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RESEARCH ARTICLE

Synthesis, Molecular Docking and Antimicrobial Activity of 2-(3, 4-dihydro-3-oxo-2H-benzo [b] [1, 4] thiazin-2-yl) acetic acid Esters

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ABSTRACT

Esters of 2-(3, 4-dihydro-3-oxo-2H-benzo[b] [1, 4] thiazin-2-yl) acetic acid are synthesized and evaluated for their antimicrobial activity in vivo. All compounds are characterized by spectroscopic techniques such as FTIR, H^1 NMR, C^{13} NMR and Mass. Molecular docking of these compounds is carried out in silico. The Molecular docking study provides detailed information about the nature and reactivity of the compounds. This helps to judge the biological activity of compounds using computer softwares. Molecular docking of these compounds is done with alpha amylase enzyme responsible for microbial attack. The result shows that all these esters are excellent inhibitors of alpha amylase.

KEYWORDS

2-(3, 4-Dihydro-3-oxo-2H-Benzo[b][1,4]Thiazin-2-yl)Acetic Acid, Antimicrobial, Docking

INTRODUCTION

1, 4- benzothiazine is a prominent nucleus and plays vital role in heterocyclic chemistry. Due to the presence of a fold along nitrogen-sulphur axis; 1,4 - benzothiazine possesses wide spectrum of biological activities such as antimicrobial, antiulcer, blood cholesterol lowering, sedative, antispasmodic, anti-inflammatory, anti-helminthic, CNS depressant, antioxidant, antimalerial, antidiabetic, antiviral , analgesic and antipyretic, antagonist, vasorelaxant, antiproliferative, potassium channel opener, anti-rheumatic, anti-hepatitis, anti-edema[1-14], antifungal[15-17], beta-ribosidases[10] inhibitor anticancer agents[19-20], antiviral[21], antihypertensive[22], anticonvulsant[23], vasodialator [24] antitubercular[25] etc. These derivatives are also used as lipoxygenase[26], inhibitors. 1, 4 benzothiazine derivatives are very effective against pick disease[27], epilepsy, ischemia, and neurodegenerative disorders and perform major and crucial role in Parkinson's disease, Alzheimer disease, acetylcholine esterase inhibitors[28]. These derivatives are efficient in treatment of breast cancer, leukaemia[18,] auto immune diseases, osteoporosis[19]. They possess lots of pharmacological properties, in vivo antitumor, cytotoxic activity, calcium channel blockers, phosphodiesterase, anticataracts agents, dopamine D_4, Na^+/H^+ exchange inhibitors, matrix metalloproteinase inhibitors etc[11]. 1, 4-benzothiazine derivatives play very important role in medicinal chemistry, organic synthesis and 1, 4- benzothiazine acetic acid is used as starting material in many organic transformations. By making a slight change in substitution pattern of 1, 4- benzothiazines results into enormous change in biological activity. These derivatives are also useful as dyes, photographic developers, ultraviolet absorbers, semiconductors[10,13,29], 3-hydroxy- 3-methylglutaryl-CoA reductase inhibitors,

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diruetics[30] etc. Esterification of 1, 4 - benzothiazine acetic acid can be done by using thionyl chloride, trimethyl amine as a catalyst in dichloromethane; this is a two-step reaction[31]. For direct esterification of 1, 4 - benzothiazine acetic acid with alcohol EDC is very efficient coupling agent as its urea is soluble in water. In the present study, esters of 1, 4 - benzothiazine acetic acid are synthesized using coupling agent EDC[32] and they are screened for their antimicrobial activity in vivo. These derivatives are exposed to four bacterial strains including two gram positive, two gram negative bacteria and two fungal strains. Molecular docking was also carried out in silico to know the potency of these molecules using the software hex 6.0[33,34]. Molecular docking is the method of determining the drug activity against the enzyme responsible for that particular disease. In our study for molecular docking, we have taken the enzyme alpha amylase[35] from Bacillus substitis as a receptor responsible for microbial attack and our synthesized derivatives as ligands.

MATERIAL AND METHODS

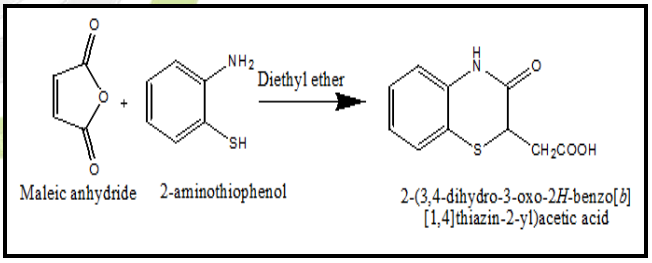
All the reagents and solvents were purchased from Sigma-Aldrich and they were used as received. Reactions and purity of products were monitored by column chromatography with a hexane /ethyl-acetate 4:1 mixture as eluent for 2-(3, 4-dihydro-3-oxo-2H-benzo[b] [1, 4] thiazin-2-yl) acetic acid esters; Melting points were determined using open capillaries method and the reported values are uncorrected. Infrared spectra (ATR) were recorded on FT-IR spectrometer Shimdzhu 8400S FT-IR in the range of 4,000-400 cm−1. The NMR spectra were recorded on a 500MHz instrument at ambient temperature using deuterated dimethylsulfoxide (DMSO-*d6*) solutions of the samples .The chemical shifts δ are given in ppm, with respect to tetramethylsilane as an internal standard. The structures of compounds are drawn with the help of chem draw 8.0. The mass spectra were recorded on 6460 Triple Quadrupole LC/MS model.

Step 1: Synthesis of (3-oxo-3, 4-dihydro-2H-1, 4-benzothiazin-2-yl) acetic acid (1)

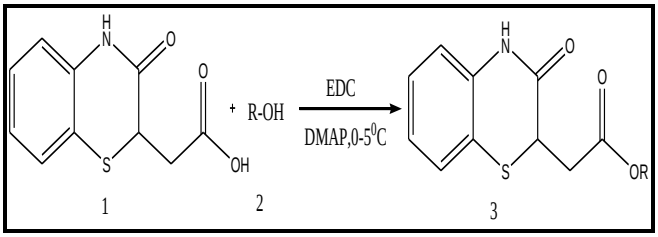
To a solution of pure maleic anhydride (4.9g, 0.05 mole) in diethyl ether (20 ml), a solution of *o*-amino-thiophenol (6.25 ml, 0.05 mole) in ether (20 ml) was added drop wise with constant stirring at room temperature for 3 hrs in cool water bath. When a colourless product was separated out, it was filtered off, washed with ether and the crystalline product was purified by recrystallization from ethanol which gave colourless needles. Yield- 95%, m.p-183-185^{0}c.

Step 2: Synthesis of ester derivatives of (3-oxo-3, 4-dihydro-2H-1,4-benzothiazin-2-yl) acetic acid (3)

To a solution of appropriate carboxylic acid derivative (1 mmol) and phenol or amine derivative (1 mmol) in DCM (10 mL), DMAP (0.2 mmol) and EDC (1.1 mmol) were added and the resulting solution was stirred for 4 hrs at 0-5^{0}C temperature. The reaction mixture was quenched with 0.5 N HCl and extracted with DCM. The organic phase was washed with a 1% $NaHCO_3$ solution and brine, dried over Na_2SO_4, and evaporated under vacuum. The residue was purified by column chromatography using hexane-EtOAc as eluents.



Scheme 1: Synthesis of 2-(3, 4-dihydro-3-oxo-2H-benzo[b][1,4]thiazin-2-yl)acetic acid



R, a=C_6H_5, b=nC_3H_7 c=C_6H_{12}, d=C_5H_{11}, e=CH_2CH_2Cl, f=$C_6H_5CH_2$, g=m-$CH_3C_6H_4$, h=o-$CH_3C_6H_4$, i=secC_3H_7

Scheme 2: Synthesis of 2-(3, 4-dihydro-3-oxo-2H-benzo[b][1,4]thiazin-2-yl)acetic acid ester derivatives

Computational Method, Molecular Docking

Molecular docking is the method which predicts the preferred orientation of small molecule (ligand) and large molecule (receptor, enzyme) to form complex with one another in three dimensional spaces[36]. It plays key role in designing of rational drugs to minimise toxicity and side-effects. It is the most important tool for selecting potential therapeutic drugs[37]. Synthesis of the bioactive molecules without knowing their activity is very difficult task and time consuming process. To overcome this, computer added drug design (CADD) is a specialized, prominent drug discovery tool for the screening of thousands of compounds[38]. It is a valuable tool to save the time and at the same time to improve the selection efficiency.

For our present study we used bioinformatics tools, biological databases like Drug Bank, PDB (Protein Data Bank) and software's like Hex, Biova discovery studio 4.5 visualizer. Chem. draw is used for effective drawings of 2D-3D structures of compounds and it helps the chemists to draw molecules, reactions and schematic diagrams, calculate chemical properties conveniently. The 2D-3D structures of 2-(3, 4-dihydro-3-oxo-2H-benzo[b] [1, 4] thiazin-2-yl) acetic acid and its ester derivatives are constructed on chem. draw 8.0.Then these chem. draw files are converted into protein data bank files using Biova discovery studio 4.5visualizer. We obtained crystal structure of alpha amylase protein (pdb id-1BAG) for antibacterial activity from the Protein Data Bank. All these structures are utilised for molecular docking process using software Hex 6.0.

The following parameters were used for the molecular docking process.

- Correlation type – Shape + Electrostatics
- FFT Mode – 3D
- Post Processing–MM Energies
- Grid Dimension – 0.6
- Receptor range – 180
- Ligand Range – 180
- Twist range – 360
- Distance Range – 40

Using all these parameters 2-(3, 4-dihydro-3-oxo-2H-benzo[b][1,4]thiazin-2-yl) acetic acid and its esters derivatives are docked against alpha amylase enzyme. The three dimensional structure of alpha amylase enzyme formed complex with 2-(3, 4-dihydro-3-oxo-2H-benzo[b][1,4] thiazin-2-yl) acetic acid and its ester derivatives with good binding site. This study enabled us to predict the interaction and orientation of 2-(3, 4-dihydro-3-oxo-2H-benzo [b] [1,4] thiazin-2-yl)acetic acid and its ester derivatives into alpha amylase active sites. We carried out 10 docking operations for each ligand and selected top score for binding affinity of each ligand-enzyme complex.

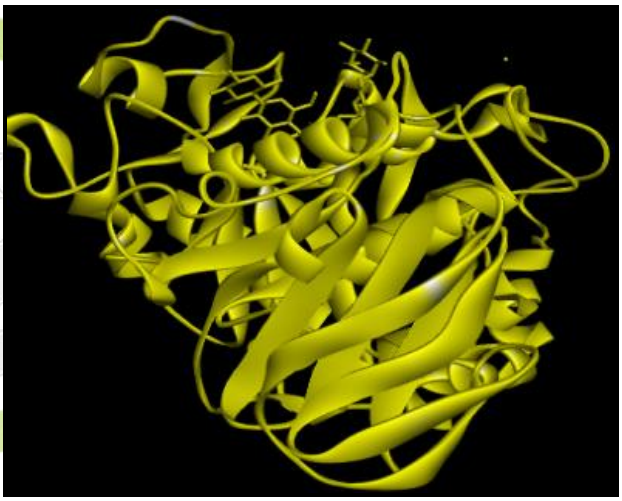


Figure 1: Structure of alpha amylase receptor Complex protein (PDB ID: 1BAG)

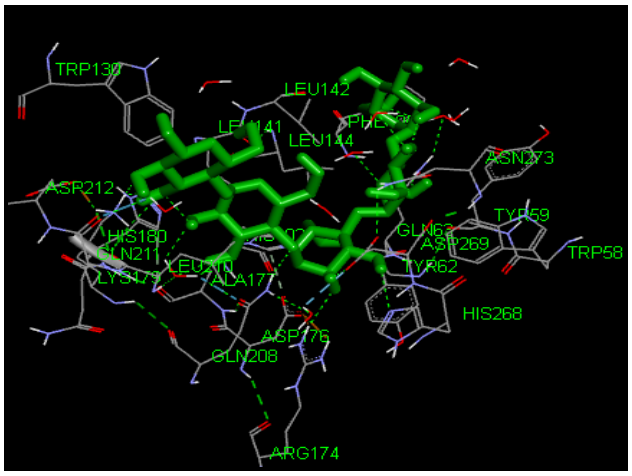


Figure 2: Compound 3c with active site of 1BAG

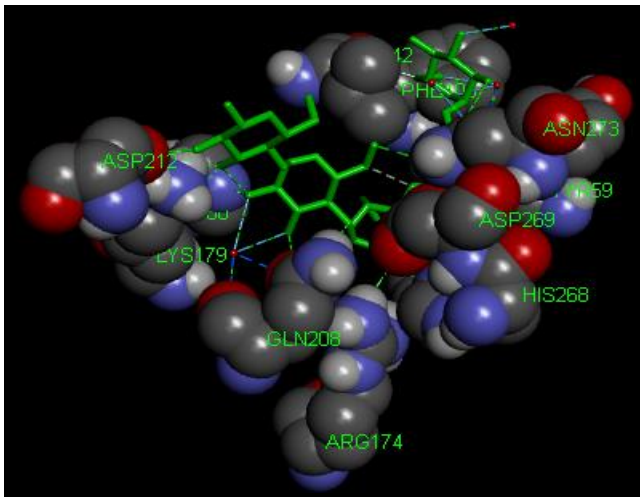


Figure 3: Docking complex of 1BAG with parent acid (ball and stick)

Table 1: Calculated free binding energy and hydrogen bonds values of the synthesized compounds

Sr. No.	Compound	E-Value	Hydrogen bond
1	3a	-988.89	1
2	3b	-813.23	1
3	3c	-1052.5	1
4	3d	-1009.2	1
5	3e	-819.01	1
6	3f	-1035.6	1
7	3g	-1025.2	1
8	3h	-1043.7	1
9	3i	-807.47	1
10	Parent acid	-2241.6	1

We have noted that there is a variation in the results obtained from the experimental and theoretical data. From the table, the results showed that parent acid and all of its ester derivatives show significant binding energy with strong hydrogen bonding. The parent acid has less binding energy than all other ester derivatives i. e parent acid form more stable complex with alpha amylase enzyme with minimum energy. Among all the ester derivatives compounds **3c, 3g, 3f** and **3h** show good binding affinity with alpha amylase.

In vitro Antimicrobial Evaluation

Antimicrobial properties of nine esters of 2-(3, 4-dihydro-3-oxo-2H-benzo [b] [1, 4] thiazin-2-yl) acetic acid derivatives were assayed against bacterial strain. This testing is carried out using disc diffusion method. Various bacterial strains - *Staphylococcus aureus* (NCIM 2079), *Escherichia coli* (NCIM 2109) and fungal strains *Candida albicans* (NCIM 3471) were used as test microorganism to evaluate the antimicrobial testing of newly synthesized compounds. Pure culture of test bacterial strain was picked with a loop, and the growth was transferred into a tube containing 5 ml nutrient broth medium, while pure culture of test fungal strain was transferred into a tube containing 5 ml of a MGYP medium. The broth culture was incubated at 37°C until it achieves or exceeds the turbidity of the 0.5 McFarland standard (usually up to 6 hours). The turbidity of the actively growing broth culture is adjusted with sterile saline or broth to obtain turbidity optically comparable to that of the 0.5 McFarland standard. This resulting suspension contains 2 x 10^8 CFU/ml of microbial cells. Within 15 minutes after adjusting the turbidity of the inoculum suspension, a sterile cotton swab was dipped into the adjusted suspension. The swab was rotated several times and pressed firmly on the inside wall of the tube above the fluid level. The surface of a nutrient agar plate was inoculated by streaking the swab over the entire sterile agar surface. This procedure is repeated by streaking several times, rotating the plate approximately through 60° each time to ensure an even distribution of inoculums Stock solution [1000 microgram per ml] of each newly synthesized compounds were prepared in dimethylsulfoxide (DMSO). The sterile discs of 6 mm diameter were used in this assay. The disc diffusion assay was carried out by taking concentration 100 microorganism per disc**.** The discs immersed with compounds were dispensed onto the surface of the inoculated agar plate. Also, Ciprofloxacin (10 microgram/disk, Amphotericin-B (100 units/disk) [Hi-media, Mumbai, disc diameter 6 mm] moistened with DMSO were placed on agar plate as standard. Each disc was pressed down to ensure complete

contact with the agar surface. The plates were placed in a refrigerator at to 8°C for 30 minutes and then incubated at 37°C for 24 hours. After 24 hours of incubation, each plate was examined.

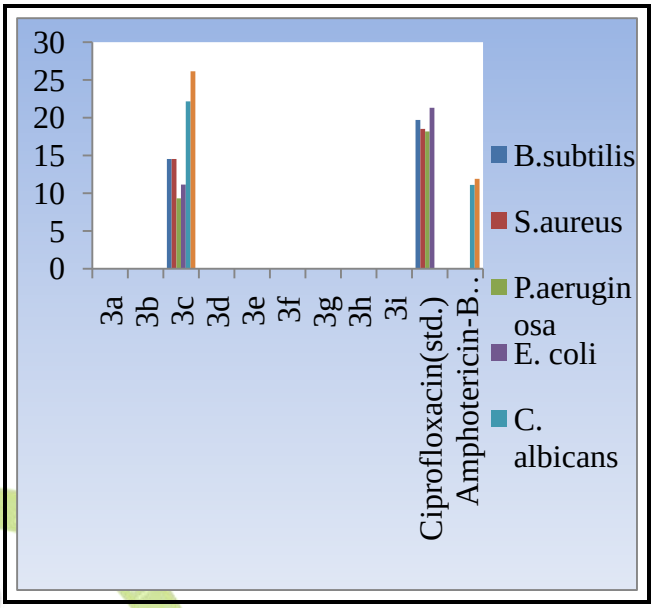
The diameters of the zones of complete inhibition including the diameter of the disc were measured using Vernier calliper, which is held on the back of the inverted Petri plate. Table 1 overviews the antibacterial and antifungal properties.

RESULTS AND DISCUSSION

Synthesis

Firstly 1, 4-benzothiazine acetic acid is prepared by carrying out the reaction between 2-aminothio phenol and maleic anhydride at room temperature for three hours. This acid was then esterified in proper proportion with different aromatic and aliphatic alcohols using EDC as a coupling agent and dimethyl amino-pyridine as a base in dichloromethane at 0-5^{0}C for three hours. Out of all these derivatives of 2-(3, 4-dihydro-3-oxo-2H-benzo[b][1,4]thiazin-2-yl)acetic acid only one compound (3c) inhibited all the four bacterial strain and showed good antibacterial activity.

Compound **3c** also showed excellent antifungal activity as compared to standard reference amphotericin-B which is almost twice the activity of standard. Hence this compound can replace the standard drug amphotericin-B.



Graph 1: Biological Activities Of Esters Of 2-(3, 4-Dihydro-3-Oxo-2H-Benzo[B] [1,4]Thiazin-2-Yl)Acetic Acid

Table 2: *In Vitro* Activity f Esters of 2-(3, 4-Dihydro-3-Oxo-2H-Benzo[B][1,4]Thiazin-2-Yl)Acetic Acid Towards Bacteria And Fungi

Sr. No	Code	R	B.subtilis	S.aureus	P.aeruginosa	E. coli	C. albicans	A. niger
1	3a	C_6H_5	-	-	-	-	-	-
2	3b	C_3H_7	-	-	-	-	-	-
3	3c	C_6H_{12}	14.55	14.54	9.34	11.14	22.15	26.15
4	3d	C_5H_{11}	-	-	-	-	-	-
5	3e	CH_2CH_2Cl	-	-	-	-	-	-
6	3f	$C_6H_5CH_2$	-	-	-	-	-	-
7	3g	m-$CH_3C_6H_4$	-	-	-	-	-	-
8	3h	o-$CH_3C_6H_4$	-	-	-	-	-	-
9	3i	secC_3H_7	-	-	-	-	-	-
10	Ciprofloxacin(std.)		19.7	18.5	18.19	21.32	NA	NA
11	Amphotericin-B(std.)		NA	NA	NA	NA	11.12	11.92

Table 3: Physical Data Of Newly Synthesized Esters Of 2-(3, 4-Dihydro-3-Oxo-2H-Benzo[B][1,4] Thiazin-2-Yl) Acetic Acid

Compound	R	Mole. formula	Mol. wt	m. p.(^{0}c)	Yield%
3a	C_6H_5	$C_{16}H_{13}NO_3S$	299.34	160-164	75
3b	C_3H_7	$C_{13}H_{15}NO_3S$	265.33	112-116	79
3c	C_6H_{12}	$C_{16}H_{19}NO_3S$	305.39	122-126	83
3d	C_5H_{11}	$C_{15}H_{19}NO_3S$	293.38	70-74	69
3e	CH_2CH_2Cl	$C_{12}H_{12}ClNO_3S$	285.75	88-90	73
3f	$C_6H_5CH_2$	$C_{17}H_{15}NO_3S$	313.37	142-144	65
3g	m-$CH_3C_6H_4$	$C_{17}H_{15}NO_3S$	313.37	130-132	58
3h	o-$CH_3C_6H_4$	$C_{17}H_{15}NO_3S$	313.37	68-69	62
3i	secC_3H_7	$C_{13}H_{15}NO_3S$	265.33	135-137	82

Experimental

Phenyl 2-(3, 4-dihydro-3-oxo-2H-benzo [b][1,4]thiazin-2-yl)acetate(3a)

IR cm^{-1}, 3201(N-H), 1764 (CO ester), 1664.62 (CONH). ^{1}HNMR: ɗ 8.86 (s, 1H, NH), ɗ 7.5-6.9(m, 9ArH), ɗ 4.14 (t, 1H), ɗ 3.27 (d d, 1H), ɗ 2.86 (d d 1H).^{13}CNMR:168.71, 167.28, 150.49, 135.91, 129.41, 127.55, 125.99, 124.10, 121.41, 38.05. MS: 321.9, Anal. Calcd. for $C_{16}H_{13}NO_3S$ (299.34):C,64.20; H, 4.38;N,4.68; O, 16.03;S,10.71

Propyl 2-(3, 4-dihydro-3-oxo-2H-benzo [b][1,4]thiazin-2-yl)acetate(3b)

IR cm^{-1}: 3198.08(N-H), 1734.06(CO ester), 1672.34(CONH) ^{1}H NMR: ɗ 9.92 (s, 1H, NH) ɗ 7.3-6.9(m, 4Ar H), ɗ 4.11-4.08 (m, 1H, s 2H), ɗ 3.06 (d d, 1H) ɗ 2.(d d 1H), ɗ 1.64 (q, 2H), ɗ 0.96 (t, 3H).^{13}C NMR: 170.19, 167.60, 136.02, 127.42, 124.03, 66.79, 38.07, 21.94, 10.38.

Anal. Calcd. for $C_{13}H_{15}NO_3S$ (265.33):C,58.85; H, 5.70;N,5.28; O, 18.09;S,12.09

Cyclohexyl 2-(3, 4-dihydro-3-oxo-2H-benzo [b][1,4]thiazin-2-yl)acetate(3c)

IR cm^{-1}: 3198.08(N-H), 1755.28 (CO ester), 1650(CONH).^{1}H NMR: ɗ 9.7 (s, 1H, NH) ɗ 7.3-6.9(m, 4ArH), ɗ 4.05 (m, 1H), ɗ 4.0(m, 1H), ɗ 3.06(d d, 1H) ɗ 2.6 (d d 1H), ɗ 1.86-1.50 (m, 4H), 1.47-1.38 (m,6H). ^{13}C NMR: 169.47, 167.53, 136.0, 127.38, 124.03, 76.81, 38.20, 31.55, 31.50, 25.33, 23.67. Anal. Calcd. for $C_{16}H_{19}NO_3S$ (305.39):C,62.93; H, 6.27;N,4.59; O, 15.72;S,10.50

Pentyl 2-(3, 4-dihydro-3-oxo-2H-benzo [b][1,4]thiazin-2-yl)acetate(3d)

IR cm^{-1}: 3203.87 (N-H), 1735.99 (CO ester), 1672.34(CONH) ^{1}H NMR: ɗ 9.7 (s, 1H, NH) ɗ 7.3-6.9(m, 4ArH), ɗ 4.13 (m, 1H), ɗ 4.02(s, 2H) , ɗ 3.06 (d d, 1H) ɗ 2.63 (d d 1H), ɗ 1.33-1.302 (m, 4H), ɗ 1.66 (pentet, 2H), ɗ 0.92 (t, 3H).^{13}C NMR: 170.19, 167.18, 146.45, 127.43, 126.09, 125.11, 121.79, 65.37, 38.11, 28.32, 28.22, 22.49, 14.06 MS: 316.0. Anal. Calcd. for

$C_{15}H_{19}NO_3S$ (293.38):C,61.41; H, 6.53;N,4.77; O, 16.36;S,10.93

2-chloroethyl 2-(3,4-dihydro-3-oxo-2H-benzo [b][1,4]thiazin-2-yl)acetate(3e)

IR cm^{-1}: 3200. 01(N-H), 1741.78 (CO ester), 1666.55(CO-NH), ^{1}H NMR: ɗ 8.9 (s, 1H, NH) ɗ 7.38-6.9 (m, 4ArH), ɗ 5.1(t, 2H), ɗ 4.06 (t, 1H), ɗ 3.2 (t, 2H), ɗ 2.7 (d d 2H). ^{13}C NMR: 165.09, 162.35, 123.49, 123.31, 121.37, 72.09, 51.03, 38.30, 36.64 Anal. Calcd. for $C_{12}H_{12}ClNO_3S$ (285.75):C,50.44; H, 4.23;Cl, 12.41;N,4.90; O, 16.80;S,11.22

Benzyl 2-(3, 4-dihydro-3-oxo-2H-benzo [b][1,4]thiazin-2-yl)acetate(3f)

IR cm^{-1}: 3203. 87(N-H), 1743.71 (CO ester), 1664.62(CO-NH), ^{1}H NMR: ɗ 8.86 (s, 1H, NH) ɗ 7.4-6.9 (m, 9ArH), ɗ 5.2 (s, 2H), ɗ 4.06 (m, 1H), ɗ 3.15 (d d, 1H), ɗ 2.68 (d d 1H). ^{13}C NMR: 169.97, 166.91, 128.60, 128.38, 127.51, 124.09, 119.07, 66.99, 38.05. Anal. Calcd. for $C_{17}H_{15}NO_3S$ (313.37):C,65.16; H, 4.82;N,4.47; O, 15.32;S,10.23

m-tolyl 2-(3,4-dihydro-3-oxo-2H-benzo [b][1,4]thiazin-2-yl)acetate(3g)

IR cm^{-1}: 3209.66(N-H), 1751.42 (CO ester), 1664.62(CO-NH), ^{1}H NMR: ɗ 9.6 (s, 1H, NH) ɗ 7.37-6.9 (m, 8ArH), ɗ 4.14 (t, 1H), ɗ 3.27 (d d, 1H), ɗ 2.86 (d d 1H), ɗ 2.35(s, 3H). ^{13}C NMR: 168.87, 167.3, 150.50, 139.71, 129.19, 127.62, 126.87, 124.17, 121.13, 118.96, 34.40, 21.31. MS: 336. Anal. Calcd. for $C_{17}H_{15}NO_3S$ (313.37):C,65.16; H, 4.82;N,4.47; O, 15.32;S,10.23

o-tolyl 2-(3,4-dihydro-3-oxo-2H benzo [b][1,4]thiazin-2-yl)acetate(3h)

IR cm^{-1}: 3200.01(N-H), 1753.55 (CO ester), 1678.13(CO-NH), ^{1}H NMR: ɗ 8.2 (b s, 1H, NH) ɗ 7.37-6.9 (m, 8 Ar H), ɗ 4.11 (m, 1H), ɗ 3.3 (d d, 1H), ɗ 3.04(s, 3H), ɗ 2.6 (d d 1H). ^{13}C NMR: 176.4, 168.6, 149.18, 145.8, 131.1, 130.1, 127.6, 126.9, 126.2, 124.5, 121.8, 55.5, 35.1, 14.1.Anal. Calcd. for $C_{17}H_{15}NO_3S$ (313.37):C,65.16; H, 4.82;N,4.47; O, 15.32;S,10.23

Isopropyl 2-(3, 4-dihydro-3-oxo-2H-benzo [b][1,4]thiazin-2-yl)acetate(3i)

IR cm^{-1}: 3200.05(N-H), 1735.05 (CO ester), 1674.02(CO-NH), ^{1}H NMR: ɗ 9.4 (s, 1H, NH) ɗ 7.3-6.9 (m, 4 Ar H), ɗ 5.10-5.05, (septet1H), ɗ 4.0 (t, 1H), ɗ 3.03 (d d, 1H), ɗ 2.6 (d d 1H), ɗ 1.27 (d, 6H) ^{13}C NMR: 169.6, 167.23, 136.02, 128.07, 127.4, 123.9, 68.7, 55.6, 38.5, 21.8, 21.8.MS: 287.9, Anal. Calcd. for $C_{13}H_{15}NO_3S$ (265.33):C,58.85; H, 5.70;N,5.28; O, 18.09;S,12.09.

CONCLUSION

In summary, we have synthesized nine esters of 2-(3, 4-dihydro-3-oxo-2H-benzo[b] [1, 4] thiazin-2-yl) acetic acid successfully. All the synthesized compounds are characterized and also checked for the antibacterial and antifungal activity of these ester derivatives in vitro. The compound **3c** (cyclohexyl 2-(3, 4-dihydro-3-oxo-2H-benzo[b] [1,4] thiazin-2-yl)acetate) shows significant antibacterial activity and excellent antifungal activity compared to standard drug amphotericin-B. Further Molecular docking of all these compounds is done using alpha amylase responsible for microbial attack in silico. All the ester derivatives are good inhibitors of alpha amylase. Parent acid (2-(3, 4-dihydro-3-oxo-2H-benzo[b] [1, 4] thiazin-2-yl) acetic acid) showed better binding score as compared to its newly synthesized ester derivatives. Among all the ester derivatives, compound **3c** showed good binding interactions with alpha amylase.

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