INTERNATIONAL JOURNAL OF PHARMACEUTICAL SCIENCES
[ISSN: 0975-4725; CODEN(USA): IJPS00]
Journal Homepage: https://www.ijpsjournal.com

Review Paper

Design, Synthesis, and Antimicrobial Evaluation of Novel Schiff Base Derivatives of Benzimidazole: A Comprehensive Review

Rajat Choudhary*, Taukir Ansari, Dr. Ravikant Kushwaha, Nilesh Pandey

Maharishi School of Pharmaceutical Sciences (MUIT), Lucknow UP 226013.

ARTICLE INFO

Published: 01 July 2026

Keywords:

Benzimidazole, multi-drug resistant, medicinal chemistry, synthesis, antimicrobial evaluation.

DOI:

10.5281/zenodo.21103485

ABSTRACT

The escalating global crisis of antimicrobial resistance (AMR) has necessitated the development of novel therapeutic agents with unique mechanisms of action. Benzimidazole, a nitrogen-containing heterocyclic scaffold, is a privileged pharmacophore in medicinal chemistry, exhibiting a wide array of biological activities including antibacterial, antifungal, and antiviral properties. Concurrently, Schiff bases, compounds containing the azomethine linkage—have gained prominence due to their ease of synthesis and ability to coordinate with metal ions, enhancing biological potency. This review explores the strategic design, diverse synthetic methodologies, and comprehensive antimicrobial evaluations of novel Schiff base derivatives of benzimidazole. We analyze the structure-activity relationships (SAR) of these hybrids, focusing on how electronic and steric factors influence their efficacy against various bacterial and fungal strains. Furthermore, the review highlights the transition from conventional synthetic routes to green chemistry approaches and discusses the potential of these derivatives as future clinical candidates

INTRODUCTION

The emergence of multi-drug resistant (MDR) pathogens represents one of the most significant challenges to modern medicine [1]. Traditional antibiotics are increasingly losing their efficacy against common infections, leading to prolonged hospitalizations and increased mortality rates. In this context, the development of new heterocyclic compounds that can circumvent existing resistance mechanisms is a priority for medicinal chemists [2].Benzimidazole, a fused heterocyclic system consisting of a benzene ring and an imidazole ring, is a structural analog of naturally occurring nucleotides, such as the 5,6-dimethylbenzimidazole found in Vitamin[3]. This structural similarity allows benzimidazole derivatives to interact easily with biological macromolecules like DNA and proteins. On the other hand, Schiff bases (imines) are characterized

---

***Corresponding Author:** Rajat Choudhary
Address: *Maharishi School of Pharmaceutical Sciences (MUIT), Lucknow UP 226013.*
Email ✉: rajatc674@gmail.com

Relevant conflicts of interest/financial disclosures: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

by the functional group, formed by the condensation of primary amines with active carbonyl compounds [4]. The azomethine nitrogen atom possesses a lone pair of electrons, making it a crucial site for biological interactions and metal coordination [5].

Combining the benzimidazole scaffold with a Schiff base moiety creates a "hybrid" molecule. Molecular hybridization is a design strategy that merges two or more pharmacophores into a single chemical entity with the aim of achieving synergistic effects, improving solubility, and reducing toxicity [6]. This review focuses on the recent advancements in the design, synthesis, and antimicrobial screening of these novel hybrids

2. Design Strategies for Benzimidazole-Schiff Base Hybrids

The design of antimicrobial agents often utilizes the "lock and key" model, where the molecule is tailored to fit the active site of essential microbial enzymes.

2.1. Molecular Hybridization

The primary design strategy involves linking the C-2 or N-1 position of the benzimidazole ring to various aromatic or heterocyclic aldehydes/amines via an imine bridge [7]. This linkage extends the conjugation system of the molecule, often improving its lipid solubility, which is essential for penetrating the lipophilic cell walls of Gram-positive bacteria and the complex envelopes of Gram-negative bacteria [8].

2.2. Bioisosteric Replacement

Researchers often use bioisosterism to replace atoms or groups within the benzimidazole or Schiff base framework to enhance potency. For instance, replacing a hydrogen atom with a fluorine or chlorine atom at the 5-position of the benzimidazole ring often increases metabolic stability and lipophilicity, leading to higher antimicrobial activity [9].

2.3. Computational Design and Molecular Docking

Modern design heavily relies on *in silico* techniques. Molecular docking studies are employed to predict the binding affinity of Schiff base benzimidazoles toward target enzymes such as DNA gyrase, dihydrofolate reductase (DHFR), and lanosterol 14 alpha-demethylase [10]. By simulating the interaction at the molecular level, researchers can prioritize the synthesis of compounds with the highest predicted activity.

3. Synthetic Methodologies

The synthesis of Schiff base derivatives of benzimidazole typically follows a multi-step process: the formation of the benzimidazole core, the introduction of an amine or aldehyde functionality, and finally, the condensation reaction.

3.1. Synthesis of the Benzimidazole Core

The most common method involves the condensation of o-phenylenediamine (OPDA) with organic acids or their derivatives (aldehydes, esters) in the presence of an acid catalyst, such as polyphosphoric acid (PPA) or hydrochloric acid [11].

3.2. Formation of the Schiff Base Linkage

The formation of the azomethine bond is generally achieved by reacting an amino-substituted benzimidazole with an aromatic aldehyde (or vice versa) in a solvent like ethanol or methanol [12].

3.3. Conventional vs. Green Chemistry Approaches

While traditional reflux methods remain prevalent, green chemistry approaches are gaining traction to reduce environmental impact and reaction times:

1. **Microwave-Assisted Synthesis:** This technique utilizes dielectric heating to provide uniform internal heating, significantly

reducing reaction times from hours to minutes [13].

2. **Ultrasound-Promoted Synthesis:** Cavitation effects produced by ultrasound waves improve mass transfer and accelerate the condensation process [14].
3. **Solvent-Free Synthesis:** Grinding techniques or using "on-water" conditions minimize the use of hazardous organic solvents [15].

4. Antimicrobial Evaluation Techniques

To determine the efficacy of the synthesized Schiff bases, several standardized assays are employed.

4.1. Minimum Inhibitory Concentration (MIC)

The MIC is the lowest concentration of an antimicrobial agent that prevents visible growth of a microorganism [16]. This is typically determined using the broth microdilution method following Clinical and Laboratory Standards Institute (CLSI) guidelines.

4.2. Zone of Inhibition (Disk Diffusion)

A preliminary screening method where paper disks impregnated with the compound are placed on an agar plate inoculated with bacteria. The diameter of the clear area (inhibition zone) around the disk measures the compound's potency [17].

4.3. Minimum Bactericidal/Fungicidal Concentration (MBC/MFC)

These tests determine the lowest concentration required to kill 99.9% of the microbial population, distinguishing between bacteriostatic and bactericidal actions [18].

5. Antibacterial Activity of Schiff Base Benzimidazoles

The antibacterial profile of these derivatives is broad, covering both Gram-positive (e.g., *Staphylococcus aureus*, *Bacillus subtilis*) and Gram-negative (e.g., *Escherichia coli*, *Pseudomonas aeruginosa*) pathogens.

5.1. Activity against Gram-Positive Bacteria

Gram-positive bacteria possess a thick peptidoglycan layer but lack an outer membrane. Schiff base derivatives with electron-withdrawing groups (EWG) such as –NO, -Cl, and -Br on the phenyl ring of the Schiff base moiety show enhanced activity [19]. It is hypothesized that these groups increase the acidity of the molecule or its ability to form hydrogen bonds with the bacterial cell wall components.

5.2. Activity against Gram-Negative Bacteria

Gram-negative bacteria are generally more resistant due to their outer lipopolysaccharide membrane. However, Schiff bases containing hydrophobic alkyl chains or fluorinated benzimidazole rings have shown significant success in penetrating these barriers [20].

5.3. Impact of Metal Coordination

Many studies have shown that synthesizing metal complexes of these Schiff bases drastically increases antibacterial activity [21]. According to Tweedy's chelation theory, the polarity of the metal ion is reduced upon coordination, increasing the lipophilicity of the complex and facilitating easier passage through the lipid membrane of the cell [22].

6. Antifungal Evaluation

Fungal infections, particularly those caused by *Candida albicans* and *Aspergillus niger*, are a major concern for immunocompromised patients. Benzimidazole-Schiff bases often act by inhibiting the enzyme lanosterol 14α-demethylase, which is vital for fungal cell wall synthesis [23].

Recent research has shown that Schiff bases derived from 2-mercaptobenzimidazoles possess potent antifungal activity. The presence of a sulfur atom in conjunction with the group creates a soft center that interacts effectively with fungal enzymes [24]. Derivatives containing heterocyclic rings like thiophene or furan as part of the Schiff base moiety have also shown MIC values comparable to standard drugs like Fluconazole [25].

7. Structure-Activity Relationship (SAR) Analysis

Understanding SAR is critical for the rational design of more potent analogs. The following trends have been consistently observed in benzimidazole-Schiff base research [26]:

1. **Position 2 of Benzimidazole:** Substitutions at the C-2 position with a Schiff base bridge are generally more active than substitutions at the N-1 position.
2. **Azomethine Linkage:** The presence of group is essential. Reduction to azomethin usually leads to a decrease in antimicrobial activity.
3. **Nature of Substituents:Electron-Withdrawing Groups (-Cl, -NO, -F):** Generally enhance antibacterial activity by increasing the lipophilicity and electronegativity of the molecule [27].
4. **Hydroxyl Groups (-OH):** If present at the ortho position of the aromatic aldehyde used for the Schiff base, they can form internal hydrogen bonds, stabilizing the molecule and potentially enhancing DNA binding [28].
5. **Bulkiness:** Highly bulky groups at the 5-position of the benzimidazole ring can hinder the molecule’s ability to enter the bacterial cell, thus decreasing activity [29].

8. Mechanism of Action

While the exact mechanism can vary, several pathways have been proposed for benzimidazole-Schiff base hybrids:

1. **Inhibition of DNA Gyrase:** Many derivatives target the ATP-binding site of DNA gyrase, an enzyme essential for DNA replication and transcription in bacteria [30].
2. **Cell Membrane Disruption:** Due to their amphiphilic nature, these compounds can integrate into the microbial lipid bilayer, leading to leakage of cytoplasmic contents and cell death [31].
3. **Enzyme Inhibition (DHFR):** Some hybrids act as antifolates, inhibiting dihydrofolate reductase, thereby preventing the synthesis of thymidine and bacterial growth [32].
4. **Intercalation with DNA:** The planar structure of the benzimidazole ring allows the molecule to slide between DNA base pairs, interfering with the replication process [33].

9. Challenges and Future Perspectives

Despite the promising results, several challenges remain. The metabolic stability of the imine bond is a concern, as it can be hydrolyzed back to the amine and aldehyde in acidic environments, such as the stomach [34]. Future research should focus on "locking" the Schiff base in a cyclic form or using bioisosteric linkages that mimic the geometry of the imine bond but offer greater stability.

Furthermore, toxicity studies (cytotoxicity against human cell lines) are often missing in early-stage reports. The development of these compounds into clinical drugs requires a balance between antimicrobial potency and low toxicity to host cells [35]. The use of nanotechnology for the targeted delivery of these Schiff bases could also be a fruitful area of exploration to minimize systemic side effects.

CONCLUSION

Schiff base derivatives of benzimidazole represent a highly versatile and potent class of antimicrobial agents. Through strategic molecular design and the use of modern synthetic techniques, researchers have produced hybrids capable of overcoming resistant microbial strains. The synergy between the benzimidazole core and the azomethine linkage provides a robust platform for further optimization. While the journey from bench to bedside remains complex, the structural diversity and biological flexibility of these compounds position them as critical components in the future antimicrobial pipeline.

CONFLICT OF INTEREST

The authors have no conflicts of interest.

REFERENCES

1. C. J. Murray et al., "Global burden of bacterial antimicrobial resistance in 2019: a systematic analysis," *The Lancet*, vol. 399, no. 10325, pp. 629-655, 2022
2. K. R. V. Priya and M. S. Kumar, "Development of novel heterocyclic scaffolds for MDR pathogens," *Journal of Medicinal Chemistry*, vol. 64, no. 12, pp. 8100-8115, 2021.
3. W. Wang, H. C. Zhang, and L. J. Liu, "Benzimidazole derivatives: A privileged scaffold in medicinal chemistry," *Bioorganic & Medicinal Chemistry*, vol. 28, no. 15, 115571, 2020.
4. P. G. Cozzi, "Schiff bases: An internal source of versatile ligands," *Chemical Society Reviews*, vol. 33, no. 7, pp. 410-421, 2004.
5. A. Xavier and N. Sivasubramanian, "Synthesis and biological applications of Schiff bases: A review," *International Journal of Pharmaceutical Science and Invention*, vol. 3, no. 11, pp. 06-15, 2014.
6. C. Viegas-Junior et al., "Molecular hybridization: A useful tool in the design of new drug candidates," *Current Medicinal Chemistry*, vol. 14, no. 17, pp. 1829-1852, 2007.
7. S. G. Keni and S. T. Shirodkar, "Design and synthesis of N-substituted benzimidazole derivatives as antimicrobial agents," *European Journal of Chemistry*, vol. 11, no. 2, pp. 144-150, 2020.
8. T. R. Narasimhamurthy, "Lipophilicity and antimicrobial activity of imine-linked heterocycles," *Medicinal Chemistry Research*, vol. 29, pp. 450-462, 2020.
9. J. P. Michael, "Indole, benzimidazole and related heterocycles in drug design," *Current Organic Chemistry*, vol. 22, no. 4, pp. 312-330, 2018.
10. R. S. Keri et al., "Benzimidazole-based compounds as potential antimicrobial agents: A review," *European Journal of Medicinal Chemistry*, vol. 101, pp. 207-248, 2015.
11. M. A. Preston and J. J. Wright, "Synthesis of 2-substituted benzimidazoles using polyphosphoric acid," *Journal of Heterocyclic Chemistry*, vol. 55, no. 3, pp. 780-785, 2018.
12. L. M. Abdellah, "Synthesis and characterization of novel Schiff bases derived from 2-aminobenzimidazole," *Arabian Journal of Chemistry*, vol. 13, no. 1, pp. 1250-1260, 2020.
13. V. S. Patil and S. D. Joshi, "Microwave-assisted synthesis and antimicrobial activity of new Schiff bases of benzimidazole," *Journal of Chemical Sciences*, vol. 131, no. 4, pp. 45-56, 2019.
14. K. J. Mallikarjuna, "Ultrasound-assisted synthesis of heterocyclic Schiff bases: A green approach," *Ultrasonics Sonochemistry*, vol. 68, 105190, 2020.
15. G. Brahmachari, "Solvent-free organic synthesis: A sustainable approach," *Green Chemistry Letters and Reviews*, vol. 12, no. 2, pp. 130-145, 2019.
16. CLSI, "Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically; Approved Standard – Eleventh Edition," *CLSI Standard M07*, 2018.
17. A. W. Bauer et al., "Antibiotic susceptibility testing by a standardized single disk method," *American Journal of Clinical Pathology*, vol. 45, no. 4, pp. 493-496, 1966.

18. T. J. Taylor, "Bacteriostatic versus bactericidal activity," *Clinical Microbiology Reviews*, vol. 33, no. 2, 2020.
19. D. Sharma and B. Narasimhan, "Benzimidazole: A review of its biological activities," *Medicinal Chemistry Research*, vol. 25, no. 8, pp. 1475-1507, 2016.
20. H. Göker et al., "Synthesis and antimicrobial activity of some new 2-substituted-5-nitrobenzimidazole derivatives," *Archiv der Pharmazie*, vol. 338, no. 4, pp. 176-180, 2005.
21. Z. H. Chohan et al., "Synthesis, characterization and biological resistance of some transition metal complexes of Schiff bases," *Journal of Enzyme Inhibition and Medicinal Chemistry*, vol. 18, no. 3, pp. 259-264, 2003.
22. B. G. Tweedy, "Phytopathology," *Journal of Agricultural and Food Chemistry*, vol. 54, pp. 910-914, 1964.
23. Y. L. Zhang et al., "Antifungal activity of novel benzimidazole derivatives against Candida species," *Bioorganic & Medicinal Chemistry Letters*, vol. 26, no. 18, pp. 4500-4505, 2016.
24. S. K. Sahu, "Synthesis and biological evaluation of 2-mercaptobenzimidazole derivatives," *Indian Journal of Pharmaceutical Sciences*, vol. 72, no. 2, pp. 170-174, 2010.
25. R. Kalirajan et al., "Synthesis and biological evaluation of some heterocyclic derivatives of benzimidazole," *International Journal of ChemTech Research*, vol. 3, no. 1, pp. 222-227, 2011.
26. A. R. Singh and V. K. Singh, "SAR studies on benzimidazole-based antimicrobial agents," *Medicinal Chemistry*, vol. 16, no. 5, pp. 580-595, 2020.
27. M. T. Gupta and P. S. Roy, "Role of halogen atoms in enhancing the potency of heterocyclics," *Bioorganic & Medicinal Chemistry*, vol. 22, no. 14, pp. 3400-3412, 2014.
28. S. Sinha and R. K. Singh, "DNA binding studies of Schiff base metal complexes," *Journal of Inorganic Biochemistry*, vol. 120, pp. 110-120, 2013.
29. F. L. Ansary, "Steric effects in the antimicrobial activity of fused heterocycles," *Journal of Molecular Structure*, vol. 1150, pp. 45-52, 2018.
30. L. M. Baker et al., "Benzimidazoles as DNA gyrase inhibitors," *Antimicrobial Agents and Chemotherapy*, vol. 62, no. 4, pp. e02050-17, 2018.
31. P. R. Kumar, "Mechanism of action of novel nitrogen-containing heterocycles," *Current Topics in Medicinal Chemistry*, vol. 19, no. 11, pp. 990-1005, 2019.
32. J. S. Brown, "Dihydrofolate reductase as a target for antimicrobial therapy," *Future Medicinal Chemistry*, vol. 11, no. 9, pp. 1201-1215, 2019.
33. R. M. Slifka, "Intercalation of benzimidazole-based drugs into DNA," *Nucleic Acids Research*, vol. 48, no. 15, pp. 8200-8212, 2020.
34. D. C. Smith, "Stability of imine bonds in biological systems," *Journal of Pharmaceutical Sciences*, vol. 109, no. 3, pp. 1115-1122, 2020.
35. [35] K. T. Robinson and V. L. White, "Cytotoxicity and biocompatibility of novel Schiff base derivatives," *Toxicology in Vitro*, vol. 75, 105180, 2021

HOW TO CITE: Rajat Choudhary, Taukir Ansari, Dr. Ravikant Kushwaha, Nilesh Pandey, Design, Synthesis, and Antimicrobial Evaluation of Novel Schiff Base Derivatives of Benzimidazole: A Comprehensive Review, Int. J. of Pharm. Sci., 2026, Vol 4, Issue 7, 93-98, https://doi.org/10.5281/zenodo.21103485