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Synthesis of Polynuclear Heterocyclic Compounds Derived from 1-Biphenyl-4-yl-4-(1-methyl-1H-benzimidazol-2-yl)-But-2-en-1-One with Expected Biological Activity

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NUCLEOPHILIC addition of barbituric acid, thioglycolic acid with malononitrile and 3-Amino-5-pyrazolinone with 1-Biphenyl-4-yl-4-(1-methyl-1H-benzimidazol-2-yl)- but-2-en-1-one (1) gives pyrimidine, Thiazol and pyrazolinone derivatives (2-4) , repectively.

Compound 4 which reacted with Arylidene malononitrile in ethanol to give pyrano derivatives (5).

Cyclocondensation of chalcone (1) with thiourea under reflux in ethanolic NaOH led to thiazine derivative (6) and treatment of (6) with bromine – acetic acid mixture afforded compound (7) similarly, cyclization of chalcone (1) with orthophenylinediamine in Dimethylformamide (DMF) gave diazepine derivatives (8).

Also, chalcone (1) reacted with 3-Amino-5-pyrazolinone in ethanol to give pyrazolinone derivative (9). (Scheme 1).

The condensation of compound (1) with 2- cyanoacetohydrazide through the acid-catalyzed reaction to form the Schiff base (10) by heating in 96% acetic acid gave the diazetidine derivative (11) which upon reaction with phosphorous oxychloride gave the chlorodiazetidine derivative (12), the latter compound reacted with 2ry amines and yielded the substitueted amino diazetidine derivatives ($13_{a\text{-}c}$).

On the other hand, the Schiff bases (14) produced from condensed compound (1) with 2-Amino-2-(hydroxy methyl) propane-1,3-diol in ethyl alcohol which reacted with phosphorous oxychloride gave the propanol derivatives (15).

Finally, the cyclization reaction of compound (1) with 2-aminophenol and 2-aminothiophenol in dry benzene formed compounds (16, 17) , repectively.(Scheme 2).

Introduction

Benzimidazole derivatives are of wide interest because of their diverse biological activity and clinical application [1]. This ring system is present in numerous anti-parasitic, fungicidal, anti-inflamatory drugs [2] ,anti-hypertensive, anti¬viral, anti-tumour, anti-helminthic , anti-microbial,anti-oxidant, anti-ulcer, anti-amoebic, anti-histaminic activity, anti-bacterial, antipsychotic, antiprotozoal, antineoplastic, analgesic, anti dopaminergic, anti-hepatitis B virus, anticonvulsant, CNS depressant, anti-parkinson.

In addition, benzimidazoles are very important precursors in organic synthesis as (vitamin B12). Constituents are a milestone in the chemistry of benzimidazoles and antidiabetic activity (3-24).

Synthesis of benzimidazolyl chalcones which have been used as intermediate for the synthesis of bioactive heterocyclic compounds vis, pyrimidine-2,4,6-trione [25] , thiazol-4-one [26], pyrazol-3-one [27], 1,7a-dihydro-pyrano [2,3-c] pyrazole-5-carbonitrile [28], thiazin-2-yl-amine[29],1H-benzo[b][1,4]diazepine, benzocycloheptene and benzo[b][1,4] thiazepin

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[30], allylidene-hydrazide, 1,2 diazetidine-3-carbonitrile, 1H-isoindol-2-yl, 4-morpholin-4-yl, 4-piperidin-1-yl, allylidene amino-propan-1-ol-1,2-diol , which have high biological activity as antimicrobial and anticancer activity.

Results and Discussion

The new derivatives were prepared according to the reaction sequences depicted in Schemes 1 & 2.

Nucleophilic addition of barbituric acid, thioglycolic acid with malononitrile and 3-Amino-5-pyrazolinone [31] with 1-Biphenyl-4-yl-4-(1-methyl-1H-benzimidazol-2-yl)-but-2-en-1-one (1) gave 5-[5-Biphenyl-4-yl-3-(1-methyl-1H-benmidazol-2-yl)-5-oxo-pentyl]-pyrimidine-2,4,6-trione (2), 5-[3-Biphenyl-4-yl-1-(1-methyl-1H-benziomidazol-2-yl)-3-oxo-propyl]-2-methyl-thiazol-4-one(3),5-[1-Biphenyl-4-yl-1-(1-methyl-1H-benziomidazol-2-yl)-allylidenamino]-2,4-dihydro-pyrazol-3-one (4) which reacted with Arylidene malononitrile in ethanol to give 6-Amino-3-[1-biphenyl-4-yl-3-(1-methyl-1H-benzoimidazol-2-yl)-alylideneamino]-4-(4-methoxy-phenyl)-1,7a-dihydro-pyrano[2,3-c] pyrazole-5-carbonitrile (5).

Cyclocondensation of chalcone (1) with thiourea under reflux in dry ethanol in the presence of few drops of glacial acetic acid gave the corresponding 6-Biphenyl-4-yl-4-(1-methyl-1H-benzoimidazol-2-yl)-5,6-dihydro-4H-[1,3] thiazin-2-yl-amine (6) and by treatment with bromine , acetic acid mixture afforded compound 6-Biphenyl-4-yl-5-Bromo-4-(1-methyl-1H-benzoimidazol-2-yl)-6H-[1,3]thiazin-2-yl-amine (7).

Similarly, cyclization of chalcone (1) with orthophenylinediamine in glacial acetic acid gives 4-Biphenyl-4-yl-2-(1-methyl-1H-benzoimidazol-2-yl)-1H-benzo[b] [1,4] diazepine (8), by reaction of 3-Amino-5-pyrazolinone with chalcone (1) in ethanol containing T.E.A to form 5-Amino-4-[3-biphenyl-4-yl-1-(1-methyl-1H-benzoimidazol-2-ylmethyl)-3-oxo-propyl]-2,4-dihydro-pyrazol-3-one (9). (Scheme 1).

Also, chalcone (1) reacted with 2-cyanoacetohydrazide in acetic acid to form Schiff base cyano-acetic acid [1-biphenyl-4-yl-3-(1-methyl-1H-benzimidazol-2-yl)-allylidene]-hydrazide (10) and by heating in 96% acetic acid gave 2-[Biphenyl-4-yl-3-(1-methyl-1H-benzoimidazol-2-yl)-allyl]-4-oxo-[1,2]diazetidine-3-carbonitrile (11) , which upon reaction with phosphorous oxychloride gave 2-[1-Biphenyl-4-yl-3-(1-methyl-1H-benzoimidazol-2-yl)-allyl]-4-chloro-[1,2] diazetidine-3-carbonitrile (12) .

The latter compound reacted with 2ry amines and yielded the substitueted amino diazetidine derivative(13a-c)2-[1-Biphenyl-4-yl-3-(1-methyl-1H-benzoimidazol-2-yl)-allyl]-4-(1H-isoindol-2-yl)-[1,2]diazetidine-3-carbonitrile (13a),2-[1-Biphenyl-4-yl-3-(1-methyl-1H-benzoimidazol-2-yl)-allyl]-4-morpholin-4-yl-[1,2] diazetidine-3-carbonitrile (13b) and 2-[1-Biphenyl-4-yl-3-(1-methyl-1H-benzimidazol-2-yl)-allyl]-4-piperidin-1-yl-[1,2]diazetidine-3-carbonitrile(13c), repectively.

On the other hand, the Schiff bases of 3-[1-Biphenyl-4-yl-3-(1-methyl-1H-benziomidazol-2-yl)-allylidene amino] -propan-1-ol-1,2-diol (14) resulted from condensed compound (1) with 2- amino-2-(hydroxy methyl) propane- 1,3 diol in ethyl alcohol which reacted with phosphorous oxychloride to give [1-Biphenyl-4-yl-3-(1-methyl-1H-benziomidazol-2-yl)-allylidene]-propyl-amine (15).

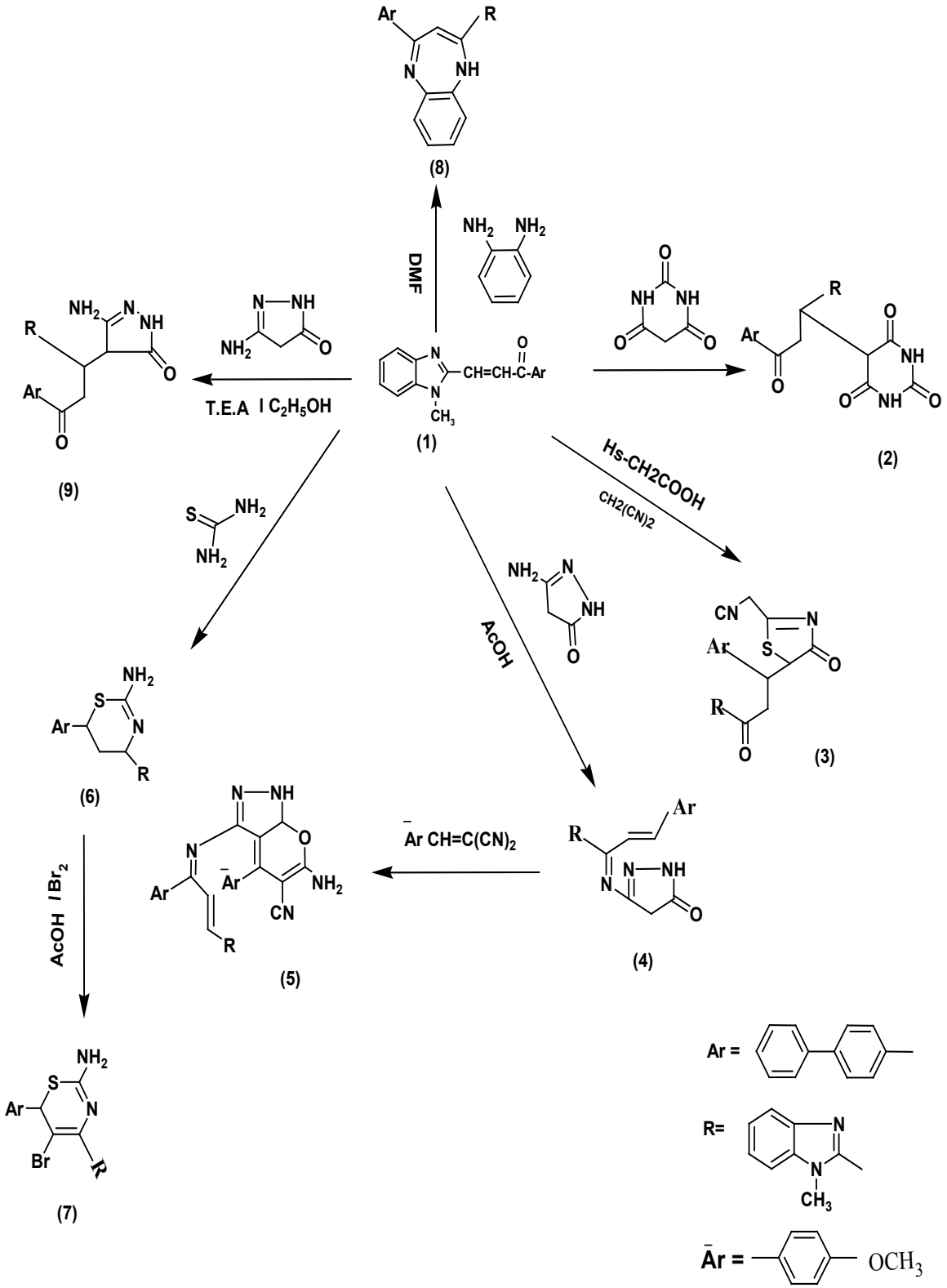
Finally, the cyclization of chalcone (1) with 2- aminophenol and 2-aminothiophenol in dry benzene formed 8-Biphenyl-4-yl-6-(1-methyl-1H-benzoimidazol-2-yl)-5-oxo-9-aza-bezocycloheptene and 4-Biphenyl-4-yl-2-(1-methyl-1H-Benzoimidazol-2-yl)-benzo[b][1,4] thiazepine (16,17), repectively (Scheme 2).

Experimental

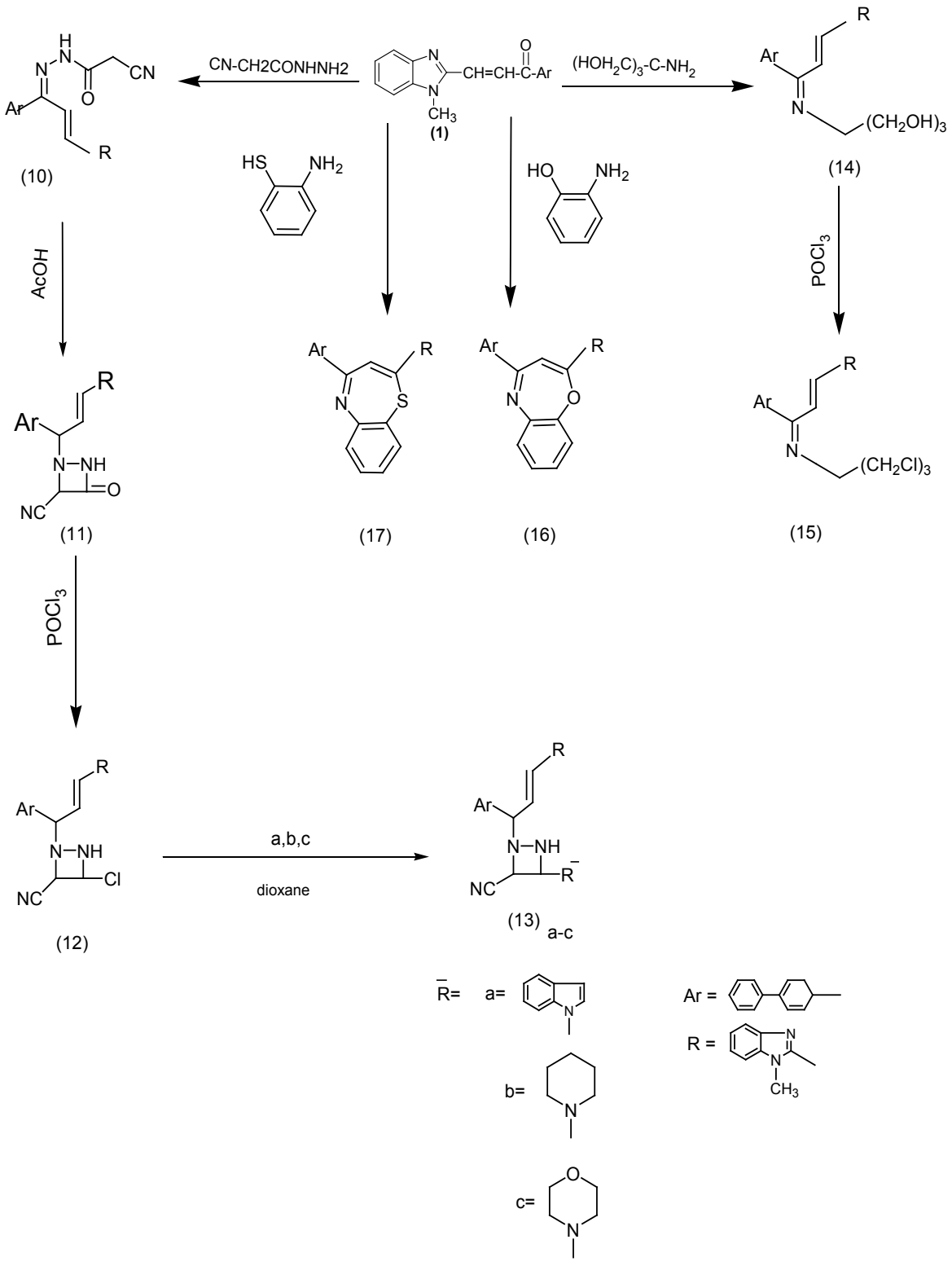
Melting points were determined in open capillary tubes on an electrothermal 9100 digital melting point apparatus (Buchi, stritzerland) Elmer 2400 analyzer (USA).

IR spectra were recorded on a Perkin-Elmer 160 FTIR (USA) as KBr. The ^{1}H-NMR and ^{13}C-NMR spectra were measured on Brüker DRX 500 and 125 MHZ at Max-Plank Institute, Germany in DMSO-d_6.

The elemental analysis were carried out at microanalytical, Faculty of Science, Cairo University by using Perkin-Elmer 2400 C, H, N elemental analyzer.


DMF
T.E.A I C_2H_5OH
AcOH
AcOH I Br_2
Hs-CH2COOH
$CH_2(CN)_2$
$\bar{A}r$ $CH=C(CN)_2$
Ar =
R=
$\bar{A}r$ = OCH_3
(1)
(2)
(3)
(4)
(5)
(6)
(7)
(8)
(9)

Scheme (1)


CN-CH2CONHNH2
(HOH2C)3-C-NH2
(1)
(10)
(14)
AcOH
POCl3
(11)
(17)
(16)
(15)
a,b,c
dioxane
(12)
(13) a-c
R= a=
b=
c=
Ar =
R =

Scheme (2)

Synthesis of 1-(Biphenyl-4-yl)-4-(1-methyl-1H-benzimidazol-2- yl)- but-2-en-1-one (1)

To a solution of [4-(1H-benzimidazol-2-yl)-(1-biphenyl-4-yl)-but-2-en-1-one] in methanol (20 ml) is treated with methanolic sodium hydroxide solution (25 ml; 20%) and then chloroacetic acid (0.01 mol) is added dropwise with continuous shaking. The reaction mixture is refluxed for 2 hr and treated with dilute HCl and the solid obtained was recrystallized from benzene to give compound (1).

Synthesis of 5-[5-Biphenyl-4-yl-3-(1-methyl-1H-benmidazol-2-yl) -5-oxo-pentyl]-pyrimidine- 2,4, 6- trione (2)

To a solution of (1) (0.01 mol) in dry benzene (20 ml), Barbituric acid (0.01 mol) was added. The reaction mixture was refluxed for 10 hr, the solid separated on cooling was crystallized from ethanol to give (2). FT-IR (KBr,υ, cm^{-1}) 3406, 3170 (2NH), 1624,1654,1685 (2C=O,(C=N),1558 (C=C),1303(CH_3). ^{1}HNMR spectrum DMSO d6), 3.8 (S,3H,CH_3),7.7-7.1 (m,15,2 CH,ArH), 10.2 (S,H,NH),13.8 (S,H,NH), 1.5 (dd,2H,CH_2).

Synthesis of 5-[3-(Biphenyl-4-yl)-1-(1-methyl-1H-benzimidazol-2-yl)-3-oxo-propyl]-4-oxo-4,5-dihydro-2-methyl-thiazol-2-yl-acetonitrile (3)

To a solution of (1) (0.01 mole) and malononitrile (0.01 mol) and thioglycolic acid (0.01 mol) in ethanol (50 ml) and acatalytic amount of pipredine was heated under reflux for 5 hr. The reaction mixture was cooled at room temperature and the solution was concentrated by evaboration of the solvent and then the product was collected and recrystallized from ethanol. FT-IR (KBr, υ,cm^{-1}) 1600 (C=O),1581 (C=C),1400 (CH3), 2110 (CN). The mass spectrum (EL/MS) shows molecular ion peak at m/z = 478 (23 %) and base peak at m/z = 190.

Synthesis of 5-[1-Biphenyl-4-yl-1-(1-methyl-1H-benziomidazol-2-yl)-allylidenamino]-2,4-dihydro-pyrazol-3-one (4)

To a solution of compound (1) (0.01 mol) in acetic acid (96%) was added 3-Amino-5-pyrazolinone (0.01 mol). The reaction mixture was stirred at room temperature for 7 and 10 hr. The solid product which formed was filtered off, washed with water, air dried and recrystallized from absolute ethanol to give the title compound. FT-IR (KBr, υ,cm^{-1}) 3402 (NH),1600 (C=O),1342 (CH3). ^{1}H -NMR spectrum (DMSO-d6), 3.3(S,3H,CH_3),7.5-7.1(m,13H,Ar-H),7.7-7.6 (dd,2H,CH=CH), 2.4(S,H,CH_2), ^{13}C-NMR 25.1 (CH_3), 192.4 (C=O),18.1 (CH_2),82.2 (CH=CH).

Synthesis of 6-Amino-3-[1-biphenyl-4-yl-3-(1-methyl-1H-benzoimidazol-2-yl)-alylideneamino]-4-(4-methoxy-phenyl)- 1,7a-dihydro-pyrano [2,3-c] pyrazole-5-carbonitrile (5)

To a solution of (4) (0.01 mol) and arylidene malononitrile (0.01 mol) in ethanol (50 ml) on heating under reflux for 7 hr, the reaction mixture was cooled and the product was collected by filtration and recrystallized from petroleum ether (40-60). FT-IR (KBr, υ,cm^{-1}) 3417, 3059 (NH2, NH), 2191(CN), 1307(CH_3), 1512(C=C),1604(C=N),1253(C-O),1114 (OCH3),The mass spectrum (EL/MS) shows molecular ion peak at m/z = 603/604 M/M^+ (3.9%) and base peak at m/z = 181.

Synthesis of 6-Biphenyl-4-yl-4-(1-methyl-1H-benzimidazol-2-yl)-5,6-dihydro-4H-[1,3] thiazin-2-yl-amine (6)

To a solution of chalcone (1) (0.01 mol) and thiourea (0.01 mol) were dissolved in ethanolic NaOH (10 ml) stirred for 3hr by using a magnetic stirrer and then pored into 200 ml water with continous stirring, this was kept in cold condition in refrigerator for 24 hr, the precipitate obtained was filtered , washed and recrystallized from ethanol to give (6). FT-IR (KBr, υ,cm^{-1}) 3421(NH2), 1608(C=N),1423 (CH3), ^{1}H -NMR spectrum (DMSO-d6), (δ,ppm), 3.3 (S,3H,CH3), 7.6-6.6 (m,14H, CH-thiazine, Ar-H), 7.8 (S,2H,NH2), 1.3 (dd,2H,CH_2- thiazine), 10.7 (S,H,CH- thiazine).

Synthesis of 6-Biphenyl-4-yl-5-Bromo-4-(1-methyl-1H-benzimidazol-2-yl)-6H-[1,3]thiazin-2-yl-amine (7)

A solution of (21) (0.01 mol , in glacial acetic acid (20 ml) was stirred and treated portionwise with Bromine (0.01 mol) at 60-70 ˚C. The solution was further stirred for 2 hr, then cooled in ice . The precipitate product was filtered, washed with light petroleum ether (40-60), stirred with concentrated ammonium hydroxide for 15 min. The solid product was recrystallized from benzene to give (7). 3431 (NH_2), 1677 (C=N), 1365(CH_3), 706 (Br).

Synthesis of 4-Biphenyl-4-yl-2-(1-methyl-1H-benzoimidazol-2-yl)-1H-benzo[b] [1,4] diazepine (8)

To a solution of chalcone (1) (0.01 mol) and orthophenylene diamine (0.01 mol) in Dimethylformamide (30 ml) containing added

amount of glacial acetic are refluxed on water bath for 9-10 hr,then cooling down to room temperature, and the precipitate obtained is filtered and recrystallized from petroleum ether (60-80). FT-IR (KBr, υ,cm^{-1}) 3029(NH), 1300(CH3), 1599(C=C), 1659(C=N), its ^{1}H -NMR spectrum (DMSO-d6) reveals signals (δ, ppm) 7.7-6.8 (m,18H, Ar-H),8.2 (S,H,NH), 3.3 (S,3H,CH_3), ^{13}C-NMR, 22.5 (CH3), 142.3-115.7 (Ar-c), 139.5 (C=N).

Synthesis of 5-Amino-4-[3-biphenyl-4-yl-1-(1-methyl-1H-benzoimidazol-2-ylmethyl)-3-oxo-propyl]-2,4-dihydro-pyrazol-3-one (9)

To a solution of 3-Amino-5-pyrazolinone (0.01 mol) in absolute alcohol (15 ml) cotaining triethylamine (0.5 ml) compound (2) is added (0.01 mol), the reaction mixture was stirred at room temperature for 7 and 10 hr.

The solid formed after adding water (20 ml) is filtered off ,washed with water, air dried and recrystallized from absolute ethanol to give the title compound . FT-IR (KBr, υ,cm^{-1}) 1678,1620 (2C=O), 3250, 3062 (NH_2lNH),1381(CH_3), 1527 (C=C).

Synthesis of cyano-acetic acid [1-biphenyl-4-yl-3-(1-methyl-1H-benzimidazol-2-yl)-allylidene]-hydrazide (10)

To a solution of compound (1) (0.01 mol) in acetic acid (10 ml) is added 2-cyanoacetohydrazide (0.01 mol) .The reaction mixture is stirred at room temperature for 30 min, the solid formed is filtered off, washed with water , air dried and recrystallized from absolute ethanol. FT-IR (KBr, υ,cm^{-1}) 3186(NH), 1685 (C=O),1624(C=O),1685,1624 (2C=O),1357(CH3), its 1H -NMR spectrum (DMSO-d6), (δ,ppm) 7.9-6.8 (m,13H,Ar-H),13.9 (S,H,NH),8.07 (dd,2H,CH=CH), 3.3 (S,2H,CH2).

Synthesis of 2-[Biphenyl-4-yl-3-(1-methyl-1H-benzimidazol-2-yl) -allyl]-4 – oxo -[1,2] diazetidine-3-carbonitrile (11)

Compound (10) (0.01 mol) is heated under reflux in acetic acid (96 %) for 5hr , the reaction mixture was then concentrated to 1/2 of its volume and left to cool at refrigerator . The product formed was collected by filtration , washed with water , air dried and recrystallized from ethanol. FT-IR (KBr, υ,cm^{-1}). 3280 (NH), 2130(CN), 1662 (C=O),1411(CH3),1597 (C=C).

Synthesis of 2-[1-Biphenyl-4-yl-3-(1-methyl-1H-benzimidazol-2-yl)-allyl]-4-chloro-[1,2] diazetidine-3-carbonitrile (12)

A solution of compound (11) (0.01 mol) in phosphorus oxychloride (50 ml)was heated under reflux for 3hr, the reaction mixture was cooled and then poured into ice-water while stirring ; the solid that formed was filtered off ,washed with water ,air dried and recrystallized from ethanol to give (12). FT-IR (KBr, υ,cm-1) 1620 (C=N),3414 (NH),1384(CH3),744(Cl). 1H -NMR spectrum (DMSO-d6),reveals signals (δ,ppm) 7.6-6.8 (m,14H,CH,Ar-H),7.7(dd,2H,CH=CH),3.5(S,3H, CH3),12.2 (S, H,NH),13.7 (S,H,CH-diazetidine).

Synthesis of 2-[1-Biphenyl-4-yl-3-(1-methyl-1H-benzimidazol-2-yl)-allyl]-4-(1H-isoindol-2-yl), 4-morpholin-4-yl and 4-piperidin-1-yl -[1,2] diazetidine-3-carbonitrile (13 a-c)

A solution of (12) and appropriate secondary amines (0.01 mol) in dry dioxane (15ml) containing few drops of triethylamine was refluxed until the material disappeared for 7-12 hr, the solvent was then evaporated till dryness and the resulting residue was triurated with water (50 ml) and set aside overnight at refrigerator .The solid that formed is filtered off, and recrystallized from the proper solvent to give the title compounds. FT-IR(KBr,υ,cm^{-1}) 3417(NH),1635(C=N),1523(C=C), 1384 (CH_3). 2121(CN), 1620(C=N),1384 (CH3),1516 (C=C), 3414 (NH), 2121 (CN).

Synthesis of 3-[1-Biphenyl-4-yl-3-(1-methyl-1H-benzimidazol -2-yl)-allylidene amino]-propan-1-ol-1,2-diol (14)

To a solution of (2) (0.01 mol) in acetic acid (96 %) (10 ml) is added to 2-amino-2-(hydroxy methyl) propane-1,3 diol (0.01 mol), and the solid formed is filtered off, washed with water ,air dried and recrystallized from absolute ethanol. FT-IR (KBr, υ,cm^{-1}) 1604(C=N),1338(CH3), 3433-3302 (3 OH), its 1H -NMR spectrum (DMSO-d6),3.4(t, H,CH2OH),3.7(dd,2H,CH2OH),8.17.2(m,13H, ArH),9.8(d,2H,CH=CH),2.5(S,2H,CH2),13C-NMR , 56.1 (CH2OH), 23.1(CH3), 129.5-127.3 (Ar-C),85.2 (CH=CH).

Synthesis of [1-Biphenyl-4-yl-3-(1-methyl-1H-benzimidazol-2-yl)-allylidene]-propyl-amine (15)

A solution of compound (14) (0.01 mol) in phosphorus oxychloride (50 ml) is heated under reflux for 3hr, the reaction mixture is cooled down and then poured onto ice-water while

stirring , the solid formed is filtered off, washed with water , air dried and recrystallized from ethanol to give (15). FT-IR (KBr, υ,cm^{-1}) 1620 (C=N),1384 (CH3),759 (Cl). The mass spectrum (EL/MS) shows molecular ion peak at m/z = 497 (2.8%) and base peak at m/z =68.

Synthesis of 8-Biphenyl-4-yl-6-(1-methyl-1H-benzimidazol-2-yl)-5-oxo-9-aza-benzocy-cloheptene and 4-Biphenyl-4-yl-2-(1-methyl-1H-Benzimidazol-2-yl)-benzo[b][1,4]thiazepine (16,17)

To a solution of compound (1) (0.01 mol) in dry benzene (30 ml) is added to 2- aminophenol and 2-aminothiophenol (0.01 mol) with drops of glacial acetic acid ,the reaction mixtures is refluxed on the water bath (8-10 hr), the solids separated on cooling at room temperature, are recrystallized from ethanol to give the title compounds. FT-IR (KBr, υ,cm^{-1}) 1431 (CH_3), 1696 (C=N), 1111 (C-O-C)., 1684 (C=N), 1313 (CH3), 1571(C=C). its 1H-NMR spectrum (DMSO-d6), (δ,ppm) 2.5 (S,3H,CH3),7.8-7.2 (m,17H,Ar-H),8.2 (S, H, CH-thiazepine)

Biological Activity

Antimicrobial activity

The antimicrobial activity of the synthesized compounds is determined in vitro using the disc diffusion [31].

All the synthesized compounds are screened for their antibacterial and antifungal activity in vitro by disc diffusion method using nutrient agar medium against following microorganisms, *Staphylococcus aureus* [ATCC 29213] *Bacillus subtilis* (Gram positive bacteria), *Escherichia coli* [ATCC 27853] , *Pseudomonas aeuroginosa* [ATCC 25922] (Gram negative bacteria) and fungal species like *Candida albicans*, *Aspergillus flavin* organisms.

Ampicillin is used as a positive control for antibacterial screening and an Amphotericin B is used as a positive control for antifungal screening. DMSO (Dimethylsulfoxide) is diluent which does not affect the growth of microbes (negative control). The tested compounds were dissolved in (DMSO). Filter paper discs (Whatman, 5 mm diameter) were saturated with former solution for bacterial test. The saturated filter paper discs were placed on the surface of solidified Czapek's Dox agar dishes seeded by test bacterial and fungal strains.The inhibition zones were measured in mm at the end of an incubation period of 48 hr at 28°C. The screening results have been tabulated in Table 1 and represented in Fig. 1 & 2.

TABLE 1. Antimicrobial study of the newly synthesized compounds of Part (II)

Comp. No.	Antibacterial activity				Antifungal Activity	
	Gram positive		Gram negative			
	B.subtilis	*S.aureus*	*E.coli*	*P.aeruginosa*	*A. flavin*	*C.albicans*
	Inhibition zone diameter (mm)					
1	10	6	11	8	10	12
2	13	6	9	7	-	--
3	9	12	--	--	9	11
4	11	7	--	8	8	10
5	12	9	10	10	11	9
6	10	8	-	--	12	10
7	12	11	10	14	-	-
8	16	11	16	12	9	11
9	14	15	11	-	9	11
10	10	12	12	--	-	15
11	-	--	-	-	11	14
12	16	9	-	-	13	16
13	15	12	--	--	14	9
14	-	-	11	10	-	16
15	11	14	19	14	13	16
16	13	10	11	13	12	-
Ampicillin (antibacterial drug)	20	18	22	17	-	-

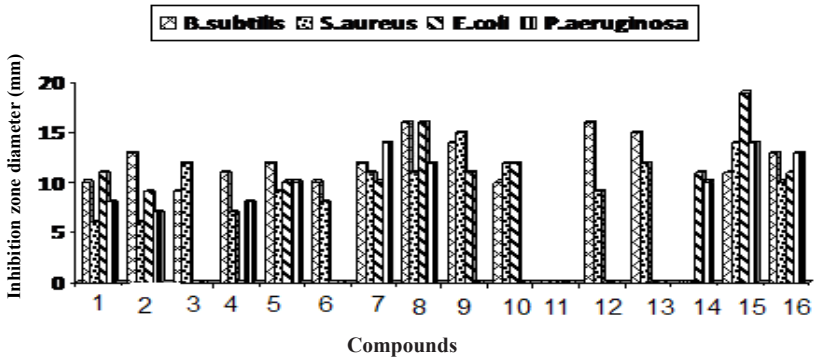


Fig.1 . Antibacterial activity of newly synthesized compounds.

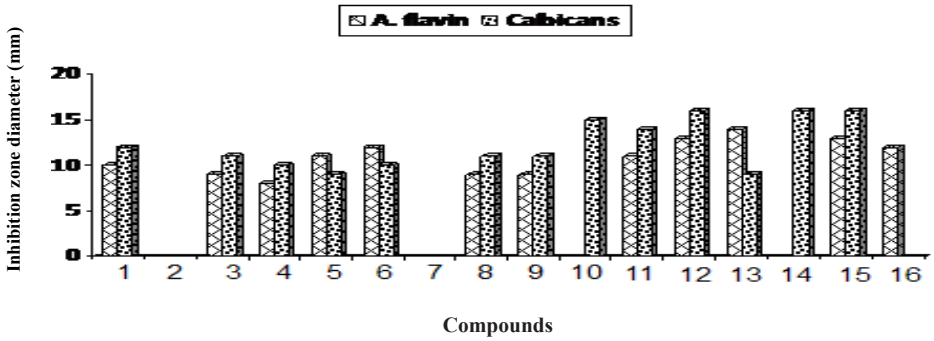


Fig.2 . Antifungal activity of newly synthesized compounds.

Antiproliferative activity

All the newly synthesized compounds were tested for antiproliferative activity in vitro against different types of cancer cell lines as (HEPG2) human liver carcinoma cell line, (MCF-7) human breast cancer cell line and (HCT-116) human colon cancer cell line at different concentrations (100, 50, 25, 12.5, 6, 25.0)ug/ml, and the Doxorubicin, one of the most effective anticancer agents ,was used as a reference drug . These cell lines were grown in RPMI -1640 medium supplemented with 2mg/ml sodium bicarbonate,4.5 mg/ml glucose, 100 ug/ml streptomycin sulphate ,40 ug/ml gentamycin,100 U/ml pencillin, as well as 10% (vol/vol) foetal bovine serum (FBS). All cell lines incubated at 37 ºC in humidified air containing 5% CO_2. Cytotoxicity was determined by MTT (3-[4,5¬ dimethylthiazol-2-yl]-2,5-diphenyltetrazolium bromide) is a water soluble tetrazolium salt, which is converted to an insoluble purple formazan by cleavage of the tetrazolium ring by succinate dehydrogenase within the mitochondria. The formazan product is impermeable to the cell membranes and therefore it accumulates in healthy cells [32] .

Breifly, the Plate cells (104 –106 cells) in 200 µl PBS in 96-well (flat bottom). Add 20 ul of MTT solution, mix well, incubate for 4hr in 37°C and incubate additional 1hr in 37 ºC in the dark. Then read plate in ELISA Reader – measure OD in 570 nm (background wavelength is 570 nm). IC50% (µg/ml), *i.e.* (50% inhibition concentration) was calculated for the tested compounds by using (sigmaplot software). The results of the IC50% of the compounds was tabulated (Table 2) and represented in Fig. 3.

Results and Discussion

From Table 1, the results showed that all the compounds have moderate sensitivity towards Gram positive bacteria compared to positive control (Ampicillin) except the compounds (11,

14) and all compounds showed slight to moderate sensitivity toward the Gram negative bacteria except (3,4,6, 9,11,12,13) which showed no antibacterial activity.

Antiproliferative activity of the newly synthesized compounds against human carcinoma cell lines Part (II): IC_{50} %(µg/ml) of newly synthesized compounds on human liver carcinoma cell line (HEPG2), human breast cancer cell line (MCF-7) (Table 2 & Fig.3).

Inhibition concentration (50%) (IC_{50}) µg/ml		
Comp. No.	**HEPG-II**	**MCF-7**
1	33.6	120.09
2	63.6	103.22
3	8.24	60.34
4	12.81	8.51
5	35.1	5.68
6	57.9	95.02
7	3.8	68.7
8	20.2	21.5
9	7.6	6.9
10	10.3	125.5
11	140.98	32
12	45.3	87.2
13	66.9	21.7
14	23.3	34.2
15	74.6	42.5
16	49.7	11.2

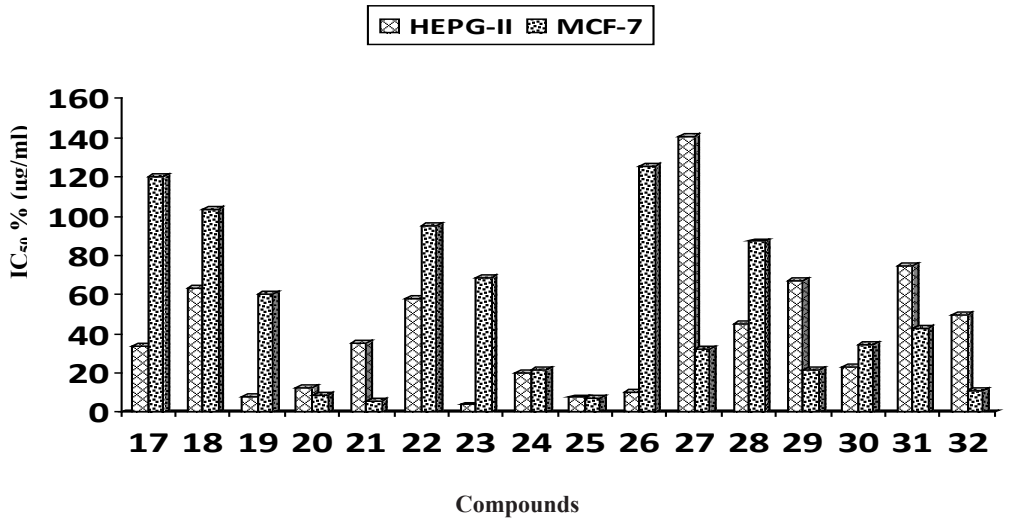


Fig. 3. Antiproliferative activity of the compounds against human carcinoma cell lines (HEPG2), (MCF-7).

On the other hand, all compounds showed moderate to good antifungal activity towards (*Candida albicans and Aspergillus flavin*) except compounds (2,7,10,14,16) which showed no antifungal activity compared to positive control (Amphotericin B).

From Table 2, the results showed that compounds (7,9,2,10,3) were the most active recording IC50% values in the order (3.8, 7.6, 8.24, 10.3, 12.81) µg/ml against HEPG-II cell line. On the other hand,the compounds (5,9,3,16) showed to be the most active recording IC50% values in the order (5.68, 6.9, 8.51, 11.2) µg/ml against MCF-7 cell line.

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تحضير بعض المركبات الحلقية الغير المتجانسة عديدة الأنوية من1 -باى فينيل4--يل4--(-1ميثيل-بنزيميدازول2--يل)-بيوت- -2اين1-- أون مع توقع النشاط البيولوجى لهذه المركبات

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الإضافة النيوكليوفيلية لحمض الباربيتوريك ، حمض الثيوجليكوليك مع المالونونيتريل وايضا -3 أمينو5--بيرازولينون مع -1 باى فينيل -4يل-4-(-1 ميثيل- -1 بنزيميدازول2--يل)- بيوت2--إين1--أون (1) ليعطوا مشتقات البريميدين، الثيازول والبيرازولينون (4-2) على التوالى.
ويتفاعل المركب (4) مع الأريليدين- مالونيتريل فى الميثانول يعطى مشتق البيرينو (5).
ويتكاثف الشالكون (1) مع ثيويوريا فى ايثانوليك هيدروكسيد الصوديوم مكونا مشتق الثيازين (6) التى عولجت بخليط من (البرومين وحمض الخليك) مكونا مركب (7).
وايضا، بحولقة شالكون (1) مع اورثوفينيلين داى امين فى حمض الخليك الثلجى مكونا مشتق الديازيبين (8) .
ايضا تفاعل شالكون (1) مع -3 أمينو5-- بيرازولينون فى الكحول الإيثيلى مكونا مشتق البيرازولينون (9) . (مخطط 1).
وبتكاثف مركب (1) مع -2سيانو اسيتوهيدرازيد فى وجود حامض مكونا قاعدة شيف (10). التى بالتسخين فى حمض الخليك (96%) تكون مشتق البيرازولينون (11) والتى بتفاعلها مع كلوريد اكسيد الفوسفور مكونا مشتق الكلوروبيرازول (12) الذى تفاعل مع الأمينات الثانوية وأعطى مشتقات البيرازول ذات المستبدلات الأمينية (13 أ-ج) على التوالى.
من جهة اخرى ، تكونت قاعدة شيف (14) من تكاثف المركب (1) مع -2 أمينو2--(هيدروكسى ميثيل) بروبان 3،1- – دايول فى الكحول الإيثيلى والذى تم تفاعله مع كلوريد أكسيد الفوسفور ليعطى مشتق البروبانول (15).
وأخيرا، تم حولقة المركب (1) مع -2 أمينو فينول و-2 أمينو ثيازول فى البنزين الجاف ليعطى المركبات (16 ، 17) على التوالي (مخطط 2).