susceptibility Testing of Yeast: Proposed Guideline M44-P, NCCLS, Wayne, PA, USA, 2003.
[35]. Liebowitz, L. D.; Ashbee, H. R.; Evans, E. G. V.; Chong, Y.; Mallatova, N.; Zaidi, M.; Gibbs, M. *Diagn. Microbiol. Infect. Dis.* **2002**, *4*, 27-33.
[36]. Matar, M. J.; Ostrosky-Zeichner, L.; Paetznick, V. L.; Rodriguez, J. R.; Chen, E.; Rex, J. H. Antimicrob. Agents Chemother. **2003**, *47*, 1647-1651.
[37]. Skhen, P.; Storeng, R.; Scudiero, D.; Monks, A.; McMahon, J.; Vistica, D.; Warren, J. T.; Bokesch, H.; Kenney, S.; Oyd, M. R. *J. Nat. Cancer Inst.* **1990**, *82*, 1107-1112.