Synthesis, Characterization, and in *silico* Studies of Novel Alkanoylated 4-Methylphenyl sulphonamoyl Carboxylic Acids as Potential Antimicrobial and Antioxidant Agents

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ABSTRACT

The synthesis of new alkanoylated 4-methylphenyl sulphonamoyl carboxylic acids, their molecular docking, and antimicrobial and antioxidant activities were reported. The process involved the mild reaction of acetic anhydride and sodium acetate with 4-methylsulphonamoyl carboxylic acids. The characterization of the compounds was done using FTIR, ^{1}H-NMR, ^{13}C-NMR, and elemental analysis. They were tested for their antimicrobial activities against selected human pathogens such as *Pseudomonas aeruginosa, Salmonella typhi, Candida albicans, Aspergillusniger*, *Staphylococcus aureus, Escherichia coli,* and *Bacillus subtilis.* The antioxidant study of the compounds was evaluated *in vitro* by the inhibition of generated stable 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical. The *in silico* study was carried out in order to study five different prevalent disease conditions, namely: bacterial infections, fungal infections, oxidative stress, trypanosomiasis, and malaria. The findings demonstrated that compounds **2b and 2c** exhibited the best *in vitro* antibacterial and antifungal activities compared to ofloxacin and fluconazole. The antioxidant study revealed that compound **2a** was the most excellent antioxidant agent. The molecular docking study revealed that compounds **2a, 2b, 2c, 2e,** and **2f** exhibited excellent antibacterial, antifungal, antioxidant, antitrypanosomal, and antimalaria activities compared to the corresponding standard drugs such as Penicillin, Ketoconazole, α-Tocopherol, Melarsoprol, and Chloroquine, respectively. The physicochemical evaluations showed that all the compounds were drug-like according to "Lipinski's rule of five".The synthesized compounds were found to be potent antibacterial, antifungal, antioxidant, and antitrypanosomal agents.

Key Words: *Alkanoylation; 4-methylphenyl sulphonamoyl carboxylic acids; Antibacterial activity; antifungal activity; Antioxidant activity, molecular docking.*

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INTRODUCTION

Alkanoylation is the process of incorporating an acyl group (R-C=O-) into a compound. This reaction method is also known as acylation, in which the acylating agent provides the needed acyl group. Acid anhydrides and acyl halides are generally utilized as acylating agents. Acyl moiety is often gotten by removing one or more hydroxyl groups from oxoacid [1]. Alkanoylation process leads to the prevention of rearrangement reactions that would have been possible with alkylation [2]. It has also been found to protect the amino group of sulphonamide during aminolysis in order to ensure regioselectivity [3, 4]. Fundamentally, sulphonamides are commonly employed in the management of acute systematic or local infections.

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Many critical disease conditions including, coccidiosis, mastitis, metritis, respiratory infections, and toxoplasmosis are also treatable with sulphonamoyl compounds [5, 6]. Early and prompt administered of these drugs in the course of chronic infections give better results. However, in severe cases, the first doses are to be intravenously administered to minimize the time lag between dose and effect [7]. Compounds having sulphonamoyl functional group are crucial in medicinal chemistry for the design of drugs with a broad spectrum of biological activities. Also in organic chemistry study and application, sulfonamides have numerous importances. Industrially, sulphonamides are useful in the production of some health products, food colorants, and others. Considering the inherent ability of sulfonamides to cure high profile diseases such as arthritis, cancer, and osteoporosis, their syntheses are very crucial [8]. In spite of the fact that the incorporation of acyl group into sulphonamide compounds enhances their biological activities and improves their drug potency, only a few research have been reported on the successful acylation and biological evaluation of these important pharmaceutical compounds. Moreover, a little or no attention has been given to the synthesis of alkanoylated sulphonamides despite the fact that the acyl group component has been found to exist in major biochemical molecules, and are essential in the formation of molecules like DNA, proteins, carbohydrates, and lipids [9, 10]. Biochemically, acyl group-containing compounds such as Acetyl-*CoA* are required in several biosynthetic processes [11]. Considering the numerous pharmacological importances of acyl and sulphonamide functionalities, it was found necessary to synthesize alkanoylated 4-methylphenyl sulponamoyl carboxylic acids and evaluate their antimicrobial and antioxidant activities, as well as molecular docking, in order to maximize the synergistic actions arising from their successful coupling and this underscores the novelty of this study.

MATERIALS AND METHODS

Chemistry

Reagents were purchased from Sigma Aldrich. Melting points of the synthesized compounds were determined using electrothermal melting point apparatus and were uncorrected. Infrared spectra data were recorded on 8400s Fourier Transform Infrared (FTIR) (ABU,Zaria, Kaduna State, Nigeria). Nuclear Magnetic Resonance (^{1}H-NMR and ^{13}C-NMR) were run on 400MHz using NMR spectrophotometer at Sandeep VermaLabouratory, Department of Chemistry, Indian Institute of Technology, Kanpur. Chemical shifts were reported in δ scale (neat) using tetramethylsilane as a standard. The elemental analysis was carried out with elemental analyzer (Exeter Analytical Inc.model:CE440). Precipitation of the tittle compounds was in analytical grade and the reactions were monitored using TLC. The antimicrobial studies were carried out at the Department of Microbiology, University of Nigeria, Nsukka while the antioxidant studies were carried out at the Biochemistry Department, University of Nigeria, Nsukka

SYNTHESIS OF ALKANOYLATED 4-METHYLPHENYL SULPHONAMIDES

2g of various 4-methyphenyl sulphonamoyl carboxylic acids was weighed into a beaker, 9ml of concentrated hydrochloric acid and 25ml of distilled water were added to the beaker followed by vigorous stirring to ensure homogeneous mixture. 16.0g of sodium carbonate was dissolved in 50ml distilled water in a separate beaker. Then, 13.5ml of acetic anhydride was added in small portion over an interval of 1 hour to the 4-methyphenyl sulphonamoyl carboxylic acid solution after which it was poured into the sodium acetate solution. The reaction mixture was stirred thoroughly with a glass rod and immersed in an ice bath for 2 hours and filtered to afford the various alkanoylated 4-methyphenyl sulphonamoyl carboxylic acids (**2a-2f**) in good to excellent yields (70-97%).

2-{Acetyl[(4-methylphenyl)sulfonyl]amino}propanoic acid (2a)

Yield=2.03g (91.4%), mp=108-109 ˚C, IR (KBr) cm^{-1}: 3312 (O-H of COOH); 3094 (N-H); 3001 (C-H); 1923 (C-H aromatic); 1703, 1691(C=O); 1491, 1402 (C=C); 1311, 1292 (2S=O);1170 (SO_2-NH); 1115 (C-N); 741 (Ar-H). ^{1}HNMR (CD_3CN, 400MHZ) δ: 7.968 (d, J= 8.Hz, 2H, ArH), 7.523 (d, J= 8.4 Hz, 2H, ArH), 2.732 (s, 3H, CH_3-C=O), 2.492 (s,3H, CH_3-Ar), 1.985-1.968 (m, IH, CH), 1958 (d, J=1.2Hz, 3H, CH_3-CH). ^{13}CNMR (CD_3N, 400MHz) δ: 171.253, 170.244 (C=O); 147.937, 141.130, 130.613, 129.474, 126.355, 117.356 (aromatic carbons); 58.141, 51.344, 49.553 43.915 (aliphatic carbons). Anal.calcd(%)for $C_{12}H_{15}NO_5S$ (285.32): C, 50.48; H, 5.27; N, 4.92; S, 11.23. Found: C, 50.52; H, 5.31; N, 4.93; S, 11.26.

2-{Acetyl[(4-methylphenyl)sulfonyl]amino}-3-sulfanylpropanoic acid (2b)

Yield=1.95g (89.8%), mp=181-182 ˚C, IR (KBr) cm^{-1}: 3308 (O-H of COOH), 3004 (N-H), 2919 (C-H alphatic); 2960 (S-H); 2088 (C-H aromatic); 1793, 1622 (C=O); 1660, 1656 (C=C); 12987,1193 (S=O); 1155 (SO_2NH); 1137 (C-N); 785 (Ar-H). ^{1}HNMR (DMSO, 400MHz) δ: 7.930 (m, 2H, ArH),7.713 (m, 2H, ArH), 7.645-7.627(m, IH, ArH), 7.598-7.571 (m, IH, ArH); 6.577 (s, IH, NH);

3.354 (s, 2H, NH_2); 2.487 (s,3H, CH_3-C=O). ^{13}CNMR (DMSO, 400MHz) δ: 172.212, 170.122 (C=O); 137.176, 137.063, 133.822, 133.689. 133.625, 132.692 (aromatic carbons); 40.637, 40.425, 40.212, 39.988 (aliphatic carbons). Anal.calcd (%) for $C_{1`2}H_{15}NO_5S_2$ (317.39): C, 45.38; H, 4.74; N, 4.42; S, 20.18. Found: C, 45.41; H, 4.76; N, 4.39; S, 20.22.

2-{Acetyl[(4-methylphenyl)sulfonyl]amino}-4-(methylsulfanyl)butanoic acid(2c)

Yield=2.17g (93.7%), mp.217-218 ˚C, IR(KBr) Cm^{-1}: 3448(O-H of COOH); 3251(N-H); 2923(C-H), 1919 (C-H aromatic), 1719, 1671 (C=O), 1654,1623 (C=C), 1232, 1165 (S=O); 1089 (SO_2NH), 1040 (C-N); 822 (Ar-H). ^{1}HNMR ($CDCL_3$ /C_6D_6, 400MHz) δ: 7.172 (m, 2H, ArH); 7.175 (m, 2H, ArH). ^{13}(NMR ($CDCL_3$/C_6D_6, 400MHz) δ:176.135, 170.123 (C=O); 137.412, 133.830, 133.648, 128.458, 127.980, 127.737 (aromatic carbons); 77.464, 77.145, 76.811, 71.342, 68.566, 63.457 (aliphatic carbons). Anal.calcd (%) for $C_{14}H_{19}NO_5S_2$ (345.43): C, 48.65; H, 5.51; N, 4.06; S, 18.54. Found: C, 48.68; H, 5.48; N, 4.10; S, 18.49.

2-{Acetyl[(4-methylphenyl)sulfonyl]amino}--3-hydroxypropanoic acid(2d)

Yield=2.31g (96.4%); mp.201-202 ˚C, IR (KBr) cm^{-1}: 3653 (free OH); 3297 (OH of COOH); 3073 (C-H aliphatic); 1998 (C-H aromatic); 1820, 1735 (C=O); 1649, 1605 (C=C); 1329, 1240 (S=O); 1172(SO_2-NH), 1078 (C-N); 733(Ar-H). ^{1}HNMR (DMSO/$CDCL_3$) 400MHz) δ: 10.245 (m, 2H, OH); 7.495-7.475 (d, J= 8.0Hz, 2H, ArH); 7.033-7.012 (d, J= 91.2 Hz, 2H, ArH); 2.210 (S, 3H, CH_3-C=O); 2.176 (S, 3H, CH_3Ar). ^{13}CNMR (DMSO/$CDCl_3$, 400 MHz) δ: 171.133, 170.122 (C=O); 141.859, 140.318, 129.497, 128.890, 126.864, 125.756, (aromatic carbons); 78.693, 78.367, 78.040, 39.282 (aliphatic carbons). Anal.calcd (%) for $C_{12}H_{15}NO_6S$ (301.33): C, 47.80; H, 4.98; N, 4.66; S, 10.63. Found: C, 47.76; H, 4.96; N4.70, S, 10.59.

2-{Acetyl[(4-methylphenyl)sulfonyl]amino}-3-hydroxybutanoic acid(2e)

Yield=2.30g (96.0%); mp.217-218 ˚C, IR (KBr) cm^{-1}: 3387 (O-H of COOH); 3093 (N-H); 2932 (C-H aliphatic); 1995 (C-H aromatic); 1708, 1691 (C=O); 1650, 1640 (C=C); 1381, 1294 (S=O); 1180 (SO_2-NH); 1131 (C-N); 746 (Ar-H); ^{I}HNMR (DMSO, 400MHz) δ: 7.772-7.755 (d, J= 6.8Hz, 2H, ArH); 7.564-7.548 (d, J= 6.4Hz, 2H, ArH); 4.099 (s, IH, OH); 3.892 (s, IH, COOH); 2.489 (s, 3H, CH_3-C=O); 0.957-0.944 (d, J= 5.2Hz,3H, CH_3-CH) ^{13}(NMR (DMSO, 400MHz) δ:171.222; 170.233 (C=O); 140.918, 132.895, 129.444, 126.970, 124,688, 120.233 (aromatic carbons); 67.675. 61.612, 40.061, 39.848, 39.272 (aliphatic carbons). Anal.calcd (%)for $C_{13}H_{17}NO_6S$ (315.35): C, 49.48; H, 5.39; N, 4.45; S, 10.15. Found: C, 49.52; H, 5.43; N, 4.48; S, 10.21.

2-{Acetyl[(4-methylphenyl)sulfonyl]amino}-4-methylpentanoic acid(2f)

Yield=2.29g (95.8%); mp.217-218 ˚C, IR (KBr) cm^{-1}: 3273 (OH of COOH); 3073 (N-H); 2970 (C-H aliphatic); 1992 (C-Haromatic); 1871, 1695 (C=O); 1670, 1641 (C=C); 1336, 1135 (S=O); 1121 (SO_2-NH); 1088 (C-N); 741 (Ar-H). ^{1}HNMR (C_6D_6, 400MHz) δ: 7.411 (m, 2H, ArH); 7.043 (m, 2H, ArH); 3.261 (s,IH, CH-CO_2H); 2.147 (s,3H, CH_3-C=O); 2.860 (s,3H,CH_3-Ar); 1.747 (s, 2H, 2CH); 1.277 (s, 6H, 2CH_3-CH). ^{13}C-NMR (C_6D_6,400MHz) δ:171.244, 170.142(C=O); 138.557, 133.883, 133.688 133.056, 132.821, 132.578 (aromatic carbons); 78.349, 72.463, 69.789, 65.892, 56.453, 45.152 (aliphatic carbons). Anal.calcd (%) for $C_{15}H_{21}NO_5S$ (327.41): C, 54.99, H, 6.42, N, 4.29, S, 9.78. Found: C, 54.97, H, 6.46, N, 4.31, S, 9.83.

BIOLOGICAL STUDIES

ANTIMICROBIAL STUDIES

Preparation of media

Nutrient agar: 27.5g of nutrient agar powder was dissolved in 1000ml of distilled water and was allowed to soak for 15mins. The agar suspension was melted by boiling in a water bath. 20ml of the molten nutrient agar was dispensed into a bijou bottle, cocked, and sterilized in an autoclave at 121ºC for 15mins. The sterile molten nutrient agar was stored at 42ºc until the time of use.

Potato Dextrose Agar (PDA): 47g of PDA powder was dissolved in 1000ml of distilled water and was allowed soaking for 15mins. The agar suspension was melted by boiling in a water bath. 20ml of the molten PDA was dispensed into a bijou bottle, cocked, and sterilized in an autoclave at 121ºC for 15mins. The sterile molten potato dextrose agar was stored at 42ºc until use.

The used test microorganisms: The antimicrobial screening was done according to the agar dilution method [12]. The test microorganisms used (*Staphylococcus aureus*, *Escherichia coli*, *Bacillus subtilis*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Candida albicans*, and *Aspergillusniger*) were clinical isolates obtained from the department of pharmaceutical microbiology and biotechnology laboratory, University of Nigeria, Nsukka.

Standardization of the test organism suspension: The organisms were standardized using 0.5 McFarland turbid equivalents.

Preparation of different concentrations of the synthesized compounds used: 5mg/ml of the stock concentration of the compounds was prepared by dissolving 10g of the extract in 2ml of 50% DMSO. 1.0mg/ml, 0.9mg/ml, 0.8mg/ml, 0.7mg/ml, 0.6mg/ml, 0.5mg/ml, 0.4mg/ml, 0.3mg/ml, 0.2mg/ml, and 0.1mg/ml, concentrations were obtained using $C_1V_1=C_2V_2$ formula.

Where C_1 (initial concentration) =5mg/ml
V_1 (Initial volume) = X
C_2 (final concentration) = 1.0mg/ml
V_2 (final volume) = 20ml,

Control test (standard): The standard antibiotic used was Ofloxacin and Fluconazole.

Experimental: 4.0ml of sample suspension of stock concentration 50mg/ml was transferred to the sterile Petri dish, 16.0ml of double strength sterile molten agar was transferred to the same plate to mix uniformly and thus, 1mg/ml concentration was obtained. The other concentrations 0.9mg/ml, 0.8mg/ml, 0.7mg/ml, 0.6mg/ml, 0.5mg/ml, 0.4mg/ml, 0.3mg/ml, 0.2mg/ml, 0.1mg/ml, were obtained using the same $C_1V_1=C_2V_2$ formula. The molten agar plates with different concentrations of the sample were allowed to gel. The plates were divided into seven equal parts with a permanent marker. The test microorganisms were streaked on the segments and labeled. The culture plates were incubated in an inverted position at 37 ^oC for 24 hours and at 25 ^oC for 48 hours. After the incubation period , the plates were observed for sensitivity and resistivity of the organisms to the agents and the observations were recorded. The plates were further incubated for another 24 hours at 37 ^oC, and 48 hours at 25 ^oC to determine whether the activity was bacteriostatic or bactericidal. The observations were also recorded.

ANTIOXIDANT STUDIES

Antioxidant activity by DPPH method

The antioxidant activity was determined according to Blois Method [13]. The antioxidant behavior of the synthesized compounds was measured *in vitro* by the inhibition of generated stable 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical. The DPPH solution was prepared by dissolving 1.9 mg of DPPH in 100 ml of methanol. Three different concentrations (50, 100, and 200 µg/ml) of the DPPH solution were prepared. 2 mg of each synthesized compound was weighed out and dissolved in 10 ml of an appropriate solvent. The stock solution (200 µg/ml) was diluted further to get 100 and 50 µg/ml of each sample. The standard solution of ascorbic acid was prepared in a similar manner. 1 ml of DPPH solution was added to 2 ml of each sample solution and ascorbic acid. The reaction mixture was vortexed thoroughly and left in the dark at room temperature for 30 minutes. The absorbance of the mixture was measured (in triplicate) spectrophotometrically at 517 nm against the corresponding blank solution. The percentage of DPPH radical scavenging activity was calculated by using the following formula:

DPPH radical scavenging activity (%) =

$$\frac{\mathbf{Abs_{control} - Abs_{sample}}}{\mathbf{Abs_{control}}} \times \mathbf{100} \qquad \text{Eq 2}$$

where $Abs_{control}$ was the absorbance of DPPH radical and n-hexane/methanol, Abs_{sample} was the absorbance of DPPH radical and sample/standard.

In silico methodology

Physicochemical properties

The physicochemical properties of the synthesized compounds were generated *in silico*. They included molecular weight (MW), number of hydrogen bond acceptor (HBA), number of hydrogen bond donor (HBD), number of rotatable bond (NoRB), octanol/water partition coefficient logP(o/w), aqueous solubility (SlogP), and total polar surface area (TPSA). These parameters were computed by descriptors calculator in Molecular Operating Environment (MOE, 2018). The drug-likeness was evaluated using Lipinski's rule of five.

Molecular docking

Five different disease conditions were studied, namely: trypanosomiasis, malaria, bacterial and fungal infections, and oxidative stress. A drug target was chosen for each of the disease conditions for molecular docking studies. The drug targets for anti-trypanosomiasis: *T.* bruce farnesyl diphosphate synthase complexed with minodronate (PDB code: **2EWG**); antimalarial: plasmepsin II, a hemoglobin-degrading enzyme from *Plasmodium falciparum*, in complex with pepstatin A (PDB code: 1SME); antibacterial: *E. coli* DNA gyrase in complex with 1-ethyl-3-[8-methyl-5-(2-methyl-pyridin-4-yl)-isoquinolin-3yl]urea (PDB code: **5MMN**); antifungal: urate oxidase from *Aspergillus flavus* complexed with uracil (PDB code: **1WS3**), and antioxidant: human peroxiredoxin 5 (PDB code: **1HD2).**

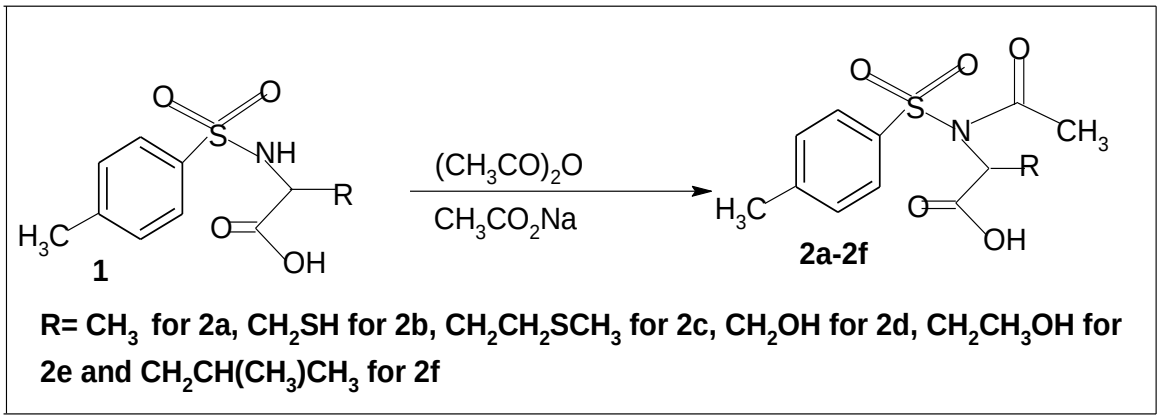
The 3-dimensional structures of these drug targets were downloaded from the Protein Data Bank (PDB), (http://www.pdb.org) database. The drug targets were loaded in Molecular Operating Environment (MOE) and prepared using the QickPrep in MOE. The MMFF94 force field was used for energy minimization of the ligand molecules. The prepared compounds were then subjected

to interact with each of the receptors through molecular docking. The protocol facilitates flexible compound docking for various compound conformers within the rigid receptor. The best conformation for each compound was chosen and the interaction was visualized in Discovery studio.

RESULTS AND DISCUSSION

SYNTHESIS

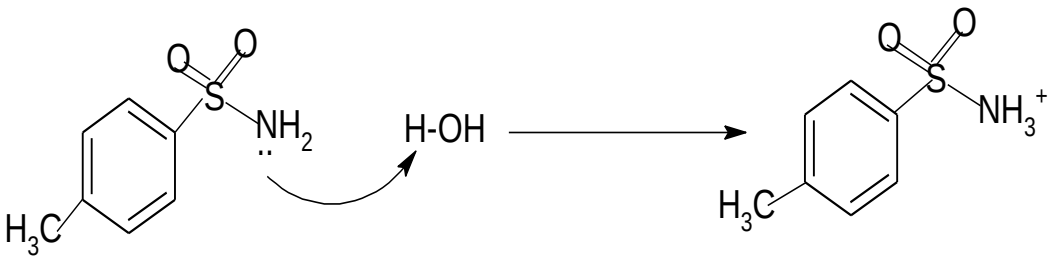
The already existing 4-methylphenyl sulphonamoyl carboxylic acids were subjected to alkanoylation reaction using acetic anhydride in the presence of sodium acetate. This reaction step was carried out for the purpose of protecting the amine functional group and incorporation of the biologically active acyl group into 4-methylphenyl sulphonamoyl carboxylic acids in order to achieve enhanced biological activities and improved drug potency.



Scheme 1: synthesis of alkanoylated 4-methylphenyl sulphonamoyl carboxylic acids

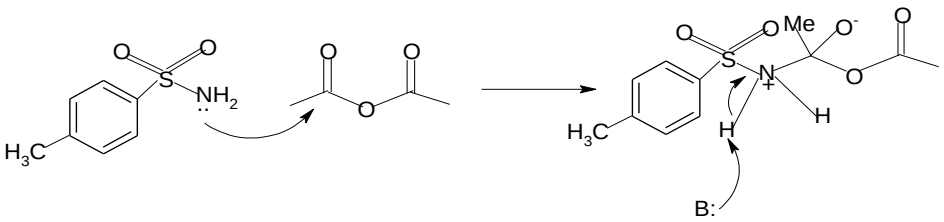
REACTION MECHANISM

In compliance with Lumiere-Barbier method, the amino group can be alkanoylated in aqueous solutions [14]. In the first step, the compound dissolves in water with one equivalent of hydrochloric acid. The amino group of 4-methylphenyl sulphonamide reacts in aqueous solution as follows:

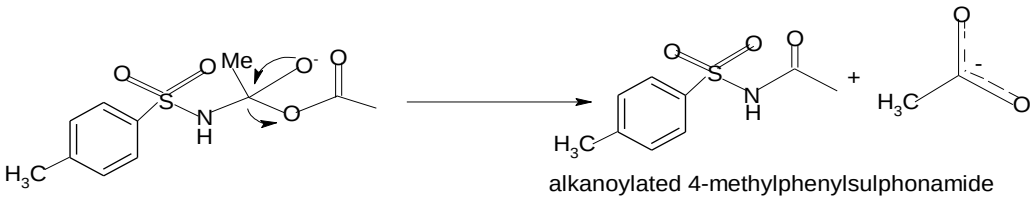


Scheme 2: Reaction of the amino group in aqueous solution

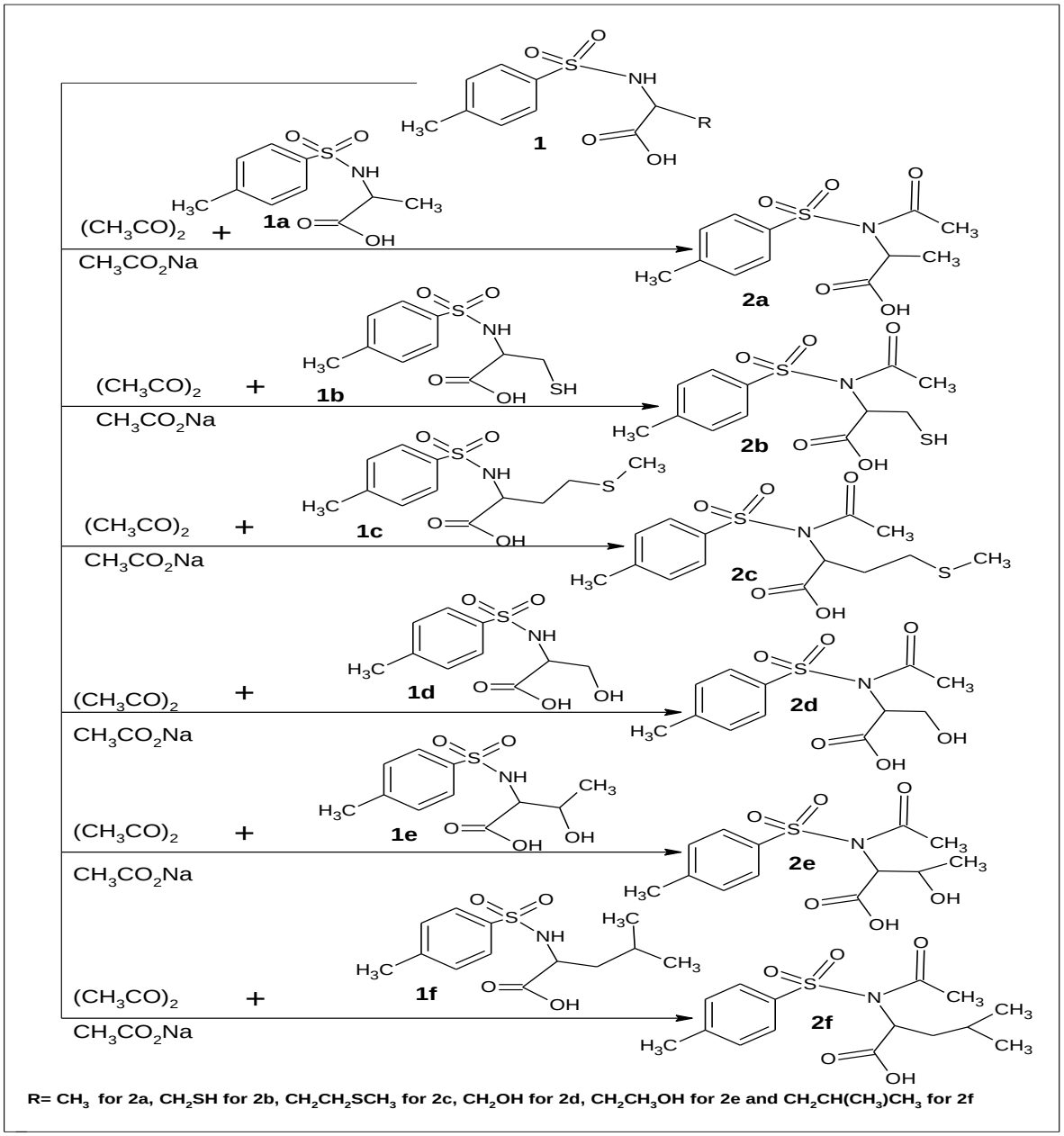
Then, 1.2 equivalent of acetic anhydride is added after which 1.2 equivalent of aqueous sodium acetate solution is added to the solution. The amino group of the 4-methylphenyl sulphonamide attacks acetic anhydride followed by deprotonation of the ammonium ion as follows (**Scheme 3**):



Then acetate acts as a living group of the substrate:



Scheme 3: reaction mechanism for the synthesis of alkanoylated 4-methylphenyl sulphonamoyl carboxylic acids



Scheme 4: a synthetic pathway for alkanoylated 4-methylphenyl sulphonamoyl carboxylic acid.

ANTIMICROBIAL ACTIVITY RESULTS

Table 1: Antimicrobial activities of compounds (2a-2f)

minimum inhibitory concentration (MIC)(mg/ml)							
COMPOUNDS	***E.coli***	***S.typhi***	***S. aureus***	***B. sub***	***Ps. aerug***	***C. albicans***	***A. niger***
2a	0.8	0.7	+	0.8	+	0.8	+
2b	0.6	0.9	0.7	0.4	0.9	0.6	0.9
2c	0.7	0.7	0.9	0.5	0.7	+	+
2e	0.9	1.0	+	+	+	0.9	+
2f	0.9	0.7	+	0.8	+	0.7	+
Ofloxacin	0.005	0.010	0.010	0.020	0.020	+	+
Fluconazole	**+**	+	**+**	**+**	**+**	**0.020**	**0.005**

Key: + = no inhibition, Ofloxacine and Fluconazole are the antibacterial and antifungal standard drugs

The antimicrobial studies (**Table 1**) revealed that the title compounds had good antimicrobial (antibacterial and antifungal) activities when compared to the commercial standard drugs. It was also observed that compounds **2b** and **2c** possess the best antibacterial activities while compound **2b** had the best antifungal activity. This implies that these alkanoylated 4-methylphenyl sulphonamoyl carboxylic acids can serve as good antibacterial and antifungal agents.

ANTIOXIDANT STUDIES

Table 2: Antioxidant activities results

	200 µg/ml		**100 µg/ml**		**50 µg/ml**	
Sample	**% inhibition**	**Std**	**% inhibition**	**Std**	**% inhibition**	**Std**
Ascorbic acid	96.83	0.001	97.68	0.001	97.31	0.001
290a	93.41	0.000	87.67	0.001	84.92	0.001
290b	75.03	0.002	71.79	0.002	77.05	0.001
290e	79.12	0.001	82.17	0.001	79.55	0.001
290f	79.67	0.000	ND	ND	75.34	0.001

The *in vitro* antioxidant studies (**Table 2**) showed that some of the tested compounds had antioxidant activities. Compound **2a** showed impressive antioxidant activities. This implies that compound **2a** was the most potent antioxidant agent and therefore could be subjected to further derivatization to improve the drug-likeness.

Drug-likeness Studies

Table 3: Physicochemical properties

comp	HBA	HBD	NoRB	logP(o/w)	SlogP	TPSA	Weight	lip_violation
290a	5	2	5	1.22	1.01	91.75	285.32	0
290b	5	2	5	1.40	0.87	130.55	303.36	0
290c	5	2	8	1.87	1.74	91.75	345.44	0
290e	6	3	6	0.64	0.37	111.98	315.35	0
290f	5	2	7	2.63	2.03	91.75	327.40	0

The physicochemical properties to evaluate the drug-likeness of the synthesized compounds are shown in **Table 3**. Lipinski's rule of five (Ro5) is used for the assessment of the drug-likeness of a molecule. According to this rule, a molecule must have a molecular weight value of ≤ 500, hydrogen bond donor (HBD)≤ 10 and partition coefficient (Log P) value ≤ 5. Violation of more than one parameter may pose a challenge to the bioavailability of the molecule in case of the oral formulation. From the results in **Table 3**, the synthesized

compounds are in agreement with (Ro5). The TPSA, a reflection of the ligand hydrophilicity, is vital in protein-ligand interaction. NoRB ≤ 10 and TPSA ≤ 140 Å^2 would have a high probability of good oral bioavailability in rats [15]. Also, a compound with TPSA of ≤ 90 Å^2 can cross the BBB and penetrate the CNS [16]. Therefore, they can be very valuable in treating CNS related diseases such as cerebral malaria, Alzheimer's diseases.

Results of Molecular Docking Studies

Table 4: In silicoAntitrypanosomal, Antimalarial, Antibacterial, Antifungal, and Antioxidant Activities.

Compound	Trypanosomiasis	Malaria	Antibacterial	Antifungal	Antioxidant
	2EWG	1SME	5MMN	1WS3	1HD2
2a	-13.35	-10.83	-9.63	-11.79	-12.66
2b	-13.68	-11.48	-10.17	-10.62	-11.81
2c	-13.68	-11.46	-9.46	-9.49	-11.33
2e	-14.54	-11.28	-10.54	-10.48	-12.28
2f	-13.04	-11.78	-11.22	-9.49	-11.45
Standard drug	-19.36	-10.08	-10.89	-10.38	-9.34

Standard drugs for **2EWG - Melarsoprol; 1SME - Chloroquine;** 5MMN - Penicillin; 1WS3 - Ketoconazole; **1HD2 - α**-Tocopherol

Molecular docking

The calculated free binding energy after molecular docking is given in **Table 4**. Our compounds showed strong binding affinities with all the receptors used in this study. Among all the compounds tested on 2EWG, **2a** and **2f** gave the lowest binding energy (highest binding affinity) of -13.35 and 13.04 kcal/mol, respectively. However, the standard drug for the treatment of trypanosomiasis (melarsoprol) showed the highest binding affinity with 2EWG (-19.36 kcal/mol). Likewise, compound **2f** showed the highest binding affinity (-11.78 kcal/mol) with the *Plasmodium falciparum* pepstatin A receptor (1SME) when compared to the standard drug (chloroquine) for malaria treatment, whose binding affinity is -10.08 kcal/mol. For the DNA gyrase receptor, compound **2f** had a more binding affinity with it (-11.93 kcal/mol) than with penicillin (-10.89 kcal/mol). The receptor for antifungal study, 1WS3, had the highest binding affinity with compound **2a** (-11.79 kcal/mol), which was better than that of ketoconazole (-10.38 kcal/mol). Finally, compounds **2a, 2b, 2c, 2e,** and **2f** outperformed α-tocopherol in their binding affinities with **1HD2** standard drug (-9.34 kcal/mol).

***In silico* antibacterial activities:** The *in silico* antibacterial studies (**Table 4**) revealed that compound **2a** with binding energy of -11.22 Kcal/mol possessed more excellent antibacterial activities than the commercial standard drug with a binding energy of -10.89 Kcal/mole. This implies that compound **2a** can serve as better antibacterial agent than standard commercial drugs and therefore should be considered accordingly.

***In silico* antifungal activities:** The *in silico* antifungal studies (**Table 4**) revealed that compounds **2a, 2b,** and **2e,** with binding energies -11.79, 10.62 and 10.48Kcal/mol respectively possess more excellent antifungal activities that the commercial standard drug with binding energy -10.38 Kcal/mole. This implies that compound **2a, 2b,** and **2e** can serve as better antifungal agents than standard commercial drugs and therefore should be considered accordingly.

***In silico* Antioxidant Activities:** The *in silico* antioxidant studies (**Table 4**) revealed that only compounds **2a, 2b, 2c, 2e** and **2f** (-12.66, 12.81, 11.33, 12.28, and 11.45 Kcal/mol) had comparable binding energy with α-tocopherol (-9.34 Kcal/mol). Compounds **2a, 2b, 2c, 2e,** and **2f** are more promising than the standard drug.

***In Silico* antitrypanosomal activities:** The *in silico* antitrypanosomal studies (**Table 4**) revealed that all the compounds possessed antitrypanosomal activities but none of the tested compounds were as effective as the standard drug.

***In Silico* antimalarial activities:** The *in silico* antimalarial studies (**Table 4**) revealed that all the compounds **2a, 2b, 2c, 2e,** and **2f** with binding energies of -10.83, -11.48, -11.46, and -11.28, -11.78 kcal/mol, respectively against *Plasmodium Falciparuim* as an excellent inhibitor of dihydrofolate reductase. These alkanoylated4-methylphenyl sulphonamoyl carboxylic acids when compared to the most potent standard antimalaria agent, Chloroquine (-10.08 Kcal/mol), showed better binding energies with compound **2f** being the most excellent compound and therefore can be used as antimalarial agents.

CONCLUSION

In conclusion, convenient synthesis of alkanoylated 4-methylphenyl sulphonamoyl carboxylic acids (**2a-2f**) was successful. The assigned structures were in perfect

agreement with the spectral data. The antimicrobial, antioxidant, and *in silico* studies carried unveiled excellent biological and pharmacological activities of the synthesized compounds. The results showed that compounds **2b and 2c** possessed the most excellent *in vitro* antibacterial and antifungal activities respectively compared to the standard drugs ofloxacin and fluconazole, therefore, these compounds are potent antibacterial and antifungal agents. Compound **2a** exhibited the most excellent antioxidant activity and therefore it can be considered as the most promising antioxidant agent. The molecular docking revealed that compounds **2a, 2b, 2c, 2e,** and **2f** exhibited excellent antibacterial, antifungal, antioxidant, antitrypanosomal, and antimalarial activities similar to their corresponding standard drugs. The synthesized alkanoylated 4-methylphenyl sulphonamoyl carboxylic acids were found to be potent antibacterial, antifungal, antioxidant, antitrypanosomal and antimalarial agents.

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Conflict of interest

The authors declare that they have no competing interest.

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