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# 6-Adenine and 2-Guanine Benzenesulphonamide: Synthesis and *In silico* Anti-cancer Properties.

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## Abstract

Sulphonamide preparation converts readily available aromatic amines into life-saving anticancer molecules. Many drugs available for the cure of cancer are no longer active due to drug resistance. Novel derivatives of adenine and guanine incorporating benzenesulphonamide have been synthesized, characterized and investigated for *in silico* anti-cancer properties. The reaction of adenine and guanine with benzenesulphonyl chloride afforded N-(9H-purin-6-yl)benzenesulphonamide **(4)** and N-(6-dihydro-1H-purin-2-yl)benzenesulphonamide **(6)** respectively in 66% and 42% yields. The compounds were characterized using Ultra-violet visible and FTIR techniques. The compounds were assayed for their in silico anticancer activities using their binding energies with hormone-bound human progesterone receptor ligand binding domain **(1A28)**, estrogen receptor alpha ligand-binding domain **(1A52)**, epidermal growth factor tyrosine kinase **(1M17)** and human androgen receptor **(1E3G)**. Diclofenac was used as the standard drug.The ligand s-protein complex showed binding energies ranges between -8.0 to -5.9. The highest and lowest binding energies were -8.0 and -5.0 recorded in the (1A28-4) complex and (diclofenac-1A52) complex respectively. All the compounds showed good binding energies comparable to the standard drug diclofenac.

**Keywords:** adenine, guanine, sulphonamides, *in silico*, diclofenac, anticancer, complex.

## Introduction

Drug molecules in the sulphonamide class have been a cornerstone in the field of antimicrobial therapy since their discovery in the early 20th century (Abdulrazzaq and Sultan, 2023). The general formula for sulphonamides is $RSO_2NH_2$, where R represents a substituent group (Yousif, 2025). Sulphonamides have the general structure shown in **Fig 1**, in which R may be alkyl, aryl or hetero aryl groups and $R_1/R_2$ may also be hydrogen, alkyl, aryl or hetero aryl groups (Pradk, 2013, Bahrami *et al*., 2010).

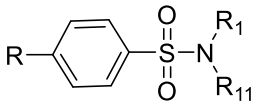


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**Figure 1:** Structure of sulphonamide.

*In silico* studies are done using computer simulations rather than traditional laboratory experiments. Over the years, there have been many reports on the applications of *in silico* studies in drug discovery (Agu *et al*., 2023; Adesina *et al*., 2025). Despite the extensive research and widespread usage of sulphonamide compounds, the emergence of bacterial resistance and the occurrence of adverse effects have underscored the necessity for developing novel sulphonamide derivatives with enhanced properties (Angeles-Hernandez *et al*., 2025). Initially, sulphonamides were thought to be great, but they faced a decline in production enthusiasm with the advent of more potent antibiotics like penicillin (Azabo *et al*., 2022; Aziz *et al*., 2022; Badawy *et al*., 2023). But even with newer antibiotics around, sulphonamides are still very useful (Hassanein, 2019). Sulphamethoxazole is a known sulphonamide which is most often used as part of a synergistic combination with trimethoprim in a 5:1 ratio in co-trimoxazole (Baglini *et al*., 2021; Das *et al*., 2015). This pairing is particularly effective against a wide range of infections. Sulfamethoxazole works by inhibiting folic acid synthesis through the same mechanism as other sulphonamides, thereby preventing bacterial growth. It also inhibits the enzyme activity of dihydropteroate synthase (Ikpa *et al*., 2020; Mohammed *et al*., 2020; Hamad *et al*., 2021). Sulphamethoxazole has shown antitumor, anti-viral, anti-fungal, anti-carbonic anhydrase, diuretic, antithyroid, hypoglycemic and protease inhibitor activity (Das *et al*., 2015; Mussi *et al*., 2021; Onoabedje *et al*., 2021; Oudaha *et al*., 2022). Sulphisoxazole is a sulphonamide antibiotic characterized by its oxazole substituent, and it is primarily used to treat a variety of bacterial infections (Iturrieta-Gonzalez *et al*., 2024; Kimera *et al*., 2020; Liu *et al*., Mittapalli et al., 2024). As a member of the sulphonamide class, sulphisoxazole operates by inhibiting the enzyme dihydropteroate synthase, which is crucial for bacterial folate synthesis (Onoabedje *et al*., 2021; Ghosh *et al*., 2023; Gentle *et al*., 2020; Feng *et al*., 2016; Eze *et al*., 2020). This inhibition occurs through competitive antagonism with para-aminobenzoic acid, a substrate necessary for the production of folic acid in bacteria (Davies *et al*., 2020; El-Gaby *et al*., 2020). By disrupting folate synthesis, sulphisoxazole effectively prevents bacterial growth and replication, making it useful against a broad spectrum of gram-positive and gram-negative organisms (Peerzada *et al*., 2018). Sulphanilamide is a sulphonamide antibacterial drug that plays a significant role in the treatment of bacterial infections. Sulphanilamide was one of the first sulphonamide antibiotics discovered in 1906 (Ovung and Bhattacharyya, 2021). Sulphadiazine is a sulphonamide antibiotic widely used for treating various bacterial infections (Paul *et al*., 2023; Rejinthala et al., 2024; Said *et al*., 2022). It exhibits bacteriostatic action, meaning it stops bacteria from multiplying but does not kill them outright (Supuran, 2021). This antibiotic is effective against a range of infections, including urinary tract infections, trachoma, chancroid, and certain forms of bacterial meningitis. It is also effective against certain forms of bacterial meningitis and is indicated for prophylaxis against rheumatic fever (Wani *et al*., 2023). The synthesis of sulphonamides involves diverse methodologies, each tailored to specific substrates and functional group tolerances (Yousif *et al*., 2022; Xing, 2025). To date, such an approach is a general method for the preparation of sulphonamide in pharmaceutical industry which still needs further improvement (Feng *et al*., 2016).

**Materials and methods.**

The chemicals and reagents utilized in this synthesis were purchased from Sigma-Aldrich. Thin layer chromatography was carried out using silica gel precoated plates. The plates were visualized using certified A.C.S iodine crystals extra pure. FTIR spectroscopy of the compounds were run on IR M530 Buck Scientific infrared spectrophotometer using KBr pellets. The absorptions values were given in wavenumbers ($cm^{-1}$). The Ultraviolet and visible spectra were obtained on a UV1 ultraviolet spectrophotometer (manufactured by UNICAM, Serial number 061408) using matched 1cm quartz cells. Ethanol was used as solvent. Absorption maximum values were provided in nanometers (nm). Melting points were determined using Melting Point Apparatus NDJ-5S Searchtech instrument. The *in silico* pharmacological evaluation for anti-inflammatory, antibacterial and anticancer were carried out at the Department of Chemistry, Hallmark University, Ijebu Itele, Ogun State, Nigeria. The structure of the two ligands that were synthesized in the laboratory as well as the standard ligand were downloaded from PubChem website (http://pubchem.ncbi. nlm.nih.gov) and were converted from a Structured Data File (SDF) to Protein Data Bank (PDB) file format (Adedotun *et al*., 2025). The ligands and standard were N-(9H-purin-6-yl)benzenesulphonamide, N-(6-oxo-6,9-dihydro-1H-purin-2-yl)benzenesulphona- mide and Diclofenac (standard). The crystal structure of target molecules were downloaded from protein database website (https://www. rcsb.org) and were saved as Protein Data Bank Format (Adedotun *et al*., 2025).

**Synthesis of N-(9H-purin-6-yl)benzenesulphonamide.**

Sodiumtrioxocarbonate (IV) ($Na_2CO_3$) (1.17 g, 11.04 mmole) was added to a solution of adenine (1.49 g, 11.04 mmole) in distilled water (30 $mL$) and dimethylformamide (20 $mL$). This was followed by continuous stirring using a magnetic stirrer until all the solutes dissolved. The solution was cooled to 2$^o$C and benzenesulphonylchloride (1.95 g, 11.04 mmole) was added in four portions over a period of 1hour. The slurry was then stirred at room temperature for 5 hours. The progress of the reaction was monitored by the use of precoated thin layer plates. The reaction mixture was diluted with water/ n-hexane and partitioned in a separating funnel. The organic and aqueous layers were separated. The organic layer was washed with brine, dried over anhydrous sodium sulphate and concentrated. Recrystallization with dichloromethane gave the pure product in (66 %) yield. $mp$ 198-200$^o$C . IR ($cm^{-1}$ KBr): Yield (3.03g, 1.99%) Colour: White crystals. Melting point: 198°-200° C. UV - Visible spectra: Xmax (EtOH): 410nm (Log Ɛ = 1.47). FTIR of N-(9H-purin-6-yl)benzenesulphonamide. (KBr, $cm^{-1}$) 1390 due to $SO_2$ , 3325 due to N-H stretching, 1623 due to C=C and C=N, 1475 due to C-N stretching, 2960 due to C-H stretching and 830 due to C-H bending.

**Synthesis of N-(6-oxo-6,9-dihydro-1H-purin-2-yl)benzenesulphonamide.**

The synthesis was carried out using the method in 1.1. $Melting\ point$ 300-301$^o$C. Yield: 41.5%, colour: White crystals, UV-Visible Spectra: Wavelength Max (EtOH): 410nm (Log Ɛ =1.47). FTIR ($cm^{-1}$ KBr): 3280 due to N-H stretching, 1710 due to C=O stretching, 1600 due to C=N/C=C aromatic stretching (aromatic and heteroaromatic), 1150 due to S=O (sulphonamide) stretch, 3050 due to aromatic C-H stretching and bending, 1020 due to C−N stretch.

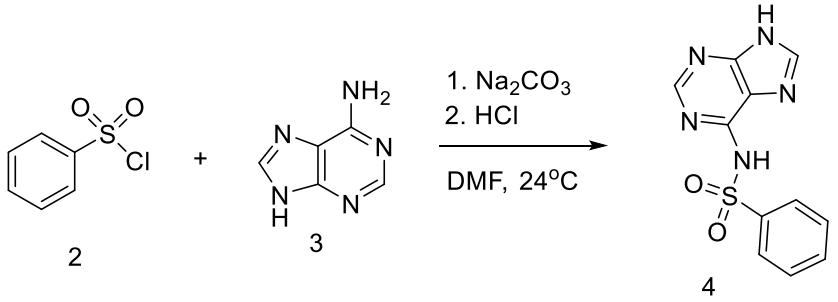
**Molecular docking.**

Computational experiment was conducted as previously described by Adedotun *etal*.,

2025 with the aid of Auto Dock (v4.2) that has been integrated into the PyRx software (v0.8 for windows). An exhaustiveness of 8 was chosen for each protein and this was utilized for docking and the generation of 9 docked binding poses. The binding energies of the synthesized compounds and the standard compound were compared to determine the most active compounds. The docking results (ligand-receptor interactions) were visualized using Biovia Discovery Studio software (v21.1.0.20289), and finally represented in 2D and 3D models (Adedotun *et al*., 2025).

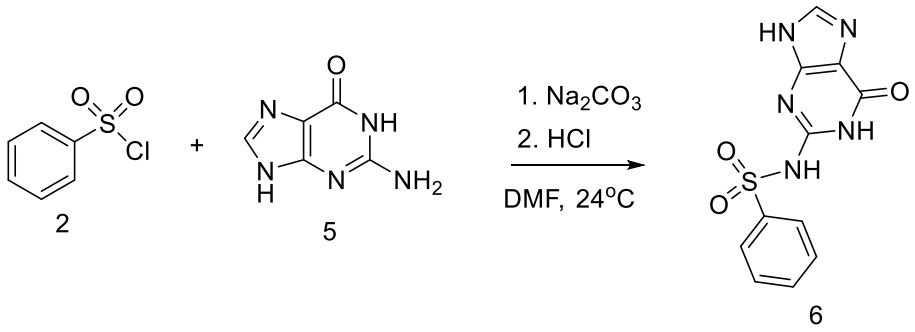
**Results and Discussion.**

**Synthesis and characterization and spectral analysis**




**Scheme 1:** Synthesis of N-(9H-purin-6-yl)benzenesulphonamide.

The structures of the compounds were characterized by IR spectra. The peaks at 3100 $cm^{-1}$ were due to asymmetric $SO_2$ stretching which confirms the presence of the sulphonamide group. The bands at 1604 $cm^{-1}$ and 1400 $cm^{-1}$ confirmed the presence of carbon to carbon double bond of aromatic compounds. The band at 759 $cm^{-1}$ is due to monosubstituted benzene ring.




**Scheme 2:** Synthesis of N-(6-oxo-6,9-dihydro-1H-purin-2-yl)benzenesulphonamide.

***In silico* anticancer and anti-inflammatory properties.**

Anticancer property against the human progesterone receptor **(1A28)** is seen in **Fig 2.** The amino acid residues of **1A28** bind to the compound **4** by four types of intermolecular bonding forces. There is pi-pi T-shaped interaction, Van der Waals forces, conventional hydrogen bonding and pi-alkyl interactions. The hydrogens of the amino groups of **3** binds to the **GLU A:695** using mainly conventional hydrogen bonding at intermolecular distance of 6.03 angstrom. The sulphonamide oxygens bonds to the PRO A:696 using conventional

hydrogen bonding at intermolecular distance of 4.46 angstrom. There is also an intermolecular interaction between the aromatic sextet and ARG A:766 and TRP A:765 by pi-alkyl and pi-pi T-shaped interaction respectively. The binding energy values shown in **Table 1** reveals that the binding energy of **1** is lower than that of diclofenac for the proteins **1A28** and **1A52** respectively which means that they are more effective than diclofenac. For 1A28, diclofenac has a binding energy of -6.6 and 1 has a binding energy of -8.0. For 1A52, diclofenac has a binding energy of -5.9 and 1 has a binding energy of -6.1. The 2D binding interaction of diclofenac is displayed in **Fig 2**.

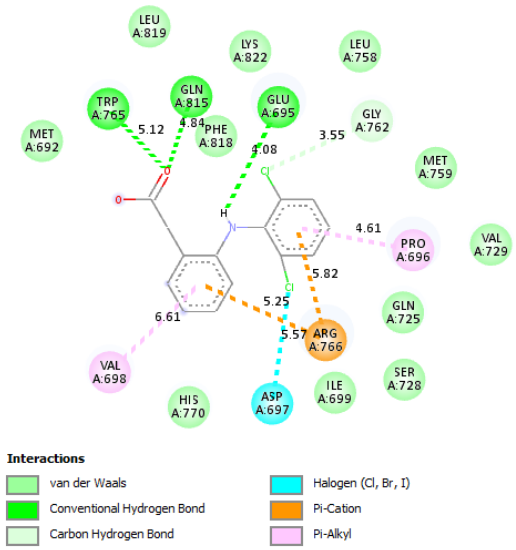



**Fig 2:** The 2D binding interaction of diclofenac with **1A28**.

The PDB entry **(1A28)** corresponds to the ligand binding domain of the human progesterone receptor in its hormone bound state contains the amino acids glycine, alanine, leucine, isoleucine, methionine, proline but the major binding interactions in the complex (**4-1A28**) are due to conventional hydrogen bonding and Van der Waals forces between the oxygen of the carboxylic acid and the amino acid proline, Pi-pi T-shaped interaction between the benzene ring of (4) and tryptophan and pi-alkyl interaction between the aromatic sextet of the benzene ring and the amino acid arginine of **(1A28) Figure 2**. Compound **4** showed a better binding energy for IE3G and 1M17 than compound 6 and diclofenac **Table 1**.

Table 1: Binding energies of **4**, **6** and diclofenac with **1A28**, **1A52**, **1M17** AND **1E3G.**

| **Compound** | **PDB-1A28** | **PDB-1A52** | **PDB-1M17** | **PDB-1E3G** |
|---|---|---|---|---|
| | **ΔG (kcal/mol)** | **ΔG (kcal/mol)** | **ΔG (kcal/mol)** | **ΔG (kcal/mol)** |
| 4 | -8.0 | -6.1 | -6.5 | -7.4 |
| 6 | -6.4 | -6.0 | -6.1 | -6.7 |
| Diclofenac | -6.6 | -5.9 | -6.3 | -6.5 |

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**Key: PDB-(1A28):** Hormone bound human progesterone receptor ligand-binding domain. **PDB-(1A52):** Estrogen receptor alpha ligand-binding domain PDB-**(1M17):** Epidermal growth factor receptor tyrosine kinase. PDB-(**1E3G)**: Human androgen receptor.

The interaction of diclofenac with the protein **1A28** is due to conventional hydrogen bonding between the centre NH of diclofenac and GLU A:695 and that between the carboxylic acid oxygen and GLU A:815and TRP A:765. There is also pi-carbon, halogen and pi-alkyl intermolecular . The binding energy of 1 and 2 are lower than that of diclofenac for the protein **1A52** which indicate better effectiveness **Fig 3**.

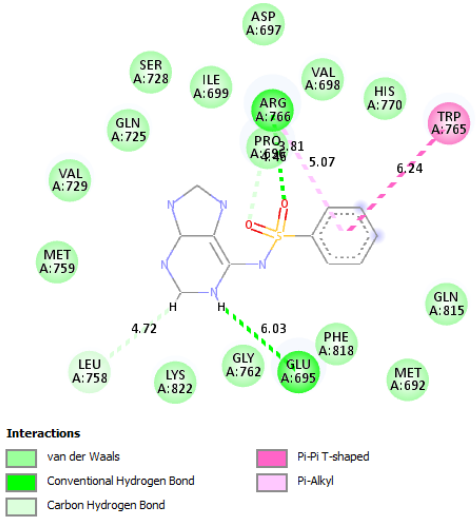


**Fig 3:** 2D view of the binding interaction of **4** with the amino acid residues of the protein **(1A28).**

The binding interactions of **6** and **(1E3G)** is displayed in **Fig 4** below. The major interaction is due to conventional hydrogen bonding.

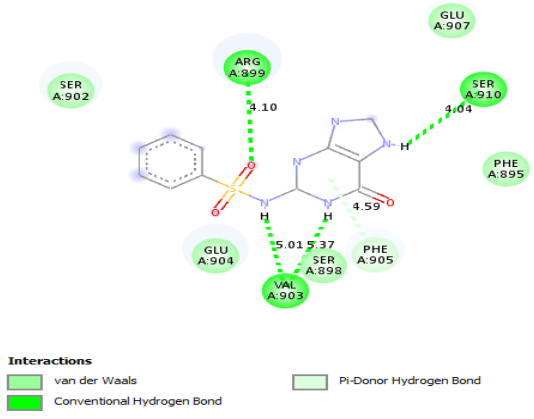


**Fig 4:** Binding interaction between the ligand 6 and **(1E3G).**

The interaction of diclofenac with (**1M17**) are mainly pi-alkyl interactions between the aromatic sextet of the benzene ring and the amino acid residues leucine and arginine which are responsible for the binding energy of -6.3 kcal/mol **Fig 5**.

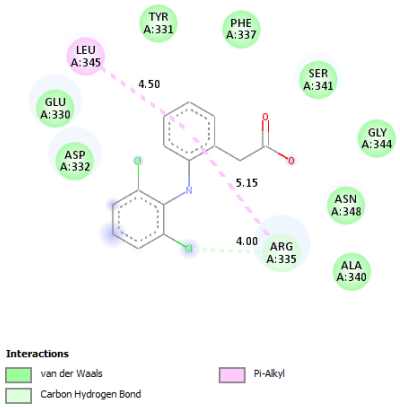


**Fig 5:** 2D view of the binding interaction of diclofenac and (**1M17**).

## Conclusion

Two new compounds were successfully synthesized and characterized. The UV-Vis and FTIR spectra of the studied compounds supported the assigned structures. The synthesized compounds exhibited low to high degree of anti-cancer activities. The compounds had good binding energies and displayed in-silico anticancer potentials. Compound 4 and 6 showed better binding energies than diclofenac for **1A28** revealing great prospect for further research in anticancer therapy.

## Conflict of Interest

The authors declare no conflict of interest.

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