Synthesis, characterization and biological screening of sulfonamides derived form 2-phenylethylamine

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Abstract: In the present study, a series of *N*-substituted derivatives of 2-phenylethylamine has been synthesized. The reaction of 2-phenylethylamine (**1**) with benzene sulfonyl chloride (**2**) yielded *N*-(2-phenylethyl) benzenesulfonamide (**3**), which further on treatment with alkyl/acyl halides (**4a-i**) in the presence of sodium hydride furnished into *N*-substituted sulfonamides (**5a-i**)**.** These derivatives were characterized by IR, ^{1}H-NMR and EI-MS and then screened against acetyl cholinesterase (AChE), butyryl cholinesterase (BChE) and lipoxygenase enzyme (LOX) and were found to be potent inhibitors of butyryl cholinesterase only.

Keywords: 2-phenylethylamine, sulfonamides, butyryl cholinesterase, ^{1}H-NMR and EI-MS.

INTRODUCTION

The sulfonamides belong to distinctive class of compounds that constitute at least five different classes of pharmacologically active agents (Supuran *et al.,* 1999). The basic sulfonamide group –SO_2NH- occurs in various biological active compounds including antimicrobial drugs, antithyroid agents, antitumor antibiotics and inhibitors of carbonic anhydrase (Remko *et al.,* 2004). Sulfonamides are widely used to treat microbial infections by inhibiting the growth of gram negative and gram positive bacteria, some protozoa and fungi (Perlovich *et al.,* 2008). Clinically sulfonamides are used to treat several urinary tract infections and gastrointestinal infections (Gaded *et al.,* 2000). Sulfonamides that are aromatic or hetroaromatic are responsible for the inhibition of the growth of tumor cells. They act as antitumor agents by inhibiting the carbonic anhydrase. Sulfonamides are structurally similar to *p*-aminobenenzoiac acid (PABA) which is a cofactor that is needed by the bacteria for the synthesis of folic acid. Sulfonamides antibiotics inhibit the conversion of PABA into folic acid and thus ultimately inhibit the synthesis of purine and DNA. Sulfonamide antibiotics are used as veterinary medicines to treat infections in livestock herdsc (El-Sayed *et al.,* 2011 and Garcia-Galan *et al.,* 2008).

Literature survey revealed that minor modification in the structure of sulfonamide can lead to quantitative as well as qualitative changes in the biological activity. It prompted us to synthesize the various *N*-substituted derivatives of sulfonamides derived from 2-phenyethylamine with the goal of having lesser toxicity and improved activity. For this, the parent sulfonamide *N*-(2-phenylethyl)benzenesulfonamide (**3**), was first prepared by reacting benzenesulfonyl chloride with 2-phenyethylamine at room temperature in basic medium. Simple stirring gave the desired compound in excellent yield. Then it was further processed to obtain different new *N*-alkyl substituted sulfonamides.

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MATERIALS AND METHODS

General

Thin layer chromatography was done on silica gel coated plates that were G-25-UV_{254}. The wavelength of detection was 254 nm, by using ceric sulphate reagent. Different combinations of ethylacetate and *n*-hexane were used to check the purity of product giving single spot. IR spectrum was recorded in KBr by instrumenting Jasco-320-A spectrophotometer (wave number in cm^{-1}). $CDCl_3$ was used to record Proton NMR by using Bruker spectrometers. The frequency applied for this purpose is 500 MHz. Finnigan MAT-112 instrument was used to measure mass spectra and JMS-HX-110 spectrometer was used to record EI-MS. Griffin & George apparatus of melting point was used to record melting points of different products.

Butyryl cholinesterase (AChE, EC 3.1.1.7) and Acetyl cholinesterase (BChE, EC 3.1.1.8) belongs to the family serine hydrolases. Both of these enzymes have different specifications for substrate because they occupy the different active sites. Acetylcholine is blocked in cholinergic synapses. The major work of BChE and

AChE is to terminate the nerve impulse by the hydrolysis of acetylcholine neurotransmitter (Cyglar *et al.,* 1993; Tougu, 2001). BChE is found in case of Alzheimer's plaques at higher quantity. Cholinesterase inhibitors have significantly raised the quantity of acetylcholine for neuromuscular transmission (Gauthier, 2001). The search for novel cholinesterase inhibitors was considered very important for the advancement of new drug that are used to treat Alzheimer's disease and other diseases (Bertaccini, 1982).

Procedure for the synthesis of sulfonamide in aqueous medium

The nucleophilic substitution reaction of amine with benzene sulfonyl chloride was carried out as follows: a mixture of benzenesulfonyl chloride (10.0 mmol; 1.27 mL) and 2-phenylethylamine (10.0 mmol; 2.61 mL) was suspended in 50 mL water. The suspension pH was maintained at 9.0 to 10.0 by adding aqueous solution of a base (Na_2CO_3) at ambient temp. The solution was stirred and monitored by using analytical technique TLC for the completion of reaction. Then concentrated HCl was added gradually to adjust the pH to 2.0. The precipitates were collected by filtration, washed with distilled H_2O and dried to afford the title compound **3**. CH_3OH was used to dissolve the product and then it was re-crystallized by slow evaporation of the solvent, to generate colorless bead like crystals of *N*-(2-Phenylethyl)benzenesulfonamide. Yield 92%.

General procedure for the synthesis of N-alkyl substituted sulfonamides in DMF

The calculated amount of *N*-(2-Phenylethyl)benzenesulfonamide (0.1 mmol; **3**) was taken in a round bottomed flask (50 mL), then *N,N*-dimethyl formamide (DMF) (10 mL) was added to dissolve it followed by the addition of sodium hydride (0.1 mmol) to the mixture. The mixture was stirred for 30 minutes at room temperature and then slowly added the alkyl halide to the mixture and the solution was further stirred for three hours. The progress of reaction was monitored *via* TLC till single spot. Distilled water was added in the flask and the product was obtained by solvent extraction.

Acetyl cholinesterase assay

The inhibition activity of AChE was performed by acquiring the procedure of (Ellman *et al*., 1961) with minor alterations. 100 µL was the total volume of the reaction. In this 60 µL buffer Na_2HPO_4 having 50 mM concentration and pH 7.7 was used. The compound which was to be tested taken as 10 µL test (0.5 mM well^{-1}), similarly 10 µL (0.005 unit well^{-1}) of enzyme was used. All the contents were well mixed and pre-read at 405 nm. Then these were pre-incubated for 10 min at 37ºC temp. The reaction was initiated by the addition of 10 µL of 0.5 mM well^{-1} substrate (acetylthiocholine iodide), followed by the addition of 10 µL DTNB (0.5 mM well^{-1}). After 15 min of incubation at 37ºC, absorbance was measured at 405 nm. Synergy HT (BioTek, USA) 96-well plate reader was used in all experiments. All experiments were carried out with their respective controls in triplicate. Eserine (0.5 mM well^{-1}) was used as a positive control. The percent inhibition was calculated by the help of following equation.

$$\text{Inhibition}\,(100) = \frac{\text{Control - Test}}{\text{Control}} \times 100$$

Butyryl cholinesterase assay

The inhibition activity BChE was done by aquiring method of (Ellman *et al*., 1961) with minute changes. 100 µL was the total volume of the reaction it contained 60 µL Na_2HPO_4 which was act as buffer, having 50 mM concentration 7.7 pH. Ten µL test compound 0.5 mM well^{-1}, followed by the addition of 10 µL (0.5 unit well^{-1}) BChE. The reaction mixture was stirred well and read prior at the wavelength 405 nm and then also prior-incubated for ten minutes at 37ºC. The reaction was started by adding ten µL of 0.5 mM well^{-1} BChE bromide 10 µL DTNB, 0.5 mM well^{-1} was also added and incubated for 15 min and wavelength 405 nm was used to record absorbance. All the experiments were read *via* Synergy HT (BioTek, USA) 96-well plate reader. Each and every experiment was conceded by their particular controls at triplicate. Eserine (0.5 mM well^{-1}) was act as +ve control. The equation that was used to calculate % inhibition was:

$$\text{Inhibition}\,(100) = \frac{\text{Control - Test}}{\text{Control}} \times 100$$

EZ–Fit Enzyme kinetics software (Perella Scientific Inc. Amherst, USA) was used to manipulate IC_{50} values of products.

Lipoxygenase assay

Activity of Lipoxygenase was carried out by acquiring the method of (Tappel, 1953; Evans, 1897 and Baylac *et al.,* 2003) with small changes. Entire volume of the mixture was 200 µL that have 150 µL Na_2PO_4 buffer (100 mM, pH 8.0), and 10 µL compound under consideration and also 15µL LOX enzyme (Sigma, USA). All the stuff was uniformly mixed and read prior at 234 nm. Preincubation was done for at least 10 min. at ambient temperature. The initiation of reaction was performed by introducing 25 µL solution of substrate. The alteration in absorbance of sample was examined for every 6 min at wavelength of 234 nm. Entire experiment was performed *via* Synergy HT (BioTek, USA) 96-well plate reader. Each and every experiment was conceded by their particular controls at triplicate. The +ve and -ve controls were incorporated in the given assay. The +ve control was used for this purpose was Quercetin (0.5 mM well^{-1}). The equation that was used to calculate % inhibition was,

$$\text{Inhibition}\,(100) = \frac{\text{Control - Test}}{\text{Control}} \times 100$$

***N*-(2-Phenylethyl)benzenesulfonamide (3)**
White needle like crystals, Yield 89%. IR (KBr): ν_{max}: 3432 (N-H), 3058 (Ar-H), 1623, 1517 (aromatic C=C), 1341 (-SO_2-NH-); ^{1}H-NMR (500 MHz, $CDCl_3$): δ 7.80 (dd, J = 8.5, 2.1 Hz, 2H, H-2′, H-6′), 7.55 (m, 2H, H-3, H-5), 7.47 (m, 1H, H-4′), 7.26 (m, 2H, H-3′, H-5′), 7.20 (m, 2H, H-2, H-6), 7.06 (dd, J = 7.5, 1.5 Hz, 1H, H-4), 3.22 (t, J = 6.5 Hz, 2H, CH_2-8) and 2.76 (t, J = 6.5 Hz, 2H, CH_2-7); EIMS: *m/z* 261 $[M]^+$, 196 $[M\text{-}SO_2]^+$, 170 $[M\text{-}C_6H_5CH_2]^+$, 141 $[C_6H_5SO_2]^+$.

***N*-Methyl-*N*-(2-phenylethyl)benzenesulfonamide (5a)**
Pale yellow greasy liquid, Yield 65%. IR (KBr): ν_{max}: 3052 (Ar-H), 1628, 1523 (aromatic C=C), 1344 (-SO_2-NH-); ^{1}H-NMR (500 MHz, $CDCl_3$): δ 7.75 (dd, J = 8.5, 2.1 Hz, 2H, H-2′, H-6′), 7.56 (m, 2H, H-3, H-5), 7.49 (m, 1H, H-4′), 7.27 (m, 2H, H-3′, H-5′), 7.21 (m, 2H, H-2, H-6), 7.15 (dd, J = 7.5, 1.5 Hz, 1H, H-4), 3.25 (t, J = 7.0 Hz, 2H, CH_2-8), 2.84 (t, J = 7.0 Hz, 2H, CH_2-7) and 2.74 (s, 3H, H-1′′); EIMS: *m/z* 275 $[M]^+$, 211 $[M\text{-}SO_2]^+$, 184 $[M\text{-}C_6H_5CH_2]^+$, 141 $[C_6H_5SO_2]^+$.

***N*-Ethyl-*N*-(2-phenylethyl) benzene sulfonamide (5b)**
Mustered colored gammy liquid, Yield 70%. IR (KBr): ν_{max}: 3054 (Ar-H), 1621, 1522 (aromatic C=C), 1342 (-SO_2-NH-); ^{1}H-NMR (500 MHz, $CDCl_3$): δ 7.77 (dd, J = 8.5, 2.1 Hz, 2H, H-2′, H-6′), 7.54 (m, 2H, H-3, H-5), 7.47 (m, 1H, H-4′), 7.28 (m, 2H, H-3′, H-5′), 7.21 (m, 2H, H-2, H-6), 7.14 (dd, J = 7.5, 1.5 Hz, 1H, H-4), 3.34 (t, J = 8.0 Hz, 2H, CH_2-8), 2.87 (t, J = 8.0 Hz, 2H, CH_2-7), 3.22 (q, J = 7.0 Hz, 2H, H-1′′) and 1.09 (t, J = 7.0 Hz, 3H, H-2′′); EIMS: *m/z* 289 $[M]^+$, 225 $[M\text{-}SO_2]^+$, 198 $[M\text{-}C_6H_5CH_2]^+$, 148 $[M\text{-}C_6H_5SO_2]^+$.

***N*-Iso-propyl-*N*-(2-phenylethyl)benzenesulfonamide (5c)**
Brown colored gammy liquid. Yield 68%. IR (KBr): ν_{max}: 3051 (Ar-H), 1630, 1523 (aromatic C=C), 1346 (-SO_2-NH-); ^{1}H-NMR (500 MHz, $CDCl_3$): δ 7.82 (dd, J = 8.5, 2.1 Hz, 2H, H-2′, H-6′), 7.78 (m, 2H, H-3, H-5), 7.53 (m, 1H, H-4′), 7.47 (m, 2H, H-3′, H-5′), 7.28 (m, 2H, H-2, H-6), 7.19 (dd, J = 7.5, 1.5 Hz, 1H, H-4), 3.00 (t, J = 8.0 Hz, 2H, CH_2-8), 2.74 (t, J = 8.0 Hz, 2H, CH_2-7), 4.10 (m, 1H, H-1′′) and 1.02 (d, J = 7.0 Hz, 6H, CH_3-2′′, CH_3-3′′); EIMS; *m/z* 303 $[M]^+$, 288, 239 $[M\text{-}SO_2]^+$, 212 $[M\text{-}C_6H_5CH_2]^+$, 162 $[M\text{-}C_6H_5SO_2]^+$

***N*-Butyl-*N*-(2-phenylethyl) benzene sulfonamide (5d)**
Pale yellow greasy liquid, Yield 78%. IR (KBr): ν_{max}: 3046 (Ar-H), 1623, 1525 (aromatic C=C), 1346 (-SO_2-NH-); ^{1}H-NMR (500 MHz, $CDCl_3$): δ 7.78 (dd, J = 8.5, 2.1 Hz, 2H, H-2′, H-6′), 7.54 (m, 2H, H-3, H-5), 7.47 (m, 1H, H-4′), 7.26 (m, 2H, H-3′, H-5′), 7.19 (m, 2H, H-2, H-6), 7.06 (dd, J = 7.5, 1.5 Hz, 1H, H-4), 3.32 (t, J = 8.0 Hz, 2H, CH_2-8), 3.12 (t, J = 8.0 Hz, 2H, CH_2-7), 2.74 (t, J = 7.0 Hz, 2H, CH_2-1′′), 1.48 (m, 2H, CH_2-2′′), 1.27 (m, 2H, CH_2-3′′) and 0.88 (t, J = 7.5 Hz, 3H, CH_3-4′′); EIMS: *m/z* 317 $[M]^+$, 253 $[M\text{-}SO_2]^+$, 226 $[M\text{-}C_6H_5CH_2]^+$, 176 $[M\text{-}C_6H_5SO_2]^+$

***N*-Pentyl-*N*-(2-phenylethyl) benzene sulfonamide (5e)**
Rust colored greasy liquid, Yield 63%. IR (KBr): ν_{max}: 3053 (Ar-H), 1625, 1523 (aromatic C=C), 1343 (-SO_2-NH-); ^{1}H-NMR (500 MHz, $CDCl_3$): δ 7.78 (dd, J = 8.5, 2.1 Hz, 2H, H-2′, H-6′), 7.55 (m, 2H, H-3, H-5), 7.49 (m, 1H, H-4′), 7.24 (m, 2H, H-3′, H-5′), 7.20 (m, 2H, H-2, H-6), 7.14 (dd, J = 7.5, 1.5 Hz, 1H, H-4), 3.32 (t, 8.0, 2H, CH_2-8), 3.12 (t, J = 8.0 Hz, 2H, CH_2-7), 2.74 (t, J = 7.0 Hz, 2H, CH_2-1′′), 1.49 (m, 2H, CH_2-2′′), 1.26 (m, 2H, CH_2-3′′, CH_2-4′′′) and 0.85 (t, J = 7.5 Hz, 3H, CH_3-5′′); EIMS: *m/z* 331 $[M]^+$, 267 $[M\text{-}SO_2]^+$, 240 $[M\text{-}C_6H_5CH_2]^+$, 190 $[M\text{-}C_6H_5SO_2]^+$.

***N*-Allyl-*N*-(2-phenylethyl)benzenesulfonamide (5f)**
Brown colored needle like crystals, Yield 72%, IR (KBr): ν_{max}: 3057 (Ar-H), 1627, 1523 (aromatic C=C), 1345 (-SO_2-NH-); ^{1}H-NMR (500 MHz, $CDCl_3$): δ 7.78 (dd, J = 8.5, 2.1Hz, 2H, H-2′, H-6′), 7.55 (m, 2H, H-3, H-5), 7.47 (m, 1H, H-4′), 7.26 (m, 2H, H-3′, H-5′), 7.19 (m, 2H, H-2, H-6), 7.12 (dd, J = 7.5, 1.5 Hz, 1H, H-4), 5.61 (m, 1H, H-2′′), 5.17 (dd, J =1.6, 17.3 Hz, H_b-3′′), 5.13 (dd , J = 1.2, 10 Hz, H_a-3′′), 3.79 (d, J = 6.5 Hz, H-1′′), 3.33 (t, J = 7.0 Hz, 2H, CH_2-8), 2.82 (t, 7.0 Hz, 2H, CH_2-7); EIMS: *m/z* 301 $[M]^+$,237 $[M\text{-}SO_2]^+$, 210 $[M\text{-}C_6H_5CH_2]^+$, 160 $[M\text{-}C_6H_5SO_2]^+$.

4-(*N*-(2-Phenylethyl)-*N*-(benzenesulfonyl)aminoethylacetoacetate (5g)
Brown amorphous powder, Yield 68%. IR (KBr): ν_{max}: 3051 (Ar-H), 1622, 1519 (aromatic C=C), 1341 (-SO_2-NH-); ^{1}H-NMR (500 MHz, $CDCl_3$): δ 7.78 (dd, J = 8.5, 2.1 Hz, 2H, H-2′, H-6′), 7.56 (t, J = 7.5, 2H, H-3, H-5), 7.47 (t, J = 7.0 1H, H-4′), 7.27 (m, 2H, H-3′, H-5′), 7.20 (m, 2H, H-2, H-6), 7.04 (dd, J = 7.5, 1.5 Hz, 1H, H-4), 4.35 (br s, 2H, CH_2-4′′), 3.27 (t, 8.0, 2H, H-8), 3.23 (s, 2H, H-2′′), 2.74 (t, J = 8.0 Hz, 2H, H-7), 4.42 (q, 2H, CH_2-1′′′) and 1.40 (t, 3H, CH_3-2′′′); EIMS: *m/z* 389 $[M]^+$, 325 $[M\text{-}SO_2]^+$, 298 $[M\text{-}C_6H_5CH_2]^+$, 248 $[M\text{-}C_6H_5SO_2]^+$.

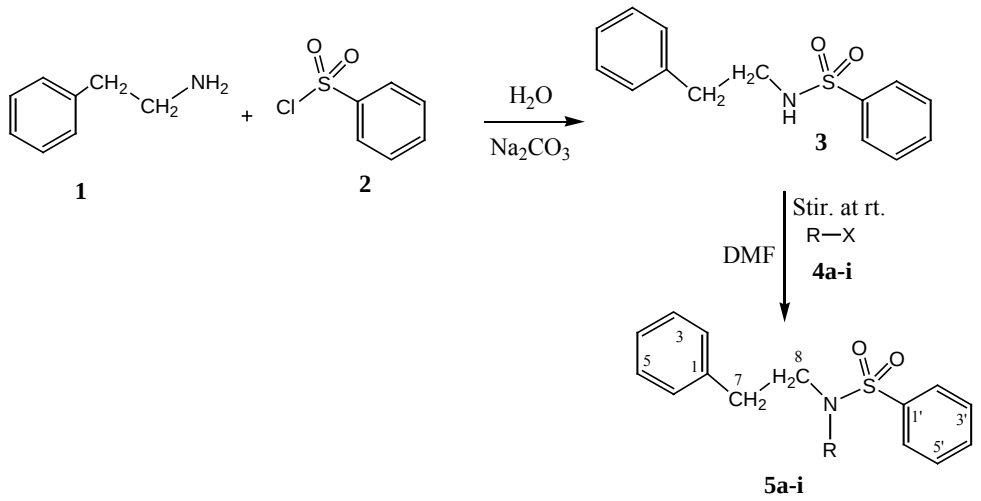
***N*-Acetyl-*N*-(2-phenylethyl)benzenesulfonamide (5h)**
Pale yellow greasy liquid, Yield 68%. IR (KBr): ν_{max}: 3051 (Ar-H), 1627, 1527 (aromatic C=C), 1349 (-SO_2-NH-); ^{1}H-NMR (500 MHz, $CDCl_3$): δ 7.87 (dd, J = 8.5, 2.1 Hz, 2H, H-2′, H-6′), 7.65 (m, 1H, H-4′), 7.56 (m, 2H, H-3′, H-5′), 7.47 (m, 2H, H-3, H-5), 7.29 (m, 2H, H-2, H-6), 7.23 (dd, J = 7.5, 1.5 Hz, 1H, H-4), 3.39 (t, J = 8.0 Hz, 2H, CH_2-8), 2.99 (t, J = 8.0 Hz, 2H, CH_2-7) and 2.28 (s, 3H, CH_3-1′′′); EIMS: *m/z* 303 $[M]^+$, 260 $(M\text{-}COCH_3)^+$, 239 $[M\text{-}SO_2]^+$, 212 $[M\text{-}C_6H_5CH_2]^+$, 162 $[M\text{-}C_6H_5SO_2]^+$.

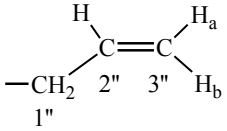
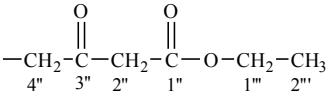
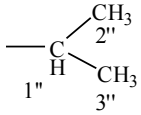
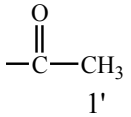
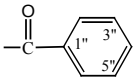
***N*-Benzoyl-*N*-(2-phenylethyl)benzenesulfonamide (5i)**
White amorphous powder, Yield 68%. IR (KBr): ν_{max}: 3056 (Ar-H), 1632, 1529 (aromatic C=C), 1349 (-SO_2-NH-); ^{1}H-NMR (500 MHz, $CDCl_3$): δ 7.87 (dd, J = 8.0,

Table: Evaluation of biological activities of the compounds (n=3, mean±sem)

Sample No.	DPPH		AChE		BChE		LOX	
	(%) at 0.5 mM	(IC_{50}) µmoles	(%)at 0.5mM	(IC_{50}) µmoles	(%) at 0.5mM	(IC_{50}) µmoles	(%) at 0.5mM	(IC_{50}) µmoles
3	12.53±0.97	Nil	41.30±0.81	Nil	57.50±0.78	288±0.91	9.78±1.21	Nil
5a	7.24±0.59	Nil	40.52±0.47	Nil	80.45±0.98	24±0.99	66.20±0.66	255±0.93
5b	8.97±0.78	Nil	41.17±0.95	Nil	89.47±0.87	2±1.01	45.98±0.45	Nil
5c	8.85±0.75	Nil	31.95±0.83	Nil	87.80±0.45	17±0.85	46.41±1.06	Nil
5d	11.46±0.91	Nil	32.34±1.01	Nil	76.21±1.02	19±0.81	24.02±0.92	Nil
5e	13.90±0.73	Nil	50.00±0.89	<500	60.50±0.78	294±0.93	60.22±0.76	278±0.87
5f	11.52±0.59	Nil	50.12±0.84	<500	82.80±0.59	13±0.78	14.02±0.69	Nil
5g	35.87±1.02	Nil	50.91±0.83	<500	55.91±0.98	293±1.10	17.93±1.05	Nil
5h	10.75±0.68	Nil	32.08±0.91	Nil	81.82±0.48	63±1.03	60.00±0.93	261±0.88
5i	22.09±1.02	Nil	38.70±0.77	Nil	72.88±0.65	21±0.77	3.80±0.84	Nil
Control	Quercetin	16.96±0.14	Eserine	0.04±0.001	Eserine	0.85±0.001	Quercetin	37.12±0.07

Note: IC_{50} values (concentration at which there is 50% enzyme inhibition) of compounds were calculated using EZ–Fit Enzyme kinetics software (Perella Scientific Inc. Amherst, USA).
DPPH = 1,1-diphenyl-2-picrylhydrazyl radical, LOX = Lipoxygenase, AchE = Acetyl cholinesterase, BChE = Butyryl cholinesterase.



Compound	R	Compound	R
5a	—CH_3 1"	5f	
5b	—CH_2—CH_3 1" 2"	5g	
5c		5h	
5d	—CH_2—CH_2—CH_2—CH_3 1" 2" 3" 4"	5i	
5e	—CH_2—CH_2—CH_2—CH_2—CH_3 1" 2" 3" 4" 5"		

Scheme: Synthesis of *N*-substituted sulfonamides derived from 2-phenylethylamine.

2.0 Hz, 2H, H-2′, H-6′), 7.78 (dd, J = 8.0, 2.0 Hz, 2H, H-2′′, H-6′′), 7.60 (m, 1H, H-4′′), 7.56 (m, 1H, H-4′), 7.49 (m, 2H, H-3′′, H-5′′), 7.43 (m, 2H, H-3′, H-5′), 7.32 (m, 2H, H-3, H-5), 7.19 (m, 2H, H-2, H-6), 7.02 (dd, J = 7.5, 1.5 Hz, 1H, H-4), 4.04 (t, J = 8.0 Hz, 2H, CH_2-8) and 2.96 (t, J = 8.0 Hz, 2H, CH_2-7); EIMS: *m/z* 365 $[M]^+$, 301 $[M-SO_2]^+$, 274 $[M-C_6H_5CH_2]^+$, 260 $(M-COC_6H_5]^+$, 224 $[M-C_6H_5SO_2]^+$.

RESULTS

In the present investigation, a series of *N*-alkyl/aryl substituted sulfonamides was synthesized derived from 2-phenylethylamine. The parent compound *N*-(2-phenylethyl)benzenesulfonamide **(3),** was prepared by using 2-phenylethylamine (**1**) and benzene sulfonyl chloride (**2**). Reaction of **3** with different electrophiles (**4a-i**) yielded a series of *N*-substituted sulfonamides (**5a-i**) as represented in Scheme 1. Synthesis of all these derivatives was performed in DMF (*N*,*N*-dimethylformamide) using sodium hydride (NaH) as the base. Complete conversion was achieved within 2 to 3 hours by simple stirring at room temperature. The product was isolated by adding cold water in the reaction mixture and subsequently, it was taken out through solvent extraction method by chloroform/ethyl acetate. The structure of the parent compound and its *N*-substituted derivatives were confirmed by ^{1}H-NMR, IR and mass spectral data as described in experimental section.

Enzyme Inhibition Studies

The results of *in vitro* enzyme inhibition activity of the synthesized compounds against acetyl cholinesterase, butyryl cholinesterase and lipoxygenase are presented in table-1.

DISCUSSION

The parent compound **3** was synthesized as white needle like crystals. The molecular formula $C_{14}H_{15}NO_2S$ was established by HR-MS showing molecular ion peak at *m/z* 261.343 (calcd for $C_{14}H_{15}NO_2S$, 261.340). The IR spectrum revealed the presence of a sulfonyl group (1338, 1447 cm^{-1}) and –NH- (3100 cm^{-1}) group in the molecule. The EI-MS gave a distinct peak at *m/z* 197 after the removal $–SO_2$ group and further a peak was observed at *m/z* 156 which showed the presence of phenylethyl group in the molecule. In the aromatic region of the ^{1}H-NMR spectrum, the signals appeared at δ 7.80 (dd, J = 8.5, 2.1 Hz, 2H, H-2′, H-6′), 7.47 (m, 1H, H-4′) and 7.26 (m, 2H, H-3′, H-5′), due to downfield shift, these protons were assigned to the mono substituted aromatic ring bearing withdrawing sulfonyl group and the signals which appeared at δ 7.55 (m, 2H, H-3, H-5), 7.20 (m, 2H, H-2, H-6) and 7.06 (dd, J = 7.5, 1.5 Hz, 1H, H-4), were assigned to the protons of other mono substituted aromatic ring. In the aliphatic region of the ^{1}H-NMR spectrum, signals which resonated at δ 3.22 (t, J = 6.5 Hz, 2H, CH_2-8) and 2.76 (t, J = 6.5, 2H, CH_2-7); indicated the presence of two adjacent methylene groups in the molecule. On the basis of above cumulative evidences, the structure of compound **3** was assigned as *N*-(2-phenylethyl)benzenesulfonamide. On the basis of similar spectral characterization, the structures of the synthesized derivatives **5a-I** were identified.

The screening of the synthesized compounds revealed that these were inactive against acetyl cholinesterase (AchE) but exhibited good inhibitory potential against butyryl cholinesterase (BChE) as it was evident from their IC_{50} values (table). Among these, *N*-ethyl-*N*-(2-phenylethyl)benzenesulfonamide (**5b**) and *N*-allyl-*N*-(2-phenylethyl) benzenesulfonamide (**5f**) were found to be the most potent inhibitor having IC_{50} value of 02±1.01 and 13±0.78 µmoles respectively, relative to eserine, a reference standard with IC_{50} value of 0.85±0.001 µmoles, probably due to the substitution of ethyl and allyl group respectively in these molecules. The screening against lipoxygenase enzyme exposed that the compounds *N*-methyl-*N*-(2-phenylethyl)benzenesulfonamide (**5a**), *N*-pentyl-*N*-(2-phenylethyl)benzenesulfonamide (**5e**) and *N*-acetyl-*N*-(2-phenylethyl)benzenesulfonamide (**5h**) exhibitted inhibitory potential, however all other compounds were inactive. Similarly all these compounds displayed no scavenging potential of the 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical.

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