

# Synthesis, Spectral Characterization, Docking Studies and QSAR Screening of 4-amino-benzenesulfonamides/N-acetyl 4-amino-benzenesulfonamide Derivatives as Antimicrobial Agents

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**Abstract:** A series of substituted 4-amino-benzenesulfonamides / N-acetyl-4-amino-benzenesulfonamide were designed & synthesized keeping in view the structural requirements of pharmacophore and were evaluated for *in-silico* antimicrobial activity. For establishing the structure, spectral characterization like FT-IR, <sup>1</sup>H NMR, GC-MS and elemental analysis (CHNS) has been performed. The antimicrobial activity of the titled compounds was assessed using *in-silico* studies (QSAR screening and Docking). It was carried out for the prediction of pharmacokinetic properties and to study the binding properties of drugs with molecular targets. Titled compounds exhibited good binding properties with molecular target. It could be concluded that molecular target responsible for the antimicrobial activity of substituted 4-amino-benzenesulfonamides / N-acetyl-4-amino-benzenesulfonamides may be pseudomonas aeruginosa exotoxin A.

**Keywords:** Sulfonamide, *in-silico* studies, statistics, quantitative structure activity relationship.

## INTRODUCTION

With serious mortality and morbidity results, drug resistance against bacteria have emerges with public health problem all over the world. The case of penicillin resistance worldwide could be considered as one of the example. Additionally, multi-drug resistance has created another problem to work with. This type of problem could be observed in Europe, Asia and America with vancomycin resistance [1-8]. The above mentioned problems and so many others like these promoted us to contribute hands a little towards solving the problems by synthesizing and screening a series of sulfonamide derivatives.

Mode of action of sulfonamide drugs observed so far is inhibition of carbonic anhydrase against a wide range of bacteria. The substituted ring of benzenesulfonamide containing –SO<sub>2</sub>NH<sub>2</sub> groups act by binding or coordination of the –SO<sub>2</sub>NH<sup>-</sup> anion to the Zn<sup>2+</sup> of the enzyme, mimicking the bicarbonate anion in the transition state [9]. The mode of action of antimetabolite sulfa drugs is the inhibition of dihydropteroate synthetase, which catalyzes an enzyme in the biosynthesis of tetrahydrofolate and then nucleotides [10].

In the same way, we have screened (*in-silico* screening) the designed compounds against Pseudomonas aeruginosa exotoxin A, Bacillus subtilis

lipase A, E-coli primosomal protein, heterodimeric hexaprenyl diphosphate synthase, Staphylococcus aureus metalloproteinase and Bacillus subtilis Lon protease.

In this study, the designed sulfonamide derivatives were synthesized, characterized and screened (*in-silico* screening with docking and QSAR study) against antimicrobial activity.

## EXPERIMENTAL

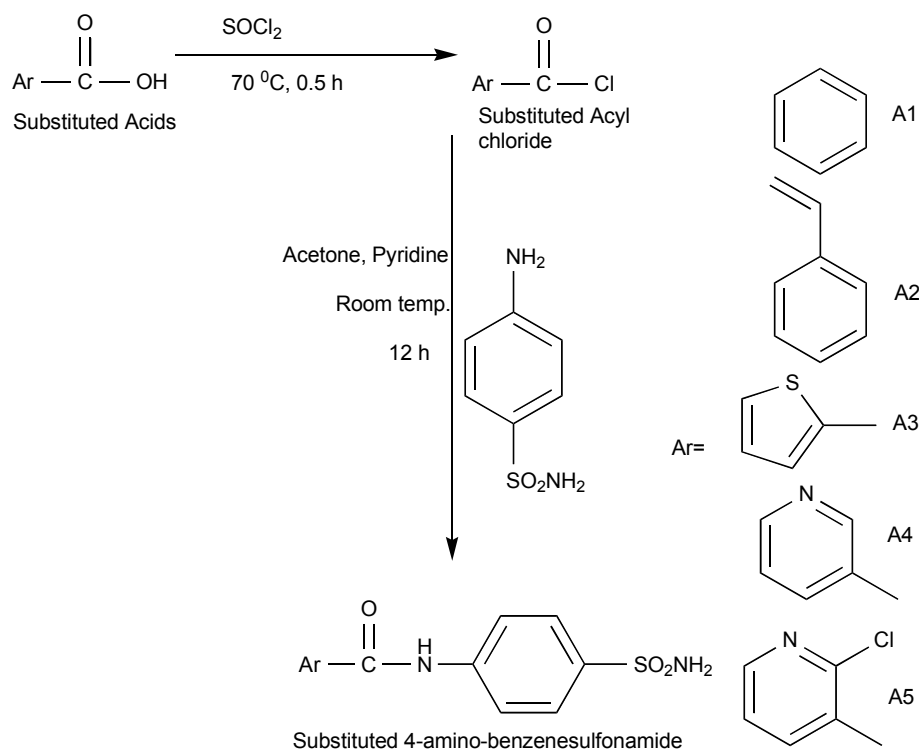
### Material and Methods

#### Synthesis and Spectral Characterization

All the chemicals and solvents, purchased from Merck (India), Spectrochem (India), Sigma-Aldrich (India), Himedia (India) and S. D. Fine were used without further purification. Thin layer chromatographic analysis of compounds was performed on silica gel G coated glass plates. The adsorbent silica gel G was coated to a thickness of about 0.3 mm on previously cleaned TLC plates of 20x5 cm using conventional spreader. The plates were placed in hot air oven at 105° C for 30 min. The solutions of compounds were applied as a spot on the activated plate about 2 cm above from the lower edge. The mobile phases were selected according to the polarity of compounds.

Melting points were determined by using open capillary melting point apparatus and are reported uncorrected. FT-IR spectra (KBr) were recorded on a Perkin-Elmer Spectrometer BX-II spectrophotometer. The <sup>1</sup>H-NMR spectra were recorded on Bruker 400 MHz High Resolution NMR spectrometer using TMS as

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**Scheme 1:** Synthetic scheme of substituted 4-amino-benzenesulfonamide from substituted acids (A1, A2, A3, A4, A5).

an internal standard. Chemical shifts were reported in ppm ( $\delta$ ) and signals were described as singlet (s), doublet (d), triplet (t) and multiplet (m). The mass spectra were recorded on a Waters Micro-Mass ZQ 2000 mass spectrometer.

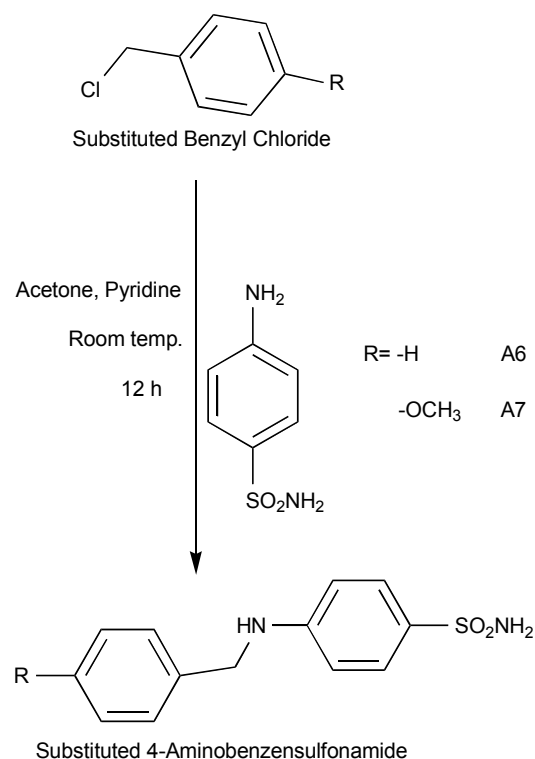
### Synthesis of Substituted Acyl Chlorides

Substituted acid (0.1 mol) and thionyl chloride (0.4 mol) were placed in a 250 ml flask equipped with a magnetic stirrer bar and a condenser with a drying tube. The reaction mixture was stirred and heated in a 70 °C oil bath. After 0.5 hours, the reaction mixture was allowed to cool at room temperature with opened flask; this facilitates the evaporation of remaining thionyl chloride and leaves acyl chloride in the flask [11, 12].

### Synthesis of Substituted 4-amino-benzenesulfonamides/N-acetyl-4-amino-benzenesulfonamide

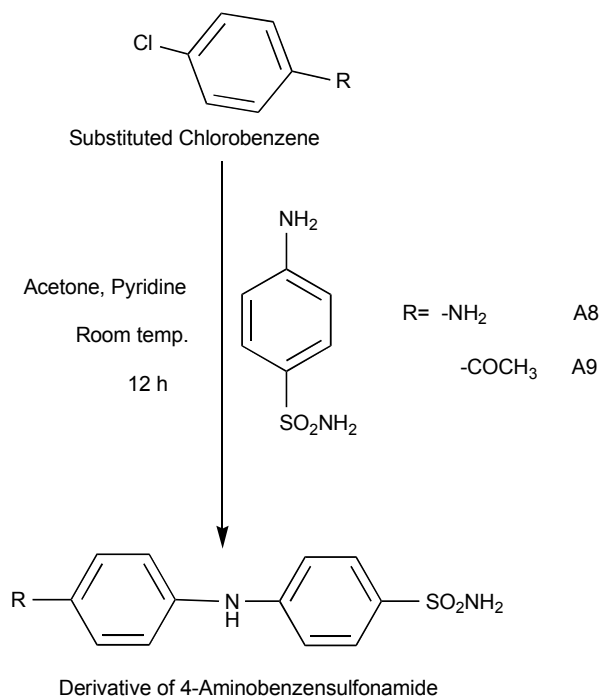
For the synthesis of an appropriate amide, the substituted acyl chloride/substituted benzyl chloride (0.009 mol) of an individual acid dissolved in 20 ml. of dry acetone was added dropwise to a stirred solution of suitable aromatic aminosulfonamide (0.0092 mol) and pyridine (0.0091 mol) in 50 ml. of dry acetone. After addition, the reaction mixture was stirred for about 12 hour at room temperature and then the solvent was evaporated under reduced pressure. The residue was dissolved in 100 ml. ethyl acetate and the organic phase washed three times with 20 ml. of distilled water.

Then 10% HCl solution was added until pH 1 was reached, and the organic phase was separated from the aqueous phase and washed three times with brine.

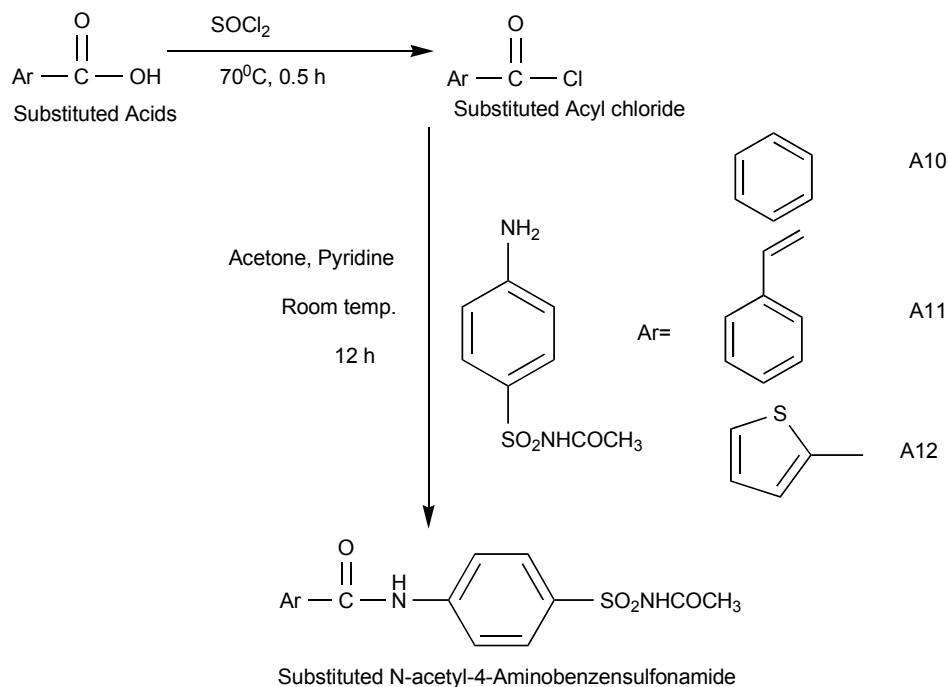


**Scheme 2:** Synthetic scheme of substituted 4-amino-benzenesulfonamide from substituted benzyl chloride (A6, A7).

The aqueous solutions were combined and extracted with ethyl acetate. The ethyl acetate extracts were combined, dried over  $\text{MgSO}_4$ , filtered and evaporated under reduced pressure. Further, the dried products have been purified by subjecting it with ethanol: petroleum ether (1:3) mixture to give white to off white crystals [11, 12].



**Scheme 3:** Synthetic scheme of substituted 4-aminobenzenesulfonamide from substituted chlorobenzene (A8, A9).



**Scheme 4:** Synthetic scheme of substituted N-acetyl-4-amino-benzenesulfonamide from substituted acids (A10, A11, A12).

## In-Silico Studies

### Docking Studies

#### Docking

Molecular docking techniques are used in modern drug design to help understand drug-receptor interaction. It has been shown in the literature that these computational procedures can strongly support and help the design of new, more potent drugs by revealing the mechanism of drug-receptor interaction. Rational drug design helps to facilitate and speedup the drug designing process, which involves variety of methods to identify novel compound, out of them one method is the docking of the drug molecule with the receptor. The therapeutic action of the clinical drug will be effective when the biochemical pathway of the enzyme can be exploited [13-18].

Docking procedures allows virtually screening a data-base of compounds and predict the strongest binder based on various scoring functions [13-18].

#### Receptor

*Pseudomonas aeruginosa* exotoxin A, *Bacillus subtilis* lipase A, *E-coli* primosomal protein, heterodimeric hexaprenyl diphosphate synthase, *Staphylococcus aureus* metalloproteinase and *Bacillus subtilis* Lon protease.

## Docking Tool

Here docking has been performed with AutoDock docking software. It is virtual screening software for computational drug discovery that can be used to screen libraries of compounds against potential drug targets. It enables medicinal chemists to run virtual screening from any platform and helps users in every steps of this process from data preparation to job submission and analysis of the results [13-18].

For performing docking, all receptors have been downloaded from NCBI website with PDB ID 1IKQ (*Pseudomonas aeruginosa* exotoxin A), 1R4Z (*Bacillus subtilis* lipase A), 2CCZ (*E-coli* primosomal protein), 3AQB (heterodimeric hexaprenyl diphosphate synthase), 3KHX (*Staphylococcus aureus* metallopeptidase) and 3M65 (*Bacillus subtilis* Lon protease), all the designed ligands have been docked with protein (receptor) with AutoDock software having its default settings.

## QSAR Studies

### QSAR

QSAR (quantitative structure-activity relationship) includes all statistical methods, by which biological activities (most often expressed by logarithms of equipotent molar activities) are related with structural elements, physicochemical properties or fields [19-26].

Classical QSAR analyses consider only 2D structures. Their main field of application is in substituent variation of a common scaffold.

3D-QSAR analysis has a much broader scope. It starts from 3D structures and correlates biological activities with 3D property fields.

Basic requirements in QSAR studies are-

- all analogs belong to a congeneric series
- all analogs exert the same mechanism of action
- all analogs bind in a comparable manner
- the effects of isosteric replacement can be predicted
- binding affinity is correlated to interaction energies
- biological activities are correlated to binding affinity

## Molecular Descriptors

Molecular descriptors can be defined as a numerical representation of chemical information encoded within a molecular structure *via* mathematical procedure. Type of QSAR is based on the dimensionality of molecular descriptors used:

- 0D- These are descriptors derived from molecular formula e.g. molecular weight, number and type of atoms etc.
- 1D- A substructure list representation of a molecule can be considered as a one-dimensional (1D) molecular representation and consists of a list of molecular fragments (e.g. functional groups, rings, bonds, substituents etc.).
- 2D- A molecular graph contains topological or two dimensional (2D) information. It describes how the atoms are bonded in a molecule, both the type of bonding and the interaction of particular atoms (e.g. total path count, molecular connectivity indices etc.).
- 3D- These are calculated starting from a geometrical or 3D representation of a molecule. These descriptors include molecular surface, molecular volume and other geometrical properties. There are different types of 3D descriptors e.g. electronic, steric, shape etc.

## Model Preparation

All the bioactivity values and information about 2D structure of sulfonamide analogues were taken from literature.  $\log 1/C$  is a variable that comprises the bioactivity parameter for the QSAR model. In order to calculate the molecular descriptors, PaDEL descriptor software, which incorporate CDK library for descriptor calculation has been used after optimizing the sulfonamide analogues. For the development of QSAR model, multiple linear regressions have been employed and all were validated through statistics [19-26].

## Modeling Parameters and Structure Optimization

The 2D structure construction, energy minimization and geometry optimization of the designed sulfonamide derivatives were carried out by using ChemDraw Ultra 7.0 and Chem3D Pro 7.0 (CambridgeSoft Corporation, 100 CambridgePark Drive, Cambridge MA, 02140 USA) on an Intel(R) Core(TM)2 Duo Central Processing Unit T6670 @ 2.20 GHz and 4.00 GB of RAM, running the Windows 7 Home Basic, 64-bit

Table 1: Physical and Elemental Data of all the Synthesized Compounds

Comp.	Molecular formula (MW)	Yield (%) [ $R_f$ ]	MP ( $^{\circ}\text{C}$ )	Elemental analysis (%) : Found (Calculated)			
				C	H	N	S
A1	$\text{C}_{13}\text{H}_{12}\text{N}_2\text{O}_3\text{S}$ (276.306)	68.75 [0.4]	205-208	56.42 (56.50)	3.260 (4.377)	10.13 (10.14)	11.16 (11.60)
A2	$\text{C}_{15}\text{H}_{14}\text{N}_2\text{O}_3\text{S}$ (302.342)	71.92 [0.84]	220-222	59.44 (59.58)	1.922 (4.667)	9.224 (9.267)	10.63 (10.60)
A3	$\text{C}_{11}\text{H}_{10}\text{N}_2\text{O}_3\text{S}_2$ (282.41)	56.73 [0.63]	210-212	39.89 (46.77)	0.969 (3.569)	8.540 (9.921)	19.16 (22.73)
A4	$\text{C}_{12}\text{H}_{11}\text{N}_3\text{O}_3\text{S}$ (277.052)	10.47 [0.34]	211-212	45.42 (51.98)	2.08 (4.00)	14.18 (15.16)	10.80 (11.54)
A5	$\text{C}_{12}\text{H}_{10}\text{ClN}_3\text{O}_3\text{S}$ (311.013)	86.18 [0.8]	230-231	46.15 (46.23)	2.90 (3.24)	13.25 (13.49)	10.03 (10.26)
A6	$\text{C}_{13}\text{H}_{14}\text{N}_2\text{O}_2\text{S}$ (262.077)	23.04 [0.24]	170-172	59.48 (59.52)	4.80 (5.38)	10.10 (10.69)	11.92 (12.20)
A7	$\text{C}_{14}\text{H}_{16}\text{N}_2\text{O}_3\text{S}$ (292.088)	41.98 [0.16]	180-182	57.21 (57.52)	3.90 (5.52)	9.20 (9.59)	10.02 (10.95)
A8	$\text{C}_{12}\text{H}_{13}\text{N}_3\text{O}_2\text{S}$ (263.072)	93.42 [0.46]	208-210	54.34 (54.74)	3.52 (4.98)	15.41 (15.97)	11.92 (12.15)
A9	$\text{C}_{14}\text{H}_{14}\text{N}_2\text{O}_3\text{S}$ (290.072)	84.88 [0.25]	216-218	57.36 (57.92)	2.98 (4.86)	9.42 (9.65)	10.87 (11.02)
A10	$\text{C}_{15}\text{H}_{14}\text{N}_2\text{O}_4\text{S}$ (318.067)	29.21 [0.18]	250-252	55.99 (56.59)	3.98 (4.44)	8.72 (8.81)	9.92 (10.05)
A11	$\text{C}_{17}\text{H}_{16}\text{N}_2\text{O}_4\text{S}$ (344.083)	62.18 [0.66]	265-266	59.12 (59.29)	3.23 (4.69)	8.11 (8.14)	9.10 (9.29)
A12	$\text{C}_{13}\text{H}_{12}\text{N}_2\text{O}_4\text{S}_2$ (324.023)	33.95 [0.2]	225-227	47.92 (48.14)	2.65 (3.73)	8.21 (8.64)	19.04 2(19.73)

compatible operating system. The energy minimization was carried out to minimum RMS Gradient of 0.100, with step interval of 2.0 Fs and frame interval of 10 Fs.

### QSAR Screening

All the designed 4-amino-benzenesulfonamides / N-acetyl-4-amino-benzenesulfonamide derivatives have been passed through the model (given below) after determining the descriptors used in model. Descriptors used are AlogP, Eccentric connectivity index (ECI) and Lipo-affinity index (LAI) for calculating  $\log 1/C$ .

### Descriptor Selection

The selection of descriptors among the calculated descriptors for the multiple linear regression analysis is based on the correlation matrix. This matrix is prepared and analyzed for the least correlated descriptors [19-26].

### Statistical Parameters

In the QSAR model, number of data points is denoted as  $n$ , squared correlation coefficient as  $r^2$  (fraction of variance), cross-validated  $r^2$  is denoted as  $q^2$ ,  $s$  is standard deviation.  $Q$  is quality factor, where  $Q = r/s$  (here  $r$  is correlation coefficient and  $s$  is standard deviation). Fischer statistics is denoted by  $F$  [19-26].

### Model Validation

The QSAR model validation was carried with statistical analysis and with internal validation [19-26].

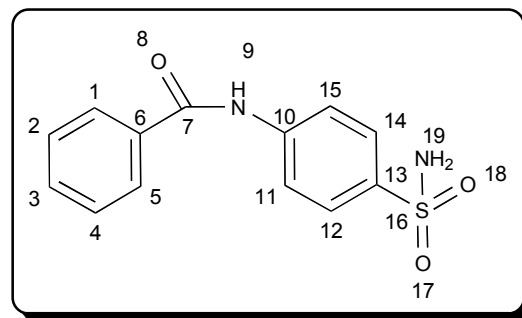
## RESULTS AND DISCUSSION

After synthesizing the designed compounds, they were treated for physical data like percentage yield, retention factor ( $R_f$ ), melting point and elemental data (CHNS analysis).

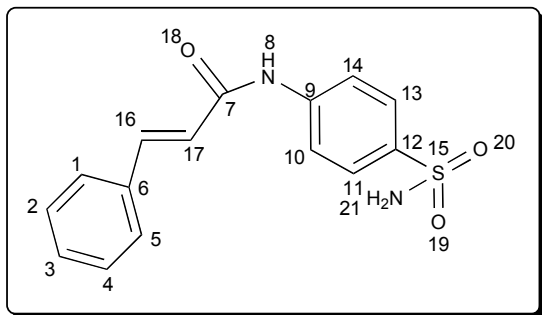
The physical and elemental data of the synthesized compounds are reported in Table 1.

### Spectral Characterization of Synthesized Substituted 4-amino-benzenesulfonamides/N-acetyl-4-amino-benzenesulfonamide Derivatives

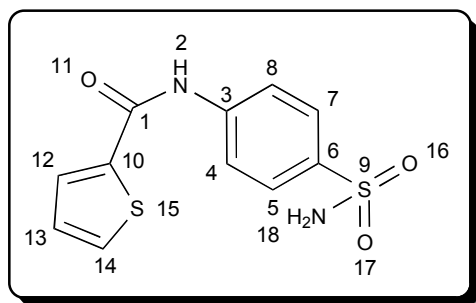
#### N-(4-Sulfamoyl-phenyl)-benzamide (A1)



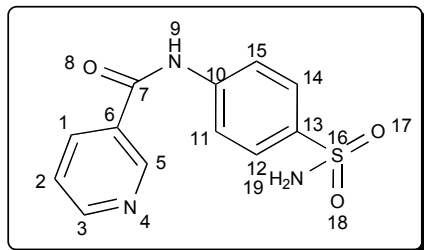
IR (KBr,  $\text{cm}^{-1}$ ,  $\nu$ ): 3340.53 ( $-\text{NH}_2$ ); 3250.94 ( $-\text{NH}-$ ); 1659.10 ( $>\text{C}=\text{O}$ ); 1397.80 ( $-\text{SO}_2-$ ).  $^1\text{H}$  NMR (DMSO, 400 MHz,  $\delta$  in ppm): 7.286(s, 2H,  $-\text{NH}_2$ ); 7.548-7.580(d, 2H, Ar-H of C12 and C14); 7.616-7.648(t, 1H, Ar-H of C3); 7.809-7.826(t, 2H, Ar-H of C2 and C4); 7.958-7.994 (d, 4H, Ar-H of C1, C5, C11 and C15); 10.564(s, 1H,  $>\text{NH}$ ). MS ( $m/z$ , %): 277.30 ( $\text{M}^+ + 1$ , 95).

**3-Phenyl-N-(4-sulfamoyl-phenyl)-acrylamide (A2)**

IR (KBr,  $\text{cm}^{-1}$ ,  $\nu$ ): 3359.69 ( $-\text{NH}_2$ -); 3182.61 ( $-\text{NH}-$ ); 1674.59 ( $>\text{C}=\text{O}$ ); 1400.63 ( $-\text{SO}_2$ -).  $^1\text{H}$  NMR (DMSO, 400 MHz,  $\delta$  in ppm): 7.271(s, 2H,  $-\text{NH}_2$ ); 2.5-3.3(d, 2H, C16 and C17); 7.489-7.492(d, 2H, Ar-H of C11 and C13); 7.631-7.663(t, 1H, Ar-H of C3); 7.791-7.809(t, 2H, Ar-H of C2 and C4); 7.857-7.875(d, 4H, Ar-H of C1, C5, C10 and C14); 10.551(s, 1H,  $>\text{NH}$ ). MS (m/z, %): 303.70 ( $\text{M}^+ + 1$ , 100).

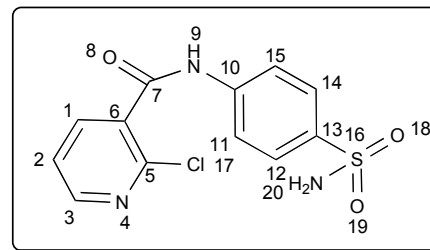
**Thiophene-2-carboxylic acid (4-sulfamoyl-phenyl)-amide (A3)**

IR (KBr,  $\text{cm}^{-1}$ ,  $\nu$ ): 3375.09 ( $-\text{NH}_2$ -); 3275.82 ( $-\text{NH}-$ ); 1650.90 ( $>\text{C}=\text{O}$ ); 1408.75 ( $-\text{SO}_2$ -).  $^1\text{H}$  NMR (DMSO, 400 MHz,  $\delta$  in ppm): 7.289(s, 2H,  $-\text{NH}_2$ ); 7.248-7.265(d, 1H, Thiophene-H of C14); 7.804-7.822(t, 1H, Ar-H of C13); 7.907-7.947(d, 4H, Ar-H of C4, C5, C7 and C8); 8.132-8.141(d, 2H, Thiophene-H of C12); 10.603(s, 1H,  $>\text{NH}$ ). MS (m/z, %): 283.71 ( $\text{M}^+ + 1$ , 100).

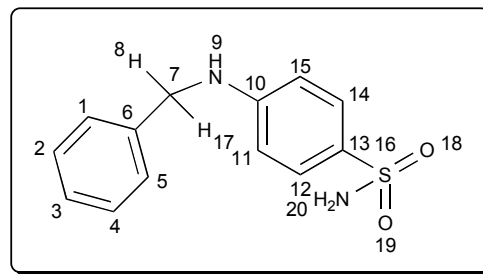
**N-(4-Sulfamoyl-phenyl)-nicotinamide (A4)**

IR (KBr,  $\text{cm}^{-1}$ ,  $\nu$ ): 3345.93 ( $-\text{NH}_2$ -); 3230.94 ( $-\text{NH}-$ ); 1659.19 ( $>\text{C}=\text{O}$ ); 1390.70 ( $-\text{SO}_2$ -).  $^1\text{H}$  NMR (DMSO, 400 MHz,  $\delta$  in ppm): 7.279(s, 2H,  $-\text{NH}_2$ ); 7.568-7.590(d, 2H,

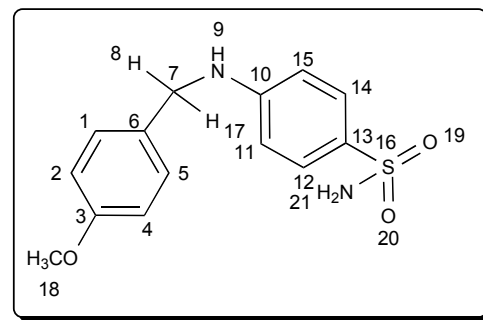
Ar-H of C12 and C14); 7.656-7.688(d, 1H, Ar-H of C3); 7.819-7.836(t, 2H, Ar-H of C2); 7.918-7.924 (d, 3H, Ar-H of C1, C11 and C15); 7.89(s, 1H, Ar-H of C5); 10.584(s, 1H,  $>\text{NH}$ ). MS (m/z, %): 278.10 ( $\text{M}^+ + 1$ , 88).

**2-Chloro-N-(4-sulfamoyl-phenyl)-nicotinamide (A5)**

IR (KBr,  $\text{cm}^{-1}$ ,  $\nu$ ): 700.10 ( $>\text{C}-\text{Cl}$ ), 3348.83 ( $-\text{NH}_2$ -); 3238.94 ( $-\text{NH}-$ ); 1657.19 ( $>\text{C}=\text{O}$ ); 1390.70 ( $-\text{SO}_2$ -).  $^1\text{H}$  NMR (DMSO, 400 MHz,  $\delta$  in ppm): 7.299(s, 2H,  $-\text{NH}_2$ ); 7.588-7.593(d, 2H, Ar-H of C12 and C14); 7.686-7.689(d, 1H, Ar-H of C3); 7.820-7.836(t, 2H, Ar-H of C2); 7.928-7.929(d, 3H, Ar-H of C1, C11 and C15); 10.594(s, 1H,  $>\text{NH}$ ). MS (m/z, %): 311.80 ( $\text{M}^+ + 1$ , 90).

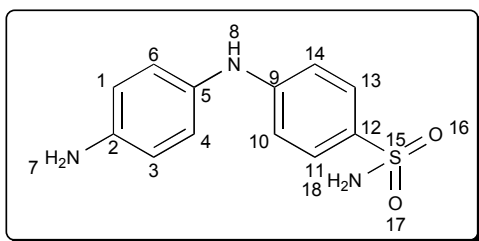
**4-Benzylamino-benzenesulfonamide (A6)**

IR (KBr,  $\text{cm}^{-1}$ ,  $\nu$ ): 3340.53 ( $-\text{NH}_2$ -); 3250.94 ( $-\text{NH}-$ ); 1397.80 ( $-\text{SO}_2$ -); 1465.76 ( $>\text{C}-\text{H}$ ).  $^1\text{H}$  NMR (DMSO, 400 MHz,  $\delta$  in ppm): 1.343(d, 2H of C7); 7.306(s, 2H,  $-\text{NH}_2$ ); 7.558-7.590(d, 2H, Ar-H of C12 and C14); 7.626-7.638(t, 1H, Ar-H of C3); 7.819-7.827(t, 2H, Ar-H of C2 and C4); 7.918-7.904(d, 4H, Ar-H of C1, C5, C11 and C15); 10.563(s, 1H,  $>\text{NH}$ ). MS (m/z, %): 263.30 ( $\text{M}^+ + 1$ , 95).

**4-(4-Methoxy-benzylamino)-benzenesulfonamide (A7)**

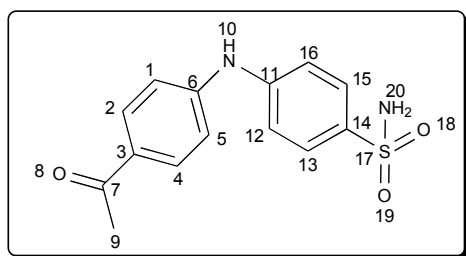
IR (KBr,  $\text{cm}^{-1}$ ,  $\nu$ ): 3340.53 ( $-\text{NH}_2-$ ); 3250.94 ( $-\text{NH}-$ ); 1397.80 ( $-\text{SO}_2-$ ); 1315.87 ( $>\text{C}=\text{O}-$ ); 1465.76 ( $>\text{C}-\text{H}$ ).  $^1\text{H}$  NMR (DMSO, 400 MHz,  $\delta$  in ppm): 0.899(s, 3H,  $-\text{CH}_3$ ); 1.383(d, 2H of C7); 7.316(s, 2H,  $-\text{NH}_2$ ); 7.508-7.510(d, 2H, Ar-H of C12 and C14); 7.828-7.829(d, 2H, Ar-H of C2 and C4); 7.910-7.914(d, 4H, Ar-H of C1, C5, C11 and C15); 10.523(s, 1H,  $>\text{NH}$ ). MS (m/z, %): 293.10 ( $\text{M}^+ + 1$ , 90).

#### 4-(4-Amino-phenylamino)-benzenesulfonamide (A8)



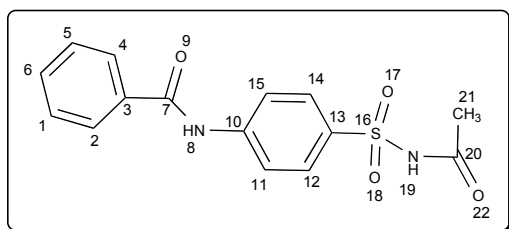
IR (KBr,  $\text{cm}^{-1}$ ,  $\nu$ ): 3337.23 ( $-\text{NH}_2-$  of C2); 3342.83 ( $-\text{NH}_2-$ ); 3190.84 ( $-\text{NH}-$ ); 1679.10 ( $>\text{C}=\text{O}$ ); 1387.90 ( $-\text{SO}_2-$ ).  $^1\text{H}$  NMR (DMSO, 400 MHz,  $\delta$  in ppm): 6.986(s, 2H,  $-\text{NH}_2$ ); 7.236(s, 2H,  $-\text{NH}_2$ ); 7.498-7.550(d, 2H, Ar-H of C11 and C13); 7.819-7.829(d, 2H, Ar-H of C1 and C3); 7.928-7.964 (d, 4H, Ar-H of C4, C6, C10 and C14); 10.524(s, 1H,  $>\text{NH}$ ). MS (m/z, %): 264.80 ( $\text{M}^+ + 1$ , 89).

#### 4-(4-Acetyl-phenylamino)-benzenesulfonamide (A9)



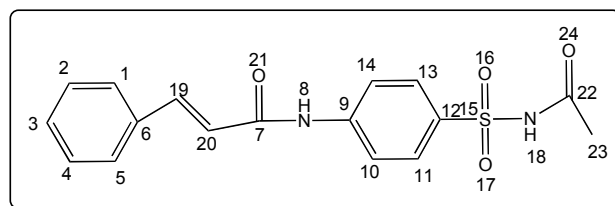
IR (KBr,  $\text{cm}^{-1}$ ,  $\nu$ ): 3348.83 ( $-\text{NH}_2-$ ); 3193.84 ( $-\text{NH}-$ ); 1639.70 ( $>\text{C}=\text{O}$ ); 1392.90 ( $-\text{SO}_2-$ ).  $^1\text{H}$  NMR (DMSO, 400 MHz,  $\delta$  in ppm): 0.909(s, 3H of C9); 7.256(s, 2H,  $-\text{NH}_2$ ); 7.428-7.510(d, 2H, Ar-H of C13 and C15); 7.839-7.859(d, 2H, Ar-H of C2 and C4); 7.978-7.994 (d, 4H, Ar-H of C1, C5, C12 and C16); 10.584(s, 1H,  $>\text{NH}$ ). MS (m/z, %): 291.30 ( $\text{M}^+ + 1$ , 98).

#### N-(4-Acetylsulfamoyl-phenyl)-benzamide (A10)



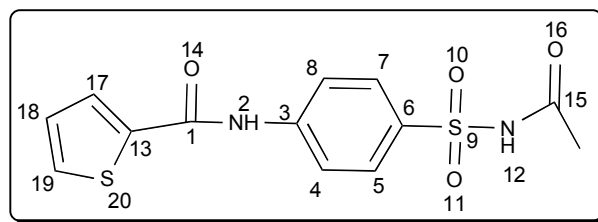
IR (KBr,  $\text{cm}^{-1}$ ,  $\nu$ ): 3245.53, 3250.94 ( $-\text{NH}-$ ); 1678.30, 1659.10 ( $>\text{C}=\text{O}$ ); 1396.50 ( $-\text{SO}_2-$ ).  $^1\text{H}$  NMR (DMSO, 400 MHz,  $\delta$  in ppm): 0.919(s, 3H,  $-\text{CH}_3$ ); 7.512-7.530(d, 2H, Ar-H of C12 and C14); 7.596-7.628(t, 1H, Ar-H of C6); 7.839-7.846(t, 2H, Ar-H of C1 and C5); 7.908-7.954 (d, 4H, Ar-H of C2, C4, C11 and C15); 10.497, 10.527(s, 1H,  $>\text{NH}$ ). MS (m/z, %): 319.90 ( $\text{M}^+ + 1$ , 70).

#### N-(4-Acetylsulfamoyl-phenyl)-3-phenyl-acrylamide (A11)



IR (KBr,  $\text{cm}^{-1}$ ,  $\nu$ ): 3188.98, 3181.61 ( $-\text{NH}-$ ); 1681.09, 1678.59 ( $>\text{C}=\text{O}$ ); 1420.63 ( $-\text{SO}_2-$ ).  $^1\text{H}$  NMR (DMSO, 400 MHz,  $\delta$  in ppm): 0.917(s, 3H,  $-\text{CH}_3$ ); 2.518-3.398(d, 2H, C19 and C20); 7.479-7.482(d, 2H, Ar-H of C11 and C13); 7.611-7.623(t, 1H, Ar-H of C3); 7.781-7.701(t, 2H, Ar-H of C2 and C4); 7.859-7.870(d, 4H, Ar-H of C1, C5, C10 and C14); 10.534, 10.550(s, 1H,  $>\text{NH}$ ). MS (m/z, %): 345.70 ( $\text{M}^+ + 1$ , 94).

#### Thiophene-2-carboxylic acid (4-acetylsulfamoyl-phenyl)-amide (A12)



IR (KBr,  $\text{cm}^{-1}$ ,  $\nu$ ): 3252.83, 3277.80 ( $-\text{NH}-$ ); 1659.07, 1654.90 ( $>\text{C}=\text{O}$ ); 1408.75 ( $-\text{SO}_2-$ ).  $^1\text{H}$  NMR (DMSO, 400 MHz,  $\delta$  in ppm): 0.927(s, 3H,  $-\text{CH}_3$ ); 7.247-7.255(d, 1H, Thiophene-H of C19); 7.807-7.820(t, 1H, Ar-H of C18); 7.937-7.949(d, 4H, Ar-H of C4, C5, C7 and C8); 8.134-8.142(d, 2H, Thiophene-H of C17); 10.567, 10.603(s, 1H,  $>\text{NH}$ ). MS (m/z, %): 325.81 ( $\text{M}^+ + 1$ , 100).

#### Docking study of 4-amino-benzenesulfonamides/N-acetyl-4-amino-benzenesulfonamide derivatives and standard drug taken Norfloxacin (CID\_4539)

Docking study of different proteins were performed with the designed inhibitors and standard drug taken is given in Tables 2 to 14 and number of hydrogen bonds & binding pattern such as element, type of bond, atom number and residue at binding site were evaluated.

**Table 2: Docking Analysis of Synthesized Compound A1**

Ligand	Receptor	Affinity Kcal/mol	H-bonds	H- Binding Ligand			H- Binding Receptor			
				Elem.	At. ID.	Type	Res.	Elem.	At.ID.	Type
A1	1IKQ	-7.6	4	O	18	Acceptor	THR 271	O	2083	Both
				O	17	Acceptor	THR 271	O	2083	Both
				O	08	Acceptor	TRP 187	N	1397	Donor
				O	08	Acceptor	SER 188	N	1411	Donor
	1R4Z	-6	5	N	19	Donor	LEU 173	O	1293	Acceptor
				N	19	Donor	ASP 72	O	546	Acceptor
				O	18	Acceptor	ASN 4	N	17	Donor
				O	17	Acceptor	ASN 98	N	736	Donor
				O	08	Acceptor	HIS 3	N	0	Donor
	2CCZ	-6.1	3	O	17	Acceptor	SER 88	O	683	Both
				N	19	Donor	SER 88	O	683	Both
				N	19	Donor	SER 88	O	681	Acceptor
	3AQB	-7.5	0	-	-	-	-	-	-	-
	3KHX	-6.4	5	N	19	Donor	ASP 311	O	2350	Acceptor
				N	19	Donor	ASP 311	O	2351	Acceptor
				N	19	Donor	PRO 273	O	2052	Acceptor
				N	19	Donor	GLY 276	O	2073	Acceptor
				O	17	Acceptor	ASN 278	N	2081	Donor
	3M65	-6.8	3	N	19	Donor	LEU 171	O	1325	Acceptor
				N	19	Donor	SER 169	O	1309	Acceptor
				O	08	Acceptor	SER 147	N	1146	Donor

On docking analysis, designed compound A1 has been found to be strongly docked with the protein 1IKQ when binding with 6 different proteins have been observed in order to study its inhibition activity. When it is docked with the protein PDB ID- 1IKQ, it forms 4 hydrogen bonds with binding affinity of -7.6 Kcal/mol. On residue study, the amino acids THR 271, TRP 187 and SER 188 were found to be significant. On the account of ligand here oxygen atom is significant in the binding with the type of binding found with acceptor bonds, whereas significant elements in receptor binding are oxygen and nitrogen both.

On docking analysis, designed compound A2 has been also found to be strongly docked with the protein 1IKQ when binding with 6 different proteins have been observed in order to study its inhibition activity. When it is docked with the protein PDB ID- 1IKQ, it forms 4 hydrogen bonds with binding affinity of -8.4 Kcal/mol. On residue study, the amino acids THR 271, TRP 187, GLU 270 and ARG 274 were found to be significant. On the account of ligand here oxygen and nitrogen both atoms are significant in the binding, the type of

binding found with acceptor bonds, whereas significant elements in receptor binding are oxygen and nitrogen both.

On docking analysis, designed compound A3 has been also found to be strongly docked with the protein 1IKQ when binding with 6 different proteins have been observed in order to study its inhibition activity. When it is docked with the protein PDB ID- 1IKQ, it forms 4 hydrogen bonds with binding affinity of -7.2 Kcal/mol. On residue study, the amino acids THR 271, TRP 187 and SER 188 were found to be significant. On the account of ligand here oxygen atom is significant in the binding with the type of binding found with acceptor bonds, whereas significant elements in receptor binding are oxygen and nitrogen both.

On docking analysis, designed compound A4 has been also found to be strongly docked with the protein 1IKQ when binding with 6 different proteins have been observed in order to study its inhibition activity. When it is docked with the protein PDB ID- 1IKQ, it forms 5 hydrogen bonds with binding affinity of -7.8 Kcal/mol.



Table 3: Docking Analysis of Synthesized Compound A2

Ligand	Receptor	Affinity Kcal/mol	H- bonds	H- Binding Ligand			H- Binding Receptor			
				Elem.	At. ID.	Type	Res.	Elem.	At.ID.	Type
A2	1IKQ	-8.4	4	O	19	Acceptor	THR 271	O	2083	Both
				O	20	Acceptor	ARG 274	N	2112	Donor
				N	21	Donor	GLU 270	O	2076	Acceptor
				O	08	Acceptor	TRP 187	N	1397	Donor
	1R4Z	-6.3	4	N	21	Donor	LEU 173	O	1293	Acceptor
				N	21	Donor	ASP 72	O	546	Acceptor
				O	19	Acceptor	ASN 98	N	736	Donor
				O	20	Acceptor	ASN 4	N	17	Donor
	2CCZ	-6.5	3	O	08	Acceptor	GLN 49	N	394	Donor
				O	19	Acceptor	LYS 89	N	692	Donor
				O	20	Acceptor	ARG 13	N	119	Donor
	3AQB	-7.5	0	-	-	-	-	-	-	-
	3KHX	-6.8	5	N	21	Donor	ASP 311	O	2350	Acceptor
				N	21	Donor	ASP 311	O	2351	Acceptor
				N	21	Donor	PRO 273	O	2052	Acceptor
				N	21	Donor	GLY 276	O	2073	Acceptor
				O	19	Acceptor	ASN 278	N	2081	Donor
	3M65	-7.2	2	N	06	Donor	THR 150	O	1171	Both
				O	08	Acceptor	THR 150	O	1171	Both

Table 4: Docking Analysis of Synthesized Compound A3

Ligand	Receptor	Affinity Kcal/mol	H- bonds	H- Binding Ligand			H- Binding Receptor			
				Elem.	At. ID.	Type	Res.	Elem.	At.ID.	Type
A3	1IKQ	-7.2	4	O	08	Acceptor	TRP 187	N	1397	Donor
				O	08	Acceptor	SER 188	N	1411	Donor
				O	17	Acceptor	THR 271	O	2083	Both
				O	16	Acceptor	THR 271	O	2083	Both
	1R4Z	-6	5	N	18	Donor	LEU 173	O	1293	Acceptor
				N	18	Donor	ASP 72	O	546	Acceptor
				O	08	Acceptor	HIS 3	N	0	Donor
				O	17	Acceptor	ASN 4	N	17	Donor
				O	16	Acceptor	ASN 98	N	736	Donor
	2CCZ	-5.8	3	N	18	Donor	SER 88	O	681	Acceptor
				N	18	Donor	SER 88	O	683	Both
				O	16	Acceptor	SER 88	O	683	Both
	3AQB	-6.9	2	N	06	Donor	ASP 152	O	1180	Acceptor
				O	17	Acceptor	SER 159	O	1243	Both
	3KHX	-5.8	5	N	18	Donor	ASP 311	O	2350	Acceptor
				N	18	Donor	ASP 311	O	2351	Acceptor
				N	18	Donor	PRO 273	O	2052	Acceptor
				N	18	Donor	GLY 276	O	2073	Acceptor
				O	16	Acceptor	ASN 278	N	2081	Donor
	3M65	-6.5	3	O	17	Acceptor	THR 150	O	1171	Both
				O	17	Acceptor	HIS 170	N	1318	Donor
				N	18	Donor	HIS 170	O	1315	Acceptor

**Table 5: Docking Analysis of Synthesized Compound A4**

Ligand	Receptor	Affinity Kcal/mol	H-bonds	H- Binding Ligand			H- Binding Receptor			
				Elem.	At. ID.	Type	Res.	Elem.	At.ID.	Type
A4	1IKQ	-7.8	5	N	20	Donor	THR 134	O	1035	Both
				N	20	Donor	GLU 270	O	2077	Acceptor
				N	14	Donor	SER 188	O	1414	Acceptor
				O	19	Acceptor	GLU 270	N	2069	Donor
				N	06	Donor	GLU 391	O	2984	Acceptor
	1R4Z	-6.2	2	O	08	Acceptor	HIS 3	N	0	Donor
				N	14	Donor	LEU 173	O	1293	Acceptor
	2CCZ	-5.9	0	-	-	-	-	-	-	-
	3AQB	-7.3	0	-	-	-	-	-	-	-
	3KHX	-6.4	2	N	14	Donor	THR 398	O	2966	Acceptor
				O	08	Acceptor	THR 398	O	2968	Both
	3M65	-6.8	3	O	19	Acceptor	THR 150	O	1171	Both
				O	19	Acceptor	HIS 170	N	1318	Donor
				N	20	Donor	HIS 170	O	1315	Acceptor

**Table 6: Docking Analysis of Synthesized Compound A5**

Ligand	Receptor	Affinity Kcal/mol	H-bonds	H- Binding Ligand			H- Binding Receptor			
				Elem.	At. ID.	Type	Res.	Elem.	At.ID.	Type
A5	1IKQ	-7.4	6	O	19	Acceptor	SER 192	O	1450	Both
				O	19	Acceptor	GLY 193	N	1451	Donor
				O	19	Acceptor	LYS 194	N	1455	Donor
				N	21	Donor	THR 371	O	2858	Both
				O	08	Acceptor	THR 396	O	3029	Both
				O	20	Acceptor	TYR 206	O	1554	Both
	1R4Z	-6.4	6	N	06	Donor	ASN 174	O	1301	Acceptor
				O	20	Acceptor	ASN 4	N	10	Donor
				O	19	Acceptor	ASN 98	N	736	Donor
				O	19	Acceptor	ASN 4	N	17	Donor
				N	21	Donor	ASP 72	O	546	Acceptor
				N	21	Donor	LEU 173	O	1293	Acceptor
	2CCZ	-6	1	N	21	Donor	GLN 49	O	410	Acceptor
	3AQB	-7.6	0	-	-	-	-	-	-	-
	3KHX	-6.9	0	-	-	-	-	-	-	-
	3M65	-6.7	3	N	21	Donor	LEU 171	O	1325	Acceptor
				N	21	Donor	SER 169	O	1309	Acceptor
				O	08	Acceptor	SER 147	N	1146	Donor

On residue study, the amino acids THR 234, GLU 270, GLU 391 and SER 188 were found to be significant. On the account of ligand here nitrogen atom is significant in

the binding, the type of binding found with donor bonds, whereas significant elements in receptor binding are oxygen.

Table 7: Docking Analysis of Synthesized Compound A6

Ligand	Receptor	Affinity Kcal/mol	H-bonds	H- Binding Ligand			H- Binding Receptor			
				Elem.	At. ID.	Type	Res.	Elem.	At.ID.	Type
A6	1IKQ	-7.6	3	N	18	Donor	SER 188	O	1416	Both
				N	18	Donor	SER 188	O	1414	Acceptor
				O	16	Acceptor	SER 188	O	1416	Both
	1R4Z	-5.8	5	O	16	Acceptor	ASN 98	N	736	Donor
				O	16	Acceptor	ASN 4	N	17	Donor
				N	18	Donor	ASP 72	O	546	Acceptor
				N	18	Donor	LEU 173	O	1293	Acceptor
				O	17	Acceptor	ASN 4	N	10	Donor
	2CCZ	-6	1	O	17	Acceptor	CYS 80	S	640	Donor
	3AQB	-7.6	0	-	-	-	-	-	-	-
	3KHX	-6.4	0	-	-	-	-	-	-	-
	3M65	-6.2	2	N	18	Donor	GLU 208	O	1630	Acceptor
				N	18	Donor	LEU 205	O	1605	Acceptor

Table 8: Docking Analysis of Synthesized Compound A7

Ligand	Receptor	Affinity Kcal/mol	H-bonds	H- Binding Ligand			H- Binding Receptor			
				Elem.	At. ID.	Type	Res.	Elem.	At.ID.	Type
A7	1IKQ	-7.5	8	O	18	Acceptor	LYS 114	N	869	Donor
				O	18	Acceptor	HIS 246	N	1894	Donor
				O	18	Acceptor	THR 134	O	1035	Both
				N	20	Donor	GLU 270	O	2077	Acceptor
				N	20	Donor	THR 134	O	1035	Both
				O	15	Acceptor	SER 188	O	1416	Both
				N	06	Donor	HIS 262	N	2015	Acceptor
				N	06	Donor	GLU 391	O	2984	Acceptor
	1R4Z	-6.2	0	-	-	-	-	-	-	-
	2CCZ	-6.1	2	O	19	Acceptor	LYS 82	N	654	Donor
				O	18	Acceptor	MET 90	N	704	Donor
	3AQB	-7.3	3	N	20	Donor	VAL 158	O	1234	Acceptor
				O	19	Acceptor	TYR 156	O	1223	Both
				O	18	Acceptor	TYR 156	O	1223	Both
	3KHX	-6.4	2	O	19	Acceptor	ASN 309	N	2330	Donor
				N	20	Donor	ASP 311	O	2350	Acceptor
	3M65	-6.6	1	O	15	Acceptor	SER 147	O	1151	Both

On docking analysis, designed compound A5 has been found to be strongly docked with the protein 1IKQ when binding with 6 different proteins have been observed in order to study its inhibition activity. When it is docked with the protein PDB ID- 1IKQ, it forms 6

hydrogen bonds with binding affinity of -7.4 Kcal/mol. On residue study, the amino acids SER 192, GLY 193, LYS 194, THR 371, THR 396 and TYR 206 were found to be significant. On the account of ligand here oxygen atom is significant in the binding, the type of binding

**Table 9: Docking Analysis of Synthesized Compound A8**

Ligand	Receptor	Affinity Kcal/mol	H-bonds	H- Binding Ligand			H- Binding Receptor			
				Elem.	At. ID.	Type	Res.	Elem.	At.ID.	Type
A8	1IKQ	-7.3	4	N	14	Donor	THR 134	O	1035	Both
				N	20	Donor	SER 188	O	1414	Acceptor
				N	20	Donor	SER 188	O	1416	Both
				O	18	Acceptor	SER 188	O	1416	Both
	1R4Z	-6.3	5	O	18	Acceptor	ASN 4	N	17	Donor
				O	18	Acceptor	ASN 98	N	736	Donor
				N	20	Donor	LEU 173	O	1293	Acceptor
				N	20	Donor	ASP 72	O	546	Acceptor
				O	19	Acceptor	ASN 4	N	10	Donor
	2CCZ	-6	1	N	20	Donor	GLN 49	O	410	Acceptor
	3AQB	-7.6	3	N	20	Donor	VAL 158	O	1234	Acceptor
				O	19	Acceptor	TYR 156	O	1223	Both
				O	18	Acceptor	TYR 156	O	1223	Both
	3KHX	-6.8	0	-	-	-	-	-	-	-
	3M65	-6.1	3	N	14	Donor	HIS 170	O	1315	Acceptor
				N	20	Donor	SER 143	O	1119	Both
				O	18	Acceptor	TYR 139	O	1088	Both

found with acceptor bonds, whereas significant elements in receptor binding are oxygen and nitrogen both.

On docking analysis, designed compound A6 has been found to be strongly docked with the protein 1IKQ when binding with 6 different proteins have been observed in order to study its inhibition activity. When it is docked with the protein PDB ID- 1IKQ, it forms 3 hydrogen bonds with binding affinity of -7.6 Kcal/mol. On residue study, the amino acid SER 188 was found to be significant. On the account of ligand here nitrogen atom is significant in the binding, the type of binding found with donor bonds, whereas significant elements in receptor binding are oxygen.

On docking analysis, designed compound A7 has been found to be strongly docked with the protein 1IKQ when binding with 6 different proteins have been observed in order to study its inhibition activity. When it is docked with the protein PDB ID- 1IKQ, it forms 8 hydrogen bonds with binding affinity of -7.5 Kcal/mol. On residue study, the amino acids LYS 114, HIS 246, THR 134, GLU 270, HIS 262, GLU 391 and SER 188 were found to be significant. On the account of ligand here oxygen and nitrogen atoms are significant in the

binding, the type of binding found with acceptor and donor bonds, whereas significant elements in receptor binding are oxygen and nitrogen both.

On docking analysis, designed compound A8 has been found to be strongly docked with the protein 3AQB when binding with 6 different proteins have been observed in order to study its inhibition activity. When it is docked with the protein PDB ID- 3AQB, it forms 3 hydrogen bonds with binding affinity of -7.6 Kcal/mol. On residue study, the amino acids VAL 158 and TYR 156 were found to be significant. On the account of ligand here oxygen atom is significant in the binding, the type of binding found with acceptor bonds, whereas significant elements in receptor binding are oxygen.

On docking analysis, designed compound A9 has been found to be strongly docked with the protein 3AQB when binding with 6 different proteins have been observed in order to study its inhibition activity. When it is docked with the protein PDB ID- 3AQB, it forms 3 hydrogen bonds with binding affinity of -8.2 Kcal/mol. On residue study, the amino acids VAL 158 and TYR 156 were found to be significant. On the account of ligand here oxygen atom is significant in the binding, the type of binding found with acceptor bonds, whereas significant elements in receptor binding are oxygen.

Table 10: Docking Analysis of Synthesized Compound A9

Ligand	Receptor	Affinity Kcal/mol	H-bonds	H- Binding Ligand			H- Binding Receptor			
				Elem.	At. ID.	Type	Res.	Elem.	At.ID.	Type
A9	1IKQ	-8	4	O	18	Acceptor	SER 188	O	1416	Both
				N	20	Donor	SER 188	O	1416	Both
				N	20	Donor	SER 188	O	1414	Acceptor
				O	15	Acceptor	THR 134	O	1035	Both
	1R4Z	-6.2	1	N	20	Donor	GLY 30	O	202	Acceptor
	2CCZ	-6.5	2	O	19	Acceptor	CYS 80	S	640	Donor
				O	19	Acceptor	HIS 81	N	641	Donor
	3AQB	-8.2	3	O	19	Acceptor	TYR 156	O	1223	Both
				O	18	Acceptor	TYR 156	O	1223	Both
				N	20	Donor	VAL 158	O	1234	Acceptor
	3KHX	-6.6	1	O	15	Acceptor	THR 358	O	2968	Both
	3M65	-6.5	2	O	18	Acceptor	LYS 178	N	1387	Donor
				N	20	Donor	SER 169	O	1309	Acceptor

Table 11: Docking Analysis of Synthesized Compound A10

Ligand	Receptor	Affinity Kcal/mol	H-bonds	H- Binding Ligand			H- Binding Receptor			
				Elem.	At. ID.	Type	Res.	Elem.	At.ID.	Type
A-10	1IKQ	-9	5	N	19	Donor	SER 188	O	1416	Both
				O	22	Acceptor	SER 188	O	1416	Both
				O	22	Acceptor	SER 192	O	1450	Both
				N	06	Donor	ALA 191	O	1443	Acceptor
				O	08	Acceptor	LEU 269	N	2061	Donor
	1R4Z	-7.1	1	N	06	Donor	GLY 30	O	202	Acceptor
	2CCZ	-7	1	O	08	Acceptor	LYS 82	N	654	Donor
	3AQB	-8.7	0	-	-	-	-	-	-	-
	3KHX	-7.1	1	O	17	Acceptor	THR 398	O	2968	Both
	3M65	-7.2	1	N	19	Donor	HIS 170	O	1315	Acceptor

On docking analysis, designed compound A10 has been also found to be strongly docked with the protein 1IKQ when binding with 6 different proteins have been observed in order to study its inhibition activity. When it is docked with the protein PDB ID- 1IKQ, it forms 5 hydrogen bonds with binding affinity of -9 Kcal/mol. On residue study, the amino acids SER 188, SER 192, ALA 191 and LEU 269 were found to be significant. On the account of ligand here oxygen atom is significant in the binding, the type of binding found with acceptor bonds, whereas significant elements in receptor binding are oxygen.

On docking analysis, designed compound A11 has been found to be strongly docked with the protein 1IKQ

when binding with 6 different proteins have been observed in order to study its inhibition activity. When it is docked with the protein PDB ID- 1IKQ, it forms 5 hydrogen bonds with binding affinity of -9.4 Kcal/mol. On residue study, the amino acids SER 188, SER 369, ALA 191 and ASN 393 were found to be significant. On the account of ligand here oxygen atom is significant in the binding, the type of binding found with acceptor bonds, whereas significant elements in receptor binding are oxygen.

On docking analysis, designed compound A12 has been found to be strongly docked with the protein 3M65 when binding with 6 different proteins have been

**Table 12: Docking Analysis of Synthesized Compound A11**

Ligand	Receptor	Affinity Kcal/mol	H- bonds	H- Binding Ligand			H- Binding Receptor			
				Elem.	At. ID.	Type	Res.	Elem.	At.ID.	Type
A-11	1IKQ	-9.4	5	O	19	Acceptor	SER 369	O	2844	Both
				O	24	Acceptor	SER 369	O	2844	Both
				O	08	Acceptor	SER 188	O	1416	Both
				O	19	Acceptor	ASN 393	N	3004	Donor
				N	21	Donor	ALA 191	O	1443	Acceptor
	1R4Z	-7	0	-	-	-	-	-	-	-
	2CCZ	-6.9	2	O	19	Acceptor	LYS 82	N	654	Donor
				O	20	Acceptor	MET 90	N	704	Donor
	3AQB	-8.7	0	-	-	-	-	-	-	-
	3KHX	-6.9	3	O	08	Acceptor	ASN 278	N	2100	Donor
				N	06	Donor	ASN 331	O	2453	Acceptor
				O	19	Acceptor	THR 329	O	2438	Both
	3M65	-7.5	2	O	08	Acceptor	THR 150	O	1171	Both
				N	06	Donor	THR 150	O	1171	Both

**Table 13: Docking Analysis of Synthesized Compound A12**

Ligand	Receptor	Affinity Kcal/mol	H- bonds	H- Binding Ligand			H- Binding Receptor			
				Elem.	At. ID.	Type	Res.	Elem.	At.ID.	Type
A-12	1IKQ	-7.8	3	O	21	Acceptor	SER 369	O	2844	Both
				O	16	Acceptor	SER 369	O	2844	Both
				O	08	Acceptor	SER 188	O	1416	Both
	1R4Z	-6.2	2	O	21	Acceptor	ASN 4	N	17	Donor
				O	17	Acceptor	HIS 3	N	0	Donor
	2CCZ	-6.4	1	O	21	Acceptor	LYS 82	N	654	Donor
	3AQB	-7.6	0	-	-	-	-	-	-	-
	3KHX	-6.5	1	O	08	Acceptor	THR 398	O	2968	Both
	3M65	-7	5	O	21	Acceptor	TYR 139	O	1088	Both
				O	16	Acceptor	TYR 139	O	1088	Both
				O	16	Acceptor	SER 143	O	1119	Both
				O	08	Acceptor	THR 150	O	1171	Both
				O	08	Acceptor	HIS 170	N	1318	Donor

observed in order to study its inhibition activity. When it is docked with the protein PDB ID- 3M65, it forms 5 hydrogen bonds with binding affinity of -7 Kcal/mol. On residue study, the amino acids TYR 139, SER 143, THR 150 and HIS 170 were found to be significant. On the account of ligand here oxygen atom is significant in the binding, the type of binding found with acceptor bonds, whereas significant elements in receptor binding are oxygen.

On docking analysis, the standard drug CID\_4539 has been found to be strongly docked with the protein 1IKQ when binding with 6 different proteins have been observed in order to study its inhibition activity. When it is docked with the protein PDB ID- 1IKQ, it forms 4 hydrogen bonds with binding affinity of -8.1 Kcal/mol. On residue study, the amino acids TYR 206, GLN 212, GLU 399, TYR 206 and GLN 212 were found to be significant. On the account of ligand here oxygen atom

Table 14: Docking Analysis of Norfloxacin (CID\_4539)

Ligand	Receptor	Affinity Kcal/mol	H- bonds	H- Binding Ligand			H- Binding Receptor			
				Elem.	At. ID.	Type	Res.	Elem.	At.ID.	Type
CID_4539	1IKQ	-8.1	4	O	23	Both	GLN 212	O	1604	Acceptor
				O	23	Both	GLU 399	O	3048	Acceptor
				O	22	Acceptor	TYR 206	O	1554	Both
				O	22	Acceptor	GLN 212	N	1605	Donor
	1R4Z	-6.2	1	O	23	Both	GLN 29	O	193	Acceptor
	2CCZ	-6.1	0	-	-	-	-	-	-	-
	3AQB	-7.3	0	-	-	-	-	-	-	-
	3KHX	-6.3	0	-	-	-	-	-	-	-
	3M65	-7.4	2	O	22	Acceptor	THR 150	O	1171	Both
				O	23	Both	HIS 170	O	1315	Acceptor

is significant in the binding, the type of binding found with both acceptor and donor bonds, whereas significant elements in receptor binding are oxygen.

### Validation of QSAR Model

The 2D structure of sulphonamide derivatives from which the QSAR model has been developed is shown in Figure 1. From the data in Table 15, QSAR equation was developed where number of data point (n) is 25 and number of descriptors used are 3. The derived QSAR model is given below. Here 95% confidence intervals are given in parantheses.

$$\log 1/C = (4.192476)(0.8976384) + (0.3564435)(\text{AlogP}) \\ (0.220396) + (0.0073582)(\text{ECI})(0.0028634) + \\ (-0.4004489)(\text{LAI})(0.1360423)$$

A comparison (multiple linear regression curve) of observed values and predicted values of  $\log 1/C$  for sulphonamide derivatives used for development of QSAR equation is shown in Figure 2.

A quantitative assessment of model robustness has been performed through model validation. All the statistical results of model validation have been given in Table 16.

### Statistical Analysis

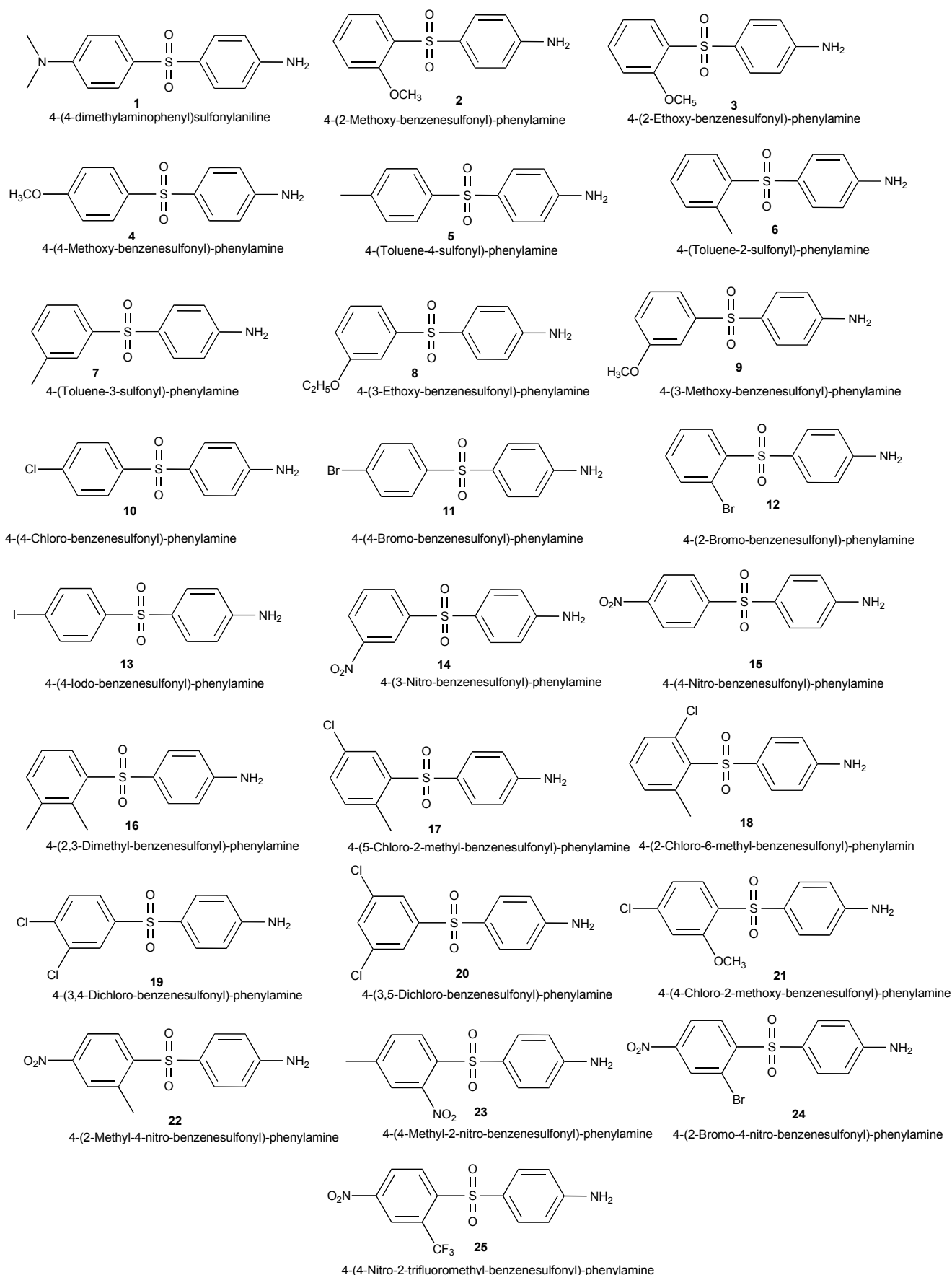
- (1) **n/p ratio:**  $n/p = \geq 4$ , where  $n$  is the number of data points and  $p$  is the number of descriptors used in the QSAR model. The model obeys the condition.
- (2) **Fraction of variance ( $r^2$ ):** The value of fraction of variance may vary between 0 (means model without explanatory power) and 1 (means perfect

model). QSAR model having  $r^2 > 0.6$  will only be considered for validation. The value for this QSAR model is 0.765.

- (3) **Cross-Validation Test ( $q^2$ ):** A QSAR model must have  $q^2 > 0.5$  for the predictive ability. The value of  $q^2$  for this QSAR model is 0.7784.
- (4) **Standard deviation (s):** The smaller s value is always required for the predictive QSAR model. The value of s for this QSAR model is 0.4.
- (6)  **$r^2 - q^2 < 0.3$ :** The difference between  $r^2$  and  $q^2$  should never be exceeding by 0.3. A large difference suggests the following: presence of outliers, over-fitted model, and presence of irrelevant variables in data. The value of  $r^2 - q^2$  for this QSAR model is -0.013.
- (7) **Quality Factor (Q):** Over fitting and chance correlation, due to excess number of descriptors, can be detected by Q value. Positive value for this QSAR model suggests its high predictive power and lack of over fitting.
- (8) **Fischer Statistics (F):** The F value of QSAR model was compared with their literature value at 95% level. The F value of this QSAR model is 19.36 (where  $F > F_{lit}$ ) suggests that the QSAR model is statistically significant at 95% level.

### Evaluation of Designed Compounds (A1-A12) from QSAR Model Developed

All the designed and synthesized compounds have been filtered with the developed QSAR model and their  $\log 1/C$  values have been predicted which is given in Table 17.



**Figure 1:** Structures of sulphonamide derivatives for developing the QSAR model.



Table 15: log1/C Values and Descriptors of Sulphonamide Derivatives Used to Derive QSAR Equation

Compound S. No.	log1/C			AlogP	ECI	LAI
	Observed	Predicted	Difference			
1	4.35	4.57	-0.22	0.0623	325	5.0764
2	4.45	4.41	0.04	-0.2073	260	4.0409
3	4.35	4.59	-0.24	0.0431	294	4.4324
4	4.47	4.76	-0.29	-0.2073	304	3.9765
5	4.66	4.68	-0.02	0.7377	262	4.2386
6	4.46	4.54	-0.08	0.7377	243	4.2519
7	4.6	4.55	0.05	0.7377	245	4.2478
8	4.8	4.79	0.01	0.0431	321	4.4250
9	4.8	4.56	0.24	-0.2073	279	4.0021
10	4.89	4.88	0.01	0.66	262	3.6797
11	4.89	4.92	-0.03	0.744	262	3.6550
12	4.99	4.84	0.15	0.744	243	3.5094
13	4.95	4.82	0.13	0.5738	262	3.7559
14	5.6	5.36	0.24	0.5046	298	3.0092
15	6	5.58	0.42	0.5046	325	2.9494
16	4.32	4.73	-0.41	1.1841	260	4.4875
17	4.8	4.92	-0.12	1.1064	260	3.9342
18	4.8	4.92	-0.12	1.1064	258	3.9074
19	5.4	5.30	0.1	1.0287	279	3.2765
20	5.55	5.15	0.4	1.0287	262	3.3275
21	5.1	4.92	0.18	0.1614	294	3.7230
22	5.55	5.73	-0.18	0.951	340	3.2527
23	5.41	5.45	-0.04	0.951	311	3.3990
24	5.64	6.10	-0.46	0.9573	340	2.3351
25	5.32	5.00	0.32	1.4071	391	6.4116

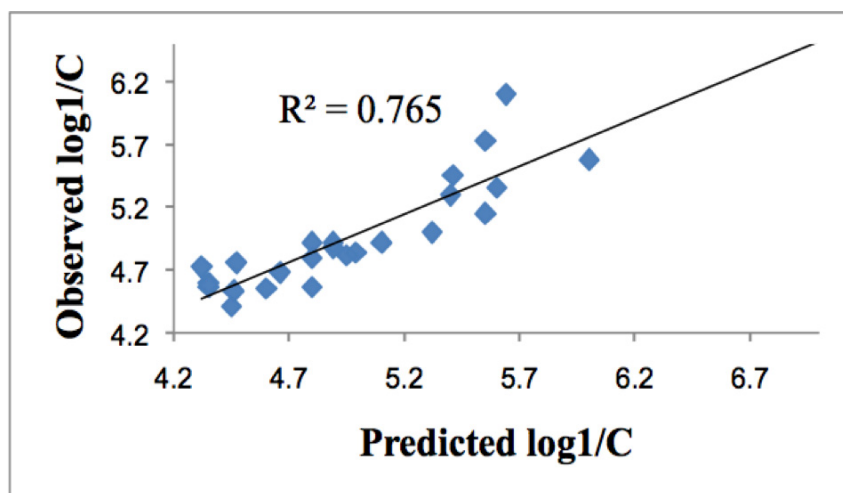


Figure 2: A plot of observed values and predicted values of log1/C for sulphonamide derivatives.

Table 16: Statistical Results of Model Validation

n/p(>=4)	r <sup>2</sup>	q <sup>2</sup>	r <sup>2</sup> -q <sup>2</sup> < 0.3	RMSD	Q	variance	F
8.33	0.765	0.7784	-0.013	0.0451	1.97	0.0607	19.36

Table 17: Descriptors and Predicted log1/C Values of Designed Compounds

Compound S. No.	Predicted log1/C	AlogP	ECI	LAI
A1	3.442427	-4.1763	341	0
A2	3.442073	-4.2087	445	0
A3	3.528493	-3.5416	298	-0.68119
A4	3.436394	-4.2531	341	0
A5	3.480161	-3.5988	360	0.147473
A6	3.450361	-3.8366	326	0.338429
A7	3.429939	-3.6508	418	1.016806
A8	3.362193	-5.192	320	0
A9	3.416152	-4.07	387	0.653385
A10	3.432445	-3.9882	455	0.498572
A11	3.432425	-4.0206	574	0.498234
A12	3.519576	-3.3535	405	-0.20488

An *in-silico* evaluation have been performed which include the QSAR and docking studies. On QSAR study the predicted bioactivity values ( $\log 1/C$ ) were found between 3.362193 and 3.528493. On the other hand, docking studies showed three proteins which significantly inhibited by the designed compounds, which are 1IKQ- Pseudomonas aeruginosa exotoxin A (inhibited by compounds A1, A2, A3, A4, A5, A6, A7, A10 and A11), 3AQB- heterodimeric hexaprenyl diphosphate synthase (inhibited by compounds A8 and A9) and 3M65- Bacillus subtilis Lon protease (inhibited by compound A12).

## CONCLUSION

In order to obtain substituted acyl chlorides, substituted acids were treated with thionyl chloride and then substituted acyl chloride/substituted benzyl chloride/substituted chlorobenzene were treated with aminosulfonamides in the presence of pyridine for obtaining the substituted 4-amino-benzenesulfonamides/N-acetyl-4-amino-benzenesulfonamides. The structures of synthesized compounds were confirmed by physical, analytical and elemental analysis.

The docking result of standard drug taken CID\_4539 (Norfloxacin) correlates well with the performance of compounds A1, A2, A3, A4, A5, A6, A7, A10 and A11 in docking study. Hence, it could be

concluded that molecular target responsible for the antimicrobial activity of substituted 4-amino-benzenesulfonamides/N-acetyl-4-amino-benzenesulfonamides may be pseudomonas aeruginosa exotoxin A. The other remaining proteins 1R4Z- Bacillus subtilis lipase A, 2CCZ- E-coli primosomal protein and 3KHX- Staphylococcus aureus metalloproteinase also showed prominent inhibition by the designed molecules. Although a systemic biochemical study of synthesized compounds is necessary to confirm the findings.

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## REFERENCES

- [1] Jones ME, Blosser-Middleton RS, Thornsberry C, Karlowsky JA, Sahm DF. The activity of levofloxacin and other antimicrobials against clinical isolates of Streptococcus pneumoniae collected worldwide during 1999-2002. Diagn Microbiol Infect Dis 2003; 47: 579-86. [http://dx.doi.org/10.1016/S0732-8893\(03\)00140-8](http://dx.doi.org/10.1016/S0732-8893(03)00140-8)
- [2] Fouda SI, Kadry AA, Shibl AM. Beta-lactam and macrolide resistance and serotype distribution among Streptococcus

- pneumoniae isolates from Saudi Arabia. *J Chemother* 2004; 16: 517-23.  
<http://dx.doi.org/10.1179/joc.2004.16.6.517>
- [3] Jenkins SG, Farrell DJ, Patel M, Lavin BS. Trends in anti-bacterial resistance among *Streptococcus pneumoniae* isolated in the USA, 2000-2003: PROTEK US years 1-3. *J Infect* 2005; 51: 355-63.  
<http://dx.doi.org/10.1016/j.jinf.2005.04.015>
- [4] Kaye KS, Engemann JJ, Fraimow HS, Abrutyn E. Pathogens resistant to antimicrobial agents: epidemiology, molecular mechanisms, and clinical management. *Infect Dis Clin North Am* 2004; 18: 467-11.  
<http://dx.doi.org/10.1016/j.idc.2004.04.003>
- [5] Cuellar-Rodriguez J, *et al.* Vancomycin-resistant enterococci, México City. *Emerg Infect Dis* 2007; 13: 798-99.  
<http://dx.doi.org/10.3201/eid1305.061421>
- [6] Tenover FC. Mechanisms of antimicrobial resistance in bacteria. *Am J Med* 2006; 119: S3-S10.
- [7] Murthy A, De Angelis G, Pittet D, Schrenzel J, Uckay I, Harbarth S. Cost-effectiveness of universal MRSA screening on admission to surgery. *Clin Microbiol Infect* 2010; 16: 1747-53.  
<http://dx.doi.org/10.1111/j.1469-0691.2010.03220.x>
- [8] Hong YL, Hossler PA, Calhoun DH, Meshnick SR. Inhibition of recombinant *Pneumocystis carinii* dihydropteroate synthetase by sulfa drugs. *Antimicrob Agents Chemother* 1995; 39: 1756-63.  
<http://dx.doi.org/10.1128/AAC.39.8.1756>
- [9] Furniss BS, Hannaford AJ, Smith PWG, Tatchell AR. In: Vogel's Textbook of Practical Organic Chemistry. Prentice Hall, New York 1989.
- [10] Naama H, Meir B, Bogdan W, Richard HF, Boris Y. Syntheses and evaluation of anticonvulsant profile and teratogenicity of novel amide derivatives of branched aliphatic carboxylic acids with 4-aminobenzenesulfonamide. *J Med Chem* 2010; 53: 4177-86.  
<http://dx.doi.org/10.1021/jm100170w>
- [11] Ajeet. Chlorambucil derivatives as antineoplastic agent: in-silico designing and docking. *Int J Cur Pharm Res* 2011; 3(4): 81-84.
- [12] Ajeet. Trans-disciplinary receptor binding of acyclovir to human phenylalanine hydroxylase: docking approach. *Int J Pharm Pharm Sci* 2012; 4(Suppl 3): 182-84.
- [13] Ajeet. In silico designing and characterization of Amiloride derivatives as ion channel modulator. *Med Chem Res* 2013; 22(2): 1004-10.  
<http://dx.doi.org/10.1007/s00044-012-0096-9>
- [14] Ajeet, Kumar P, Tripathi L. Virtual Screening Tool Based Designing and Evaluation of Novel Sulfonamide Derivatives as Anticonvulsant Agent – A Pharmacophoric Approach. *Int J Pharm Phytopharmacol Res* 2012; 2(3): 202-08.
- [15] Ajeet, Tripathi L, Kumar P. Designing of Novel 6(H)-1,3,4-Thiadiazine Derivatives as MMP12 Inhibitors: A MLR and Docking Approach. *Am J Pharmacol Sci* 2013; 1(2): 29-34.  
<http://dx.doi.org/10.12691/ajps-1-2-3>
- [16] Ajeet, Kumar A. Designing of Hybrid form of Benzothiazole-quinazoline as GABA-A Inhibitor with Anticonvulsant Profile: An in-silico Approach. *Am J Pharmacol Sci* 2013; 1(6): 116-20.  
<http://dx.doi.org/10.12691/ajps-1-6-2>
- [17] Verma RP, Hansch C. QSAR modeling of taxane analogues against colon cancer. *Eur J Med Chem* 2010; 45: 1470-77.  
<http://dx.doi.org/10.1016/j.ejmech.2009.12.054>
- [18] Verma RP, Hansch C. Matrix metalloproteinases (MMPs): Chemical-biological functions and (Q)SARs. *Bioorg Med Chem* 2007; 15: 2223-68.  
<http://dx.doi.org/10.1016/j.bmc.2007.01.011>
- [19] Ajeet. QSAR modeling of recombinant human stromelysin inhibitors– MLR approach. *Int J Adv Pharm Biol Sci* 2012; 2(2): 171-75.
- [20] Ajeet. Prediction of novel arylpiperazinyls (pyrrolidin-2-one derivatives) as antiarrhythmic agents – QSAR model development. *Int J Pharmac Arch* 2012; 1(1): 12-20.
- [21] Ajeet. Prediction of 44 novel Succinyl Hydroxamates and their carboxylic analogues against MMP2– QSAR modeling. *Novel Sci Int J Pharmac Sci* 2012; 1(11-12): 713-17.
- [22] Ajeet. Development of QSAR model for studying sulfonamide derivatives against carbonic anhydrase using multiple linear regression with McGowan volume and AlogP as molecular descriptors. *J Harmo Res Pharm* 2013; 2(1): 45-53.
- [23] Ajeet. QSAR modeling of beta-lactam antibiotic cephalosporin against transpeptidase using MLR method. *Eur J Bioinformat* 2013; 1: 1-7.
- [24] Ajeet, Kumar P, Tripathi L. QSAR Model Development for Studying Sulfonamide Derivatives as Carbonic Anhydrase Inhibitors– A MLR Approach. *Int J Pharm Res Tech* 2013; 5(1): 74-81.
- [25] Ajeet. QSAR Study of Methionine Aminopeptidase Inhibitors as Anti-cancer Agents Using MLR Approach. *Am J Pharmacol Sci* 2013; 1(5): 80-83.
- [26] Ajeet. Triterpene acid (3 $\beta$ -hydroxyurs-12-en-28-oic acid) against human lung cancer A-549 cells- multiple linear regression based QSAR modeling. *Eur J Bioinformat* 2013; 1: 8-14.