Computational Insights into Indole Derivatives as Dual Antifungal and Antibacterial Agent: Docking Studies on Key Cellular Target

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Abstract: Due to their various biological activity, indole derivatives have garnered great attention as possible dual medicines against bacterial and fungal diseases. In this work, we used computational docking methods to explore the binding relationships between indole derivatives and important cellular targets in bacteria and fungus. Indoles are organic molecules found in nature that include an aromatic heterocyclic structure. They are extensively utilized in the production of medicinal products. Software tools like Auto Dock and Glide were used to perform molecular docking simulations using crystal structures of crucial enzymes and receptors involved in protein synthesis pathways, DNA replication, and cell wall construction. Antibacterial drugs are primarily developed through empirical screening programs, similar to the "golden age" of research. However, antimicrobial resistance is increasing, necessitating the design and synthesis of unique molecules to combat this global threat. Our findings demonstrate the potential mechanisms of action and promising binding affinities of indole derivatives against targets such as bacterial DNA gyrase and fungal lanosterol 14α-demethylase. The insights from computation

Key words: Auto Dock Vina, Antimicrobial efficacy, Computational methods, Docking simulations, Indole derivatives

INTRODUCTION

Bacterial infections that are resistant to antibiotics now pose a serious danger to global public health and cause significant financial and human suffering. This issue, which was formerly exclusive to nosocomial infections, is now more frequently observed in illnesses that are acquired in the community. Prominent instances comprise of multiply-resistant Gram-negative bacilli, like Escherichia coli, Acinetobacter baumanii, Pseudomonas aeruginosa, Enterobacter cloacae, and methicillin-resistant Staphylococcus aureus, as well as vancomycin-resistant enterococci and drug-resistant Streptococcus pneumoniae. These bacteria are frequently seen nosocomiants, however they are becoming lethal pathogens in a variety of illnesses due to the advent of multidrug resistance through numerous resistance pathways. We urgently need new antibiotics since multidrug resistance is on the rise.

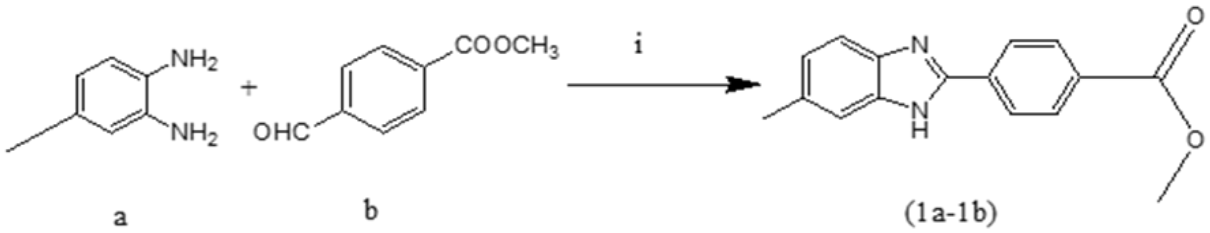
Because of their structural characteristics, heteroaromatic compounds—like indole molecules, where the benzene ring is linked to the second and third positions of the pyrrole ring—have been significant in the advancement of the chemical, pharmaceutical, medical, and agricultural industries. Most physiologically active substances, including medications and alkaloids, include indole and similar structures.

Indoles, aromatic organic compounds, are widely used in pharmaceutical synthesis due to their potent activity against fungi, viruses, Leishmania parasites, Gram-positive and Gram-negative bacteria, and mycobacterial species. This wide-spectrum activity makes them ideal candidates for developing novel anti-infectives to combat multidrug resistance, a pressing global issue.

Indoles, due to their unique structural motifs, are privileged scaffolds that can serve as ligands for various receptors. They have characteristics similar to peptides and can bind reversibly to enzymes, offering potential for discovering new drugs with different modes of action. However, further research and development are needed to develop alternative indole derivatives against common diseases.

RESULT

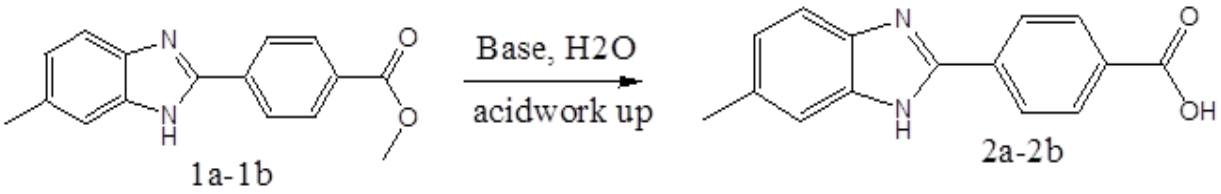
(I)Methyl 4-(5(6)-substituted-1H-benzimidazol-2-yl) benzoate derivatives (1a–1b) were synthesized by reacting methyl 4-formylbenzoate with various o-phenylenediamines in the presence of Na2S2O5. This two-step procedure involves the condensation of o-phenylenediamines with aromatic aldehydes, forming Schiff base intermediates that are then cyclodehydrogenated oxidatively to yield the benzimidazole products. Na2S2O5 was chosen as the oxidizing agent due to its reported effectiveness in achieving high yields in similar reactions according to previous studies.



(II) Esters are hydrolyzed either by aqueous base or acid to yield carboxylic acids plus alcohol. In base-catalyzed hydrolysis (saponification), a hydroxide ion attacks the ester carbonyl to produce a tetrahedral alkoxide intermediate. Elimination of the alkoxide ion generates the carboxylic acid, which is immediately deprotonated by the alkoxide ion. An acid work-up restores the carboxylic acid.

Acid-catalyzed hydrolysis can occur by more than one mechanism, depending on the structure of the ester. In a reverse Fisher esterification, the carbonyl group is protonated by the acid, which activates it for nucleophilic attack by water, yielding a tetrahedral intermediate. Transfer of a proton and elimination of alcohol yields the carboxylic acid.

- Reagents: Acid or Base
- Reactant: Ester
- Product: Alcohol, Carboxylic Acid
- Type of Reaction: Nucleophilic Acyl Substitution
- Bond Formation: RCO_2H



The reaction where an alcohol is converted to an alkyl chloride in the presence of thionyl chloride (PCL_5) at 70°C is known as the thionyl chloride reaction. This process involves the substitution of the hydroxyl group (-OH) of the alcohol with a chlorine atom (Cl), resulting in the formation of an alkyl chloride. The overall reaction can be summarized as follows:

Mechanism:
Using Phosphorus Pentachloride (PCl_5):
Procedure:

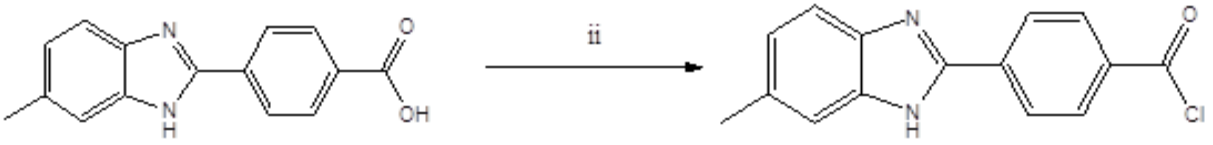
- Add phosphorus pentachloride directly to the aryl alcohol.
- The reaction often proceeds at room temperature but can be heated if necessary.
- Isolate the aryl chloride by standard workup procedures, such as extraction and distillation.

Safety Note: All these reactions should be carried out in a well-ventilated fume hood due to the release of harmful gases like HCl and SO_2. Proper personal protective equipment (PPE) should be worn, including gloves and eye protection.
These methods provide efficient pathways for the transformation of aryl alcohols into aryl chlorides.

Characteristics:
By-products: The reaction produces two gaseous by-products, SO2 and HCl, which can be removed from the reaction mixture easily.

- *Efficiency*: This method is highly efficient for converting primary and secondary alcohols to alkyl chlorides. However, it may be less effective for tertiary alcohols due to steric hindrance.

- *Mild Conditions*: The reaction generally proceeds under mild conditions without the need for extreme temperatures or pressures.



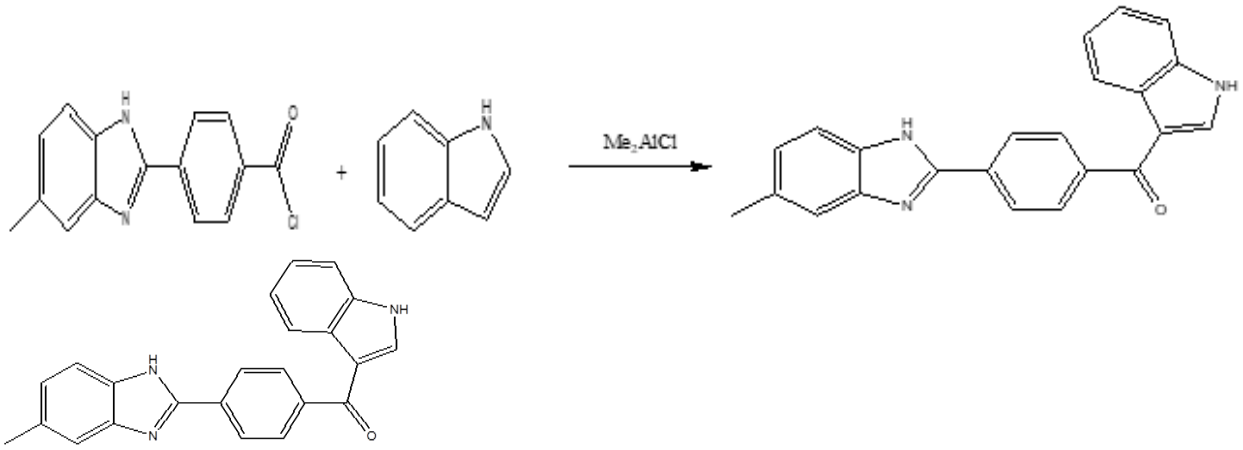
2a-2b 3a-3b

The reaction of removing a chlorine atom from an alkyl chloride in the presence of alcoholic potassium hydroxide, also known as the β-elimination reaction or dehydrohalogenation, typically results in the formation of an alkene. Here's a possible reaction:

R-Cl + KOH (alc.) -> R-H + KCl + H2O

Indoles are selectively acylated at the 3-position in high yields on treatment with a wide variety of acyl chlorides in CH2Cl2 in the presence of dimethylaluminum chloride.

The reaction proceeds under mild conditions and is applicable to indoles bearing various functional group without NH protection.



(1H-indol-3-yl)[4-(5-methyl-1H-1, 3-benzimidazol-2-yl)phenyl]methanone

Molecular Docking Studies:

The study investigates the mechanism by which new synthesized compounds, including chromenol, dihydroquinoline, and thiopyran, exhibit antimicrobial activity. In silico molecular docking studies were conducted on the bacterial target enzyme UDP-N-acetylmuramatel-alanine ligase (MurC) and Human lanosterol 14α-demethylase, to understand the binding affinity and intermolecular interactions of these compounds with the active sites of the target enzymes. Ampicillin was used as a standard drug for in silico screening of newly synthesized compounds, with the PyRx-virtual screening tool used for molecular docking. The flexible docking mode generated nine confirmations for each docked molecule.

MODE	AFFINITY (kcal/mol)	Distance from best mode rmsd l.b	Distance from best mode rmsd v.b
1	-9.2	0.000	0.000
2	-9.0	11.835	13.675
3	-8.9	22.438	26.598
4	-8.8	12.917	14.758
5	-8.7	2.465	9.168
6	-8.6	13.700	15.945
7	-8.5	5.279	8.073
8	-8.4	4.295	5.753
9	-8.2	3.307	9.480

Table. 1 Docking score for 6UEZ (PDB) enzyme

Fig 1. Human sterol 14a-demethylase (CYP51) in complex with the substrate lanosterol

Fig 2. Interaction of 4a-4b with Human sterol 14a-demethylase (CYP51) in complex with the substrate lanosterol

MODE	AFFINITY (kcal/mol)	Distance from best mode rmsd l.b	Distance from best mode rmsd v.b
1	-7.2	0.000	0.000
2	-7.2	23.051	25.239
3	-7.2	2.340	3.769
4	-7.0	23.152	25.932
5	-6.9	1.790	2.544
6	-6.7	23.011	25.597
7	-6.7	23.331	25.630
8	-6.6	22.965	24.933
9	-6.6	22.950	24.813

Table. 2 Docking score for 2F00 (PDB) enzyme

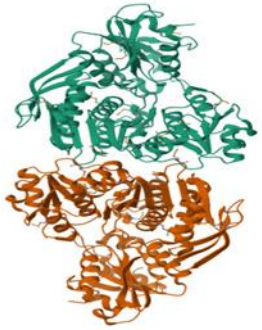


Fig 3. Structure of Escherichia coli UDP-N-acetylmuramoyl: L-alanine ligase (MurC).

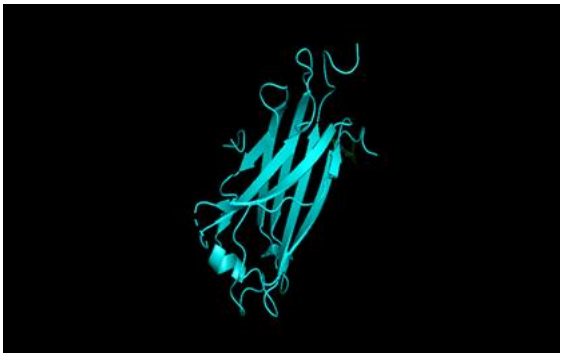
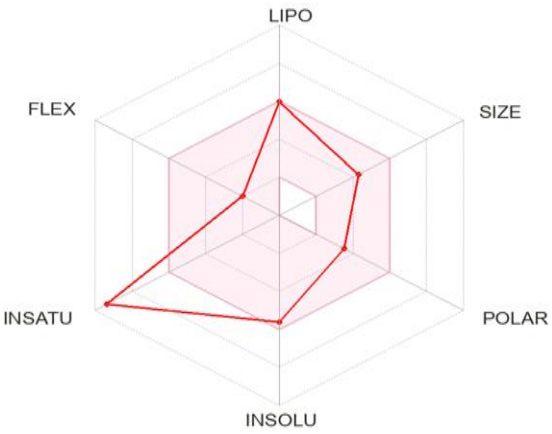


Fig 4. Interaction of 4a-4b with Structure of Escherichia coli UDP-N-acetylmuramoyl: L-alanine ligase (MurC).

Antifungal activity:

Antibacterial drugs today are primarily developed through empirical screening programs, similar to the "golden age" of antibacterial drug research. However, antimicrobial resistance is increasing worldwide, posing a threat to humans. The discovery and development of novel antimicrobial compounds are decreasing, necessitating the design and synthesis of unique molecules. Compound 4 did not show any antimicrobial activity against indicator microorganisms, while compound 5 showed intermediate antibacterial activity on three gram-positive bacteria and moderate antifungal activity against C. albicans ATCC 10231. After comparing MIC values with CLSI guidelines, compound 5 showed strong antibacterial activity against E. faecalis ATCC 29212, corresponding to a MIC of 4 µg/mL.

ADME :



Physicochemical Properties

Formula	C23H17N3O
Molecular weight	351.40 g/mol
Num. heavy atoms	27
Num. arom. heavy atoms	24
Fraction Csp3	0.04
Num. rotatable bonds	3
Num. H-bond acceptors	2
Num. H-bond donors	2
Molar Refractivity	108.23
TPSA	61.54 Å²
Lipophilicity	
Log $P_{o/w}$ (iLOGP)	2.47
Log $P_{o/w}$ (XLOGP3)	4.98
Log $P_{o/w}$ (WLOGP)	5.25
Log $P_{o/w}$ (MLOGP)	3.28
Log $P_{o/w}$ (SILICOS-IT)	6.00
Consensus Log $P_{o/w}$	4.40
Water Solubility	
Log *S* (ESOL)	-5.62
Solubility	8.51e-04 mg/ml ; 2.42e-06 mol/l
Class	Moderately soluble
Log *S* (Ali)	-6.01
Solubility	3.42e-04 mg/ml ; 9.74e-07 mol/l
Class	Poorly soluble
Log *S* (SILICOS-IT)	-9.13
Solubility	2.63e-07 mg/ml ; 7.48e-10 mol/l
Class	Poorly soluble
Pharmacokinetics	
GI absorption	High
BBB permeant	Yes
P-gp substrate	Yes
CYP1A2 inhibitor	Yes
CYP2C19 inhibitor	Yes
CYP2C9 inhibitor	No
CYP2D6 inhibitor	Yes
CYP3A4 inhibitor	Yes
Log K_p (skin permeation)	-4.91 cm/s
Druglikeness	
Lipinski	Yes; 0 violation
Ghose	Yes
Veber	Yes
Egan	Yes
Muegge	Yes
Bioavailability Score	0.55
Medicinal Chemistry	
PAINS	0 alert
Brenk	0 alert
Leadlikeness	No; 2 violations: MW>350, XLOGP3>3.5

CONCLUSION

A method for acylation of indoles at the 3-position with various acyl chlorides in the presence of Et2AlCl or Me2AlCl has been developed, which is applicable to indoles with various functional groups without NH protection. This method is simple, mild, and general, making it valuable for indole chemistry. A series of eleven novel compounds with heterocycles, such as chromenol, dihydroquinoline, and thiopyran moieties, were synthesized and characterized using elemental and spectral analyses. In silico docking studies were performed against target enzymes UDPN-acetylmuramatel-alanine ligase (MurC) and human lanosterol14α-demethylase to study binding affinities and antimicrobial activity. The most biologically active compound was compound (9) with thieno and furan moieties attached to the indole core, which could serve as a lead for further optimization to develop potent molecules targeting microbial diseases.

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