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"Synthesis, physiochemical evaluation, characterization, and anticancer activity ofderivatives containing 4-((2,3-dihydro-4H-1,4-oxazin-4-yl) sulfonyl) aniline"

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Abstract
Medicinal chemistry focuses on the exploration and advancement of new pharmaceuticals forthe treatment of diseases. The primary objective of the field is to create innovative organic molecules, whether they are naturally occurring or artificially synthesised. Heterocyclic nuclei are a substantial part of these chemical molecules. Sulphonamide based acetamide molecules are currently being utilised in the development of novel anticancer medicines. The physicochemical characteristics of the synthesised substance (SA-1 to SA-11) were documented. The chemical SA-1 to SA-11 has been characterised using infrared spectroscopy(IR), proton nuclear magnetic resonance (1HNMR), carbon-13 nuclear magnetic resonance (13CNMR), and mass spectral analysis. Additionally, all the other synthesised compounds haveexhibited a molecular ion peak that is analogous to their respective molecular formula and weight. The synthesised compounds were assessed for their anticancer activity against the NCI-H226 lung cancer cell line using the SRB test technique. The MTT experiment indicated that compounds SA-1, SA-2, SA-3, SA-4, and SA-11 had significant efficacy against the Lung cancer cell line. It demonstrated a significant cytotoxic effect on both cell types, particularly on the WI-38 normal lung fibroblast cells and NCI-H226 lung cancer cells. Evidently, all compounds that were examined demonstrate a notable induction of cell death, except for SA- 1, SA-2, SA-3, SA-4, SA-5, and SA-11, which had IC50 values of 1866.20, 1702.23, 1374.35, 1625.10, 1200, and 2959.16 µg/ml, respectively.

Keywords: Carcinogenesis, NCI-H226, DHFR, radiotherapy**,** neuroendocrine carcinoma, ect.

1. Introduction

Cancer remains to be the leading cause of death in humans second only to cardiovascular diseases and more than 70% of all cancer deaths occur in developing and under-developed countries. There is a continuous rise of deaths from various cancers worldwide, with an estimated 12 million deaths in 2030.[1] Despite the advancement in the knowledge of biochemical processes associated with carcinogenesis, the successful treatment of cancer remains a significant challenge because of the general toxicity associated with the clinical useof traditional cancer chemotherapeutic agents. Hence, the design and development of new drugs for cancer therapeutics remains to be an important and challenging task for medicinal chemists worldwide. [2] Cancer can be considered a general term that covers a plethora of different malignancies. These pathogenic conditions are characterized by uncontrolled cellularproliferation andgrowth, and under special conditions, tumor cell migration, invasion, and spreading tootherorgans and tissues occur. Different factors and conditions can transform normal cells intocancerous ones by altering the normal function of a wide spectrum of regulatory, apoptotic,and signal transduction pathways. [3] This is called loss of differentiation(Fig. 1).

Fig.1 Diagrammatic represent the present of cancer cell in human lungs

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Numerous genes andproteins that are causally involved in the initiation and progression of cancer have beenidentified in the past few years. Cancer is the result of multiple mutations thatoccur in oncogenes, tumor suppressorsand/or DNA repair genes of somatic cells. Cancer is a multifactorial disease, in which both environmental and genetic factors play arole.[2] Risk factors in cancer etiology comprise four classes of external agents incarcinogenesis (carcinogens): physical, chemical, biological agents, and diet. [4]

2. Introduction of Lung Cancer

Lung cancer is a type of cancer that starts in the lungs. It causes cells to divide in the lungs uncontrollably and form tumors to reduce a person's ability to breathe. Worldwide, about three quarters of lung cancers are attributable to smoking; others are caused by occupational workplace exposure, radon exposure, and air pollution (Fig.2). It is more common in men, and incidence increases with age. [5] Lung cancer is a heterogeneous disease comprising several subtypes with pathologic and clinical relevance. Small-cell lung carcinoma (SCLC, 15% of all lung cancer) and non-small-cell lung carcinoma (NSCLC, 85%) are the two major forms of lung cancer. In this article, we list part of targets involved in lung cancer based on the information provided by NGC. Lung cancer is a type of cancer that starts in the lungs. Cancerstarts when cells in the body begin to grow out of control. Lungs are 2 sponge-like organs in chest.

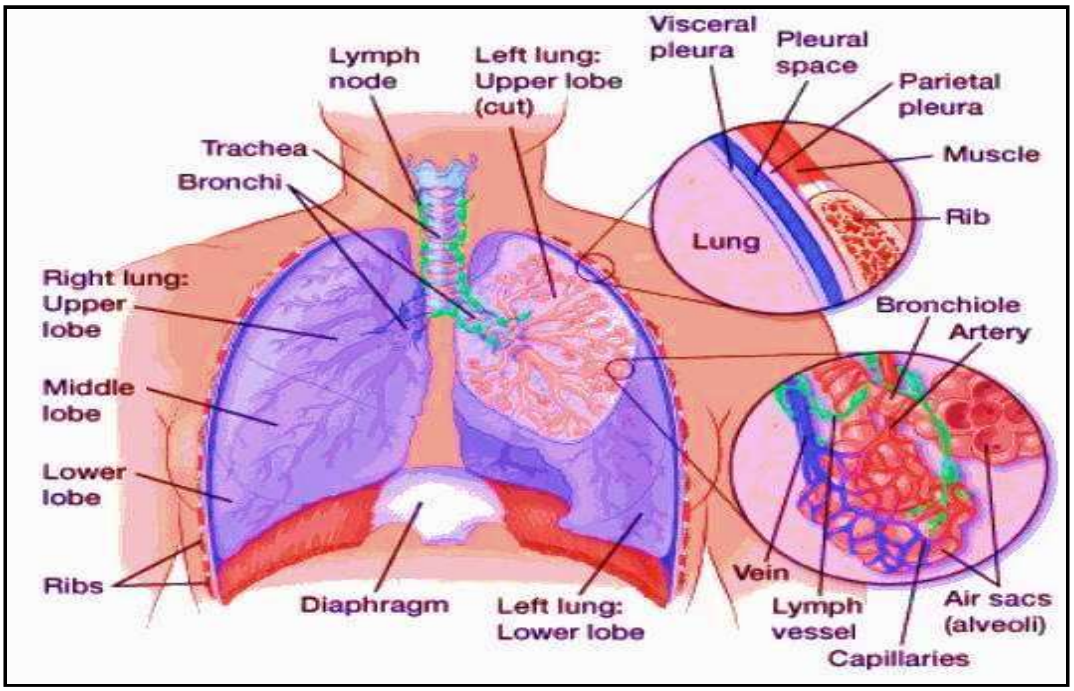

Fig.2: Normal lung description

3. Types of lungs cancer

Adenocarcinoma: Adenocarcinomas start in the cells that would normally secrete substances such as mucus. This type of lung cancer occurs mainly in people who currently smoke or formerly smoked, but it is also the most common type of lung cancerseen inpeople who don't smoke. It is more common in women than in men, and it is more likelyto occur in younger people than other types of lung cancer.[10]

Squamous cell carcinoma: Squamous cell carcinomas start in squamous cells, which are flat cells that line the inside of the airways in the lungs. They are often linked to a history of smoking and tend to be found in the central part of the lungs, near a main airway (bronchus).

Large cell (undifferentiated) carcinoma: Large cell carcinoma can appear in any partof the lung. It tends to grow and spread quickly, which can make it harder to treat. A subtype of large cell carcinoma, known as **large cell neuroendocrine carcinoma**, is afast-growing cancer that is very similar to small cell lung cancer.[11]

Small cell lung cancer (SCLC): About 10% to 15% of all lung cancers are SCLC andit is sometimes called **oat cell cancer.** This type of lung cancer tends to grow and spreadfaster than NSCLC. About 70% of people with SCLC will have cancer that has alreadyspread at the time they are diagnosed. Since this cancer grows quickly, it tends to respond well to chemotherapy andradiation therapy. Unfortunately, for most people, the cancer will return at some point. [12]

4. Rational for Chemoradiotherapy

The combination of radiotherapy and chemotherapy is an appealing approach that has ledto improved treatment results in patients with advanced solid tumors. In particular, theconcomitant use of radiotherapy and chemotherapy resulted in

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a lower recurrence rate and provided good local control for carcinoma and thus higher organ preservation rate.[3]The combination of radiotherapy and chemotherapy is mostly advocatedbecause of itsindependent cell-killing effect. Such that radiotherapy is aimed at controlling the primarytumor, while chemotherapy is used to eradicate distant metastases.A more attractive concept is the exploitation of the ability of chemotherapeutic agents tosensitize radio-resistant tumors to the lethal effect of ionizing irradiation under reduced oxygen conditions.[13]

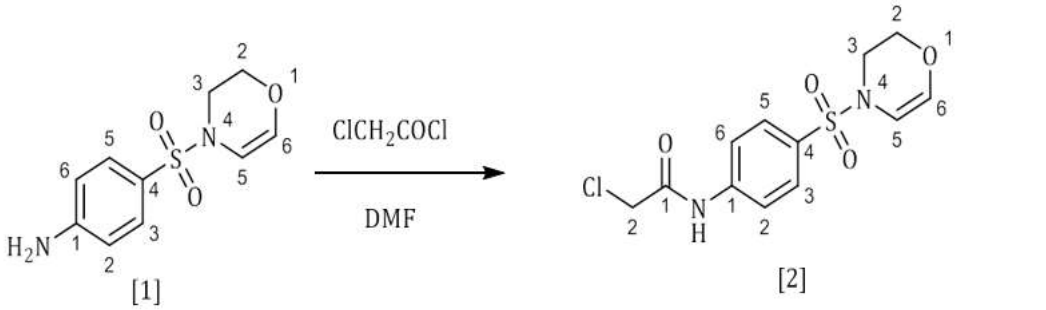
5. Medication used in cancer therapy

The antitumor activity is accomplished by the sulfonamides throughdissimilar of mechanisms, such as histone deacetylases (HDACs) inhibition,cell cycle arrest in the G1 phase, NADH oxidase inhibition, carbonic anhydrase (CA) inhibition, matrix metalloproteinase (MMPs) inhibition, cyclin-dependentkinase (CDK) inhibition, methionine aminopeptidases (MetAPs) inhibition, binding to b-Tubulinand disruption of microtubule assembly. [14] Thiazole ring asa core structural pattern found in avariety of biologically and pharmacologically activemolecules, also,it is a structural constituent of natural products such as thiamine (vitamin B1) and penicillin. In addition, thiazole derivatives demonstrated a broad spectrum of medicinal and biological activities, including antiviral, antimicrobial, anti-inflammatory, antimalarial, anti-HIVand anticanceractivities. Asan epitome heterocyclic-amines, 2-aminothiazoles and their derivativesare used as key intermediates for the synthesis of plentiful biologicallyactive compounds, such as biocides, fungicides, sulfur drugs, and asintermediates in the synthesis ofnumerous antibiotics, where a hugenumber of 2-aminothiazoles have been substituted with different groups for pharmaceutical applications. [15]. The acetamid functional group is responsible for urease inhibitory activities, antimicrobial,antioxidant, anti-inflammatory and platelet aggregation inhibitory.Thesulfonamide group linked with acetamide moiety bearing different aryl,heteroaryl substituents exhibits enormous pharmacologicalpotency,particularly sulfonamide derivatives encompassing shortamine fragments reveal promising anticancer activity.[16] Dihydrofolate reductase enzyme (DHFR) is a key enzyme in theprocess of nucleic acid synthesis in both human and bacteria. This enzyme is accountable for catalysis of the reduction of folate ordihydrofolate to tetrahydrofolate using NADPH. This function made oftheDHFR is considered as an important target for different antibacterial andcancer agent.[17]. Otherthan the well-established fluoro-nucleosides such as 5-fluoro uracil, the fluorine containing anticancer molecules include flutamide, an anti-androgen which was launched in 1983 for the treatment of prostate cancer and fluorinated anthracycline antibiotics, steroids, Vitamin D3 analogs and fluorine containing molecules have been shown to be much more effective than their parent analogs. [18]. Also, some structurally novel sulfonamide derivatives have recentlybeen reported to show substantial antitumor activity *in-vitro* and/or *in-vivo*. (E7010)
,(ER-34410) and (E7070, Indisulam) are examples for antitumor sulfonamides in advanced clinical trials.[20]

6. Experimental ProcedureMaterials and method

Different substituted 4-((2,3-dihydro-4H-1,4-oxazin-4-yl) sulfonyl) aniline was purchased from Merck, India. The different 4-Substitutedthiazol-2-amine, 2chloroacetyl chloride, and Dimethyl formamide was purchased from sigma Aldrich. All the chemicals were purchased from Sigma Aldrich and Merck India. Commercial grade solvents used for the reactions were distilled before use.The melting points of the synthesized compounds were determined in openglass capillaries. IR spectra were recorded on Bruker-alpha FTIR spectrometer. Elemental analysis was performed and found values were within 0.4% of theoretical values. 1HNMR spectra were recorded at 400 MHz, Mass Spectra were recorded using Mass Spectrometers JeolFSX-112 (FAB) by ESI.

(A) of 2-chloro-N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl) acetamide



[1] 4-((2,3-dihydro-4*H*-1,4-oxazin-4-yl)sulfonyl)aniline

[2] 2-chloro-*N*-(4-((2,3-dihydro-4*H*-1,4-oxazin-4-yl)sulfonyl)phenyl)acetamide

Procedure: A mixture 4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)aniline **[Compound 1]**(0.1 mol) and 2-chloroacetyl chloride(0.1 mol) was dissolved indimethyl formamide(DMF; 20 ml) and magnetic stirred at room temperature for 2 hrs.

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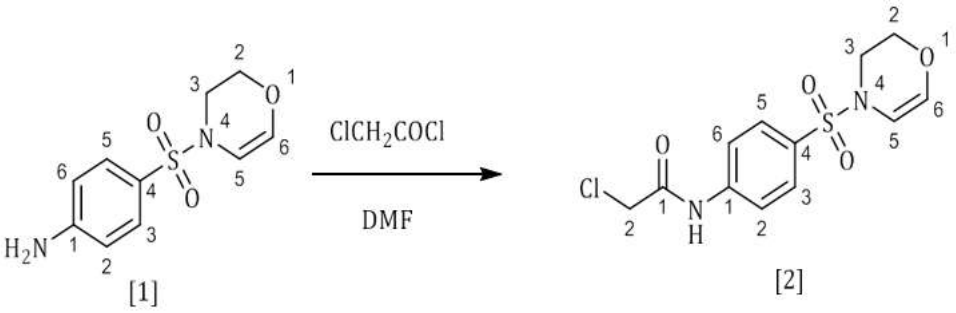
The reaction was monitored by TLCmethod using n-Hexane: ethyl acetate (2:1) as solvent system. The reaction mixture was poured onto ice cold distilled water.[53] The obtained solid was filtered off and crystallized from ethanol to form 2-chloro-N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl) acetamide **[Compound 2]**.

(B) Synthesis of N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)-2-((4-substituted thiazol-2-yl)amino)acetamide



4-((2,3-dihydro-4*H*-1,4-oxazin-4-yl)sulfonyl)aniline

2-chloro-*N*-(4-((2,3-dihydro-4*H*-1,4-oxazin-4-yl)sulfonyl)phenyl)acetamide

Procedure: The2-chloro-N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl) acetamide(compound 2) and different 4-substitutedthiazol-2-amine(0.01 mol) was dissolved in absolute ethanol was refluxed for 4-6 h. The reaction mixtures wereconcentrated under reducedpressure using rota-evaporator to obtained solid was filtered, washed withn-hexane, dried and recrystallized from ethanol to give the **Compound 3,**N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)-2-((4substituted thiazol-2-yl)amino)acetamide.

7. Characterization of the synthesized compoundsTable 1. List of Final synthesized compounds

SN	Code	Chemical name
1.	SA-1	2-((4-chlorothiazol-2-yl)amino)-N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)acetamide
2.	SA-2	2-((4-bromothiazol-2-yl)amino)-N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)acetamide
3.	SA-3	N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)-2-((4-nitrothiazol-2-yl)amino)acetamide
4.	SA-4	N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)-2-((4-methylthiazol-2-yl)amino)acetamide
5.	SA-5	N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)-2-((4-methoxythiazol-2-yl)amino)acetamide
6.	SA-6	N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)-2-((4-ethylthiazol-2-yl)amino)acetamide
7.	SA-7	N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)-2-((4-ethoxythiazol-2-yl)amino)acetamide
8.	SA-8	N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)-2-((4-propylthiazol-2-yl)amino)acetamide
9.	SA-9	N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)-2-((4-propoxythiazol-2-yl)amino)acetamide
10.	SA-10	2-((4-aminothiazol-2-yl)amino)-N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)acetamide
11.	SA-11	N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)-2-((4-fluorothiazol-2-yl)amino)acetamide

Table.2 Physicochemical properties of the synthesized compounds

SN	Code	Chemical formula	Mol. Weight	Percent Yield	Melting point
1.	SA-1	$C_{15}H_{15}ClN_4O_4S$	414.88	88%	125-127°C
2.	SA-2	$C_{15}H_{15}BrN_4O_4S_2$	459.33	76%	132-134°C
3.	SA-3	$C_{15}H_{15}N_5O_6S_2$	425.43	82%	145-147°C

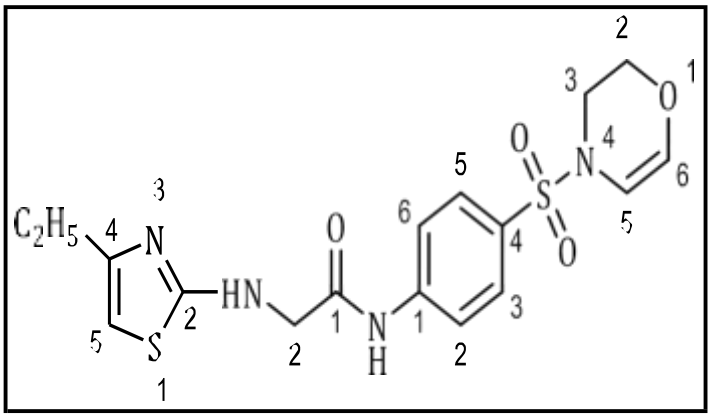
4.	SA	$C_{16}H_{18}N_4O_4S_2$	394.46	70%	114-116°C

5.	SA-5	$C_{16}H_{18}N_4O_5S_2$	410.46	68%	122-124°C
6.	SA-6	$C_{17}H_{20}N_4O_4S_2$	408.49	62%	112-114°C
7.	SA-7	$C_{17}H_{20}N_4O_5S_2$	424.49	65%	150-152°C
8.	SA-8	$C_{18}H_{22}N_4O_4S_2$	422.52	60%	138-140°C
9.	SA-9	$C_{18}H_{22}N_4O_5S_2$	438.52	69%	115-117°C
10.	SA-10	$C_{15}H_{17}N_5O_4S_2$	395.45	70%	162-164°C
11.	SA-11	$C_{15}H_{15}FN_4O_4S_2$	398.43	65%	193-195°C

8. Characterization of the synthesized compounds

Characterization by IR, NMR, mass spectral, and elemental studies were used to characterize the target structures of the synthesized compounds. The 2-chloroacetyl chloride, and dimethylformamide reacted with the 4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)aniline**(Compound 1)** to form 2-chloro-N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl) phenyl) acetamide **[Compound 2]**. The Compound 2 obtained as white crystal product having the melting point 180-190°C. The FT-IR spectrum of compound 2, denotes the characteristics peak of NH at 3296, peak of C-H aromatic at 3120, CH aliphatic at 2950, 1688 C=O at 1688 as well as 1HNMR denotes the characteristics peak of N-H at 8.53 ppm. The 13CNMR spectra shown the peak of C=O at 164.3, CH_2 at 66.1, CH_2 at 46.2,

COMPOUND 1



IUPACname: 2-((4-chlorothiazol-2-yl)amino)-N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)acetamide
Chemical Formula: $C_{15}H_{15}ClN_4O_4S_2$**;**

Molecular Weight: 414.88

Elemental Analysis:

Elements	**C**	**N**	**O**	**S**
Calculated	43.43	13.50	15.43	15.46

Found	43.30	13.48	15.42	15.45

IR (cm^{-1})

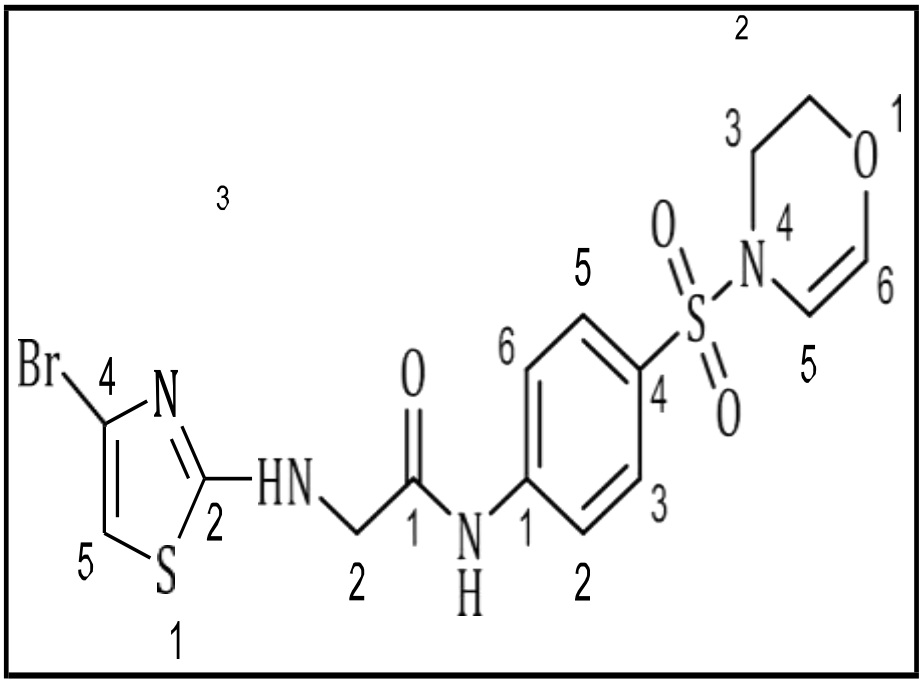
Serial Number	Propagation number (cm^{-1})	Mode of vibration
1.	3331	(N-H)
2.	3118	(N-H)
3.	3039	(Aromatic C-H)
4.	1672	(C=O)
5.	2938	(C-H aliphatic)
6.	850	(C-Cl)

1HNMR (ppm):
δ 10.23 (s, 1H, NH), 5.95 (s, 1H, NH), 6.28 (s, 1H, thiazole), 4.20 (s, 2H, CH_2), 3.25 (s, 2H,CH_2), 2.80–2.82 (m, 4H, $2CH_2$), 1.32–1.35 (m, 2H, CH_2).

13CNMR (ppm):
170.8 (C=O), 162.5 (C=N), 142.6 (C), 130.2 (C-H), 128.6 (C-H), 118.6 (C-H), 105.3 (C-H), 45.2 (CH_2), 42.0 (CH_2), 24.6 (CH_2), 22.1 (CH_2).

FAB Mass (m/z): 414.02

COMPOUND 2



IUPACNAME:2-((4-bromothiazol-2-yl)amino)-N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)acetamide

Chemical Formula: C15H15BrN4O4S2;

Molecular Weight: 459.33

Elemental Analysis:

Elements	C	N	O	S
Calculated	39.22	17.40	12.20	13.96

Found	39.20	17.38	12.18	13.92

IR (cm^{-1})

Serial Number	**Propagation number (cm^{-1})**	**Mode of vibration**
1.	3329	(N-H)
2.	3112	(N-H)
3.	3042	Aromatic C-H,
4.	1697	(C=O)
5.	2945	(C-H aliphatic)
6.	1018	(C-Br)

1HNMR (ppm):
δ 10.25 (s, 1H, NH), 5.97 (s, 1H, NH), 6.25 (s, 1H, thiazole), 4.25 (s, 2H, CH_2), 3.28 (s, 2H,CH_2), 2.82–2.84 (m, 4H, $2CH_2$), 1.30–1.32 (m, 2H, CH_2)

13CNMR (ppm):
d = 170.2 (C=O), 162.3 (C=N), 142.5 (C), 130.4 (C-H), 127.9 (C-H), 118.9 (C-H), 105.2 (CH), 45.1 (CH2), 41.9 (CH2), 24.8 (CH2), 22.3 (CH2) ppm.

FAB Mass (m/z): 498.97 and 42.8.
The final compound (N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)-2-((4- substituted thiazol-2-yl)amino)acetamide) 3, was synthesized by the reaction of compound 2 with different 4-substituted thiazol-2-amine. The physicochemical properties of the synthesized compound (SA-1 to SA-11) was represented in Table 4.4. The compound SA-1 toSA-11 has characterized by the IR, 1HNMR, 13 CNMR and Mass spectral analysis. The IR spectrum of the compounds has shown the characteristics peak (cm^{-1}) of N-H peak at 3331, N-H peak at 3118, aromatic C-H peak at 3039, C=O peak at 1672, C-H aliphatic peak at 2938, C-Cl peak at 850, C-Br peak at 1018, C-F peak at 1102, N-O peak at 1358 and N=O peak at 1562.The 1HNMR spectra of Synthesized compounds depicted the peak of N-H at 10.25 ppm, Thiazole-H peak at 6.25 ppm, CH_2 peak at 4.19. The 13 CNMR spectrum of synthesized compound (SA-1 to SA-11) denotes the peak in ppm of C=O at 170.5, C=N at 162.3 and CH_2 at 45.6.

9. Pharmacological Evaluation of Synthesize Compound
The *in-vitro* anticancer screening was done by the pharmacology unit at the NIPER, Mohali.The *in-vitro* cytotoxic activity was measured for all the newly synthesized compoundson Lung squamous carcinoma cell line (NCI-H226) by applying the Sulfo- Rhodamine-B stain (SRB) assay.Paclitaxel was chosen as a reference and standard anticancerdrug due to its potency againstNCI-H226.The eleven compound (SA-1 to SA-11) was synthesized and evaluated for the anticancer potential by SRB assay. All the synthesized compounds were evaluated for their anticancer activity against Lung cancer cell line (NCI- H226) by SRB assay method. All compounds showed anticancer activity but differ in potencyas compare to standard drug paclitaxel. The data of *In-vitro* anticancer activity of the synthesized compounds (SA-1 to SA-11) was represented in Table 3. The graphical representation was shown in Figure2 .

Table 3: *In-vitro* anticancer screening of the synthesized compounds (SA-1 to SA-11) against Lung cancer cellline (NCI-H226) at three different concentrations.

Compounds	**Compound concentration (µM)**				
	10 (µM)	**25 (µM)**	**50 (µM)**	**100 (µM)**	**IC 50 (µM)**
	Surviving fraction (mean ± SE)[a]				
Paclitaxel	0.525±0.022	0.435±0.007	0.320± 0.012	0.214 ± 0.016	31.1
SA-1	0.422±0.006	0.225±0.009	0.371± 0.005	0.345 ± 0.011	07.7
SA-2	0.810±0.018	0.548±0.012	0.331± 0.008	0.350 ± 0.015	46.0
SA-3	0.385±0.021	0.251±0.021	0.355± 0.004	0.290 ± 0.009	09.5
SA-4	0.528±0.015	0.328±0.025	0.313± 0.014	0.381 ± 0.007	14.7
SA-5	0.790±0.024	0.545±0.032	0.439± 0.005	0.415 ± 0.006	37.5
SA-6	0.855±0.012	0.608±0.012	0.360± 0.014	0.350 ± 0.009	50.5
SA-7	0.856±0.015	0.615±0.015	0.532± 0.011	0.340 ± 0.015	51.6
SA-8	0.823±0.018	0.588±0.014	0.435± 0.009	0.420 ± 0.018	40.3

SA-9	0.903±0.023	0.612±0.017	0.413± 0.021	0.395 ± 0.021	50.8
SA-10	0.825±0.012	0.668±0.015	0.307± 0.015	0.271 ± 0.017	55.4
SA-11	0.541±0.009	0.375±0.014	0.360± 0.016	0.460 ± 0.015	08.1

[a]Each value is the mean of three experiments± standard error

The result data of the synthesized compounds by SRB assay stated the IC50 value of compounds SA-1 (7.7), SA-3 (9.5) , SA-4 (14.7) and SA-11 (8.1) has shown better activity as compared to the standard drug paclitaxel (31.1) and it suggested that Cl, Br, NO_2 and F compounds enhance the activity when it attached to 4-position of the thiazole ring as well as presence of sulfonamide bearing thiazole with addition to electronegate atom enhance the anticancer activity of compounds.

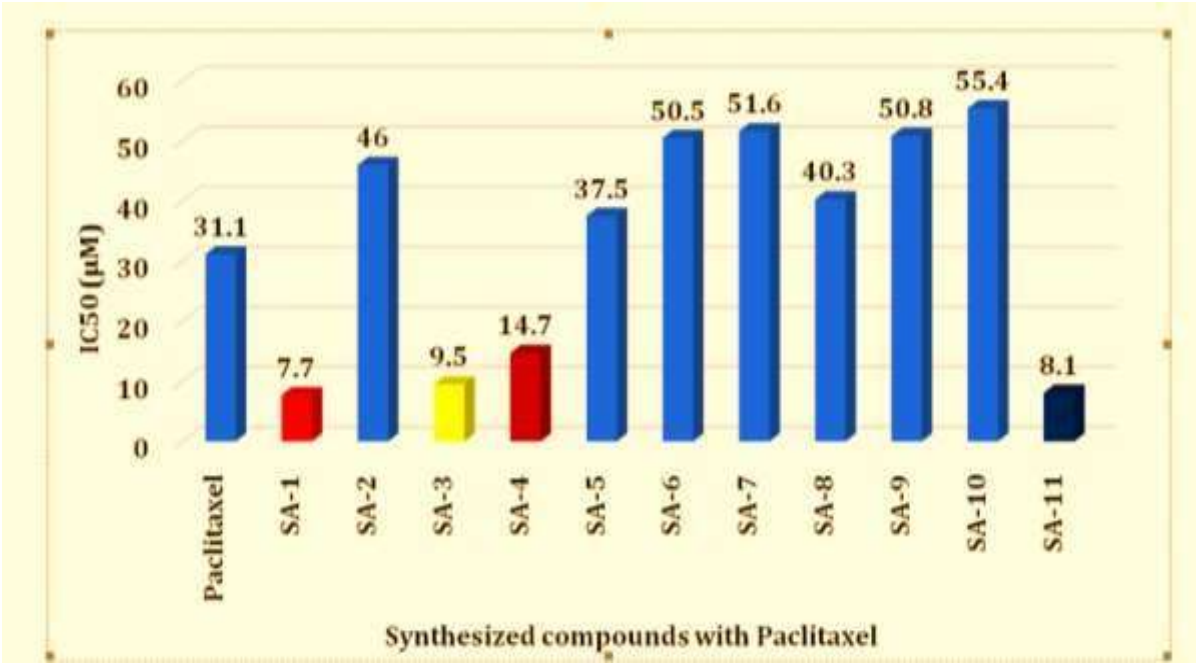


Fig..2 : Graphical representation of IC50 value of synthesized compound (SA-1 to SA-11). MTT assay suggested that the compound SA-1, SA-2, SA-3, SA-4 and SA-11 has shown the prominent effectiveness against the Lung cancer cell line and also depicted that prominent activity against WI-38.

10. Results and Discussion

Medicinal chemistry is concerned with the discovery and development of novel drugs to cure ailments. The majority of the discipline's activities are focused on developing novel natural orsynthetic organic molecules. Heterocyclic nucleus containing moieties make up a significant component of these organic compounds. New anticancer drugs are being developed using sulphonamide based acetamide compounds. Fluorine-containing compounds have attracted much interest since the introduction of fluorine atoms or fluoroalkyl moieties to an organic compound can bring about remarkable changes in the physical, chemical, and biological properties. The final compound (N-(4-((2,3-dihydro-4H-1,4-oxazin-4-yl)sulfonyl)phenyl)-2- ((4-substituted thiazol-2-yl)amino)acetamide) 3, was synthesized by the reaction of compound

2 with different 4-substituted thiazol-2-amine. The physicochemical properties of the synthesized compound (SA-1 to SA-11) was represented in Table 2. The compound SA-1 to SA-11 has characterized by the IR, 1HNMR, 13 CNMR and Mass spectral analysis. The IR spectrum of the compounds has shown the characteristics peak (cm^{-1}) of N-H peak at 3331, N-H peak at 3118, aromatic C-H peak at 3039, C=O peak at 1672, C-H aliphatic peak at 2938, C-Cl peak at 850, C-Br peak at 1018, C-F peak at 1102, N-O peak at 1358 and N=O peak at 1562. The 1HNMR spectra of Synthesized compounds depicted the peak of N-H at 10.25 ppm, Thiazole-H peak at 6.25 ppm, CH_2 peak at 4.19. The 13 CNMR spectrum of synthesized compound (SA-1 to SA-11) denotes the peak in ppm of C=O at 170.5, C=N at 162.3 and CH_2 at 45.6. COMPOUND 3 (SA-1), mass spectrum has shown peak at m/z = 414.02, which matches the chemical formula $C_{15}H_{15}ClN_4O_4S_2$. All the other synthesized compound has also shown the molecular ion peak similar to their molecular formula and weight. All the synthesized compounds were evaluated for their anticancer activity against Lung cancer cell line (NCI-H226) by SRB assay method. All compounds showed anticancer activity but differ in potencyas compare to standard drug paclitaxel. MTT assay suggested that the compound SA-1, SA-2, SA-3, SA-4 and SA-11 has

shown the prominent effectiveness against the Lung cancer cell line and also depicted that prominent activity against WI-38. Compound (SA-1 to SA-11) tested against WI-38 normal lung fibroblast cells, NCI-H226 lung cancer cells and exert a prominently cytotoxic influence on the WI-38 normal lung fibroblast cells and NCI- H226 lung cancer cells. Paclitaxel a well-known chemotherapeutic agent (IC50 = 41 and 6.25µg/mL for WI-38 and NCI-H226respectively) was used as the reference control. The cell viability of WI-38 under different concentration and IC50 calculations for each compound, clearly show that all tested compounds induce a significant cell death except SA-1, SA-2, SA-3, SA-4, SA-5 and SA-11 with IC_{50} value of 1866.20, 1702.23, 1374.35, 1625.10, 1200 and 2959.16 µg/ml, respectively. In case to NCI-H226 cancer cell line, cell viability and IC50 calculations prove that synthesized compounds have an obvious cell death except compound SA-9 and SA-10 with IC50 value of 122.5 and 151.3 µg/ml, respectively. From screening results shown in Table 3, it can figure out that compounds SA-1 (IC50 = 15.10), SA-2 (IC50 =22.65), SA-3 (IC50 = 19.15), SA-4 (IC50 = 32.65) and SA-11 (IC50 = 16.10) have a potential
anti-cancer activity compared with Paclitaxel.

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