Vol 8, Issue 6, 2025 **Impact Factor: 8.535** DOI: https://doi.org/10.31426/ijamsr.2025.8.6.8427

IJAMSR

International Journal of Advanced Multidisciplinary Scientific Research (IJAMSR) ISSN:2581-4281

Spectroscopic Characterization and Antibacterial Activity of Newly Synthesized Benzothiazole Derivatives

Wadnere Kishor Eknath [1]

[1] Research Scholar, Department of Chemistry, Mansarovar Global University, Sehore, M.P., India.

Dr. Pushpendra Tiwari [2]

[2] Supervisor, Department of Chemistry, Mansarovar Global University, Sehore, M.P., India.

ABSTRACT

Synthesis, spectral characterization, and antibacterial assessment of benzothiazole-based thiourea and sulphonamide derivatives are described in the current work. Through spectroscopic investigations using FT-IR, ´1H NMR, and 13C NMR, the synthesized compounds' structures were confirmed. The compounds' antibacterial activity was assessed by comparing them to Gram-positive Staphylococcus aureus and Gram-negative Escherichia coli germs using agar well diffusion and broth dilution techniques. Findings showed that antibacterial activity was higher in sulphonamide derivatives than in thiourea and benzothiazole precursors. Larger zones of inhibition and lower minimum inhibitory concentration values against S. aureus indicated that compound 3b, among the produced compounds, had the most powerful antibacterial action.

Keywords: *Antibacterial, Benzothiazole Derivatives, Sulphonamide, Thiourea, Bacteria.*

I. Introduction

Benzothiazole is an important family of nitrogen- and sulfur-containing heterocycles in medicinal chemistry; it is a heterocyclic molecule with a fused benzene and thiazole ring structure. Benzothiazole derivatives have attracted a lot of attention from scientists in recent decades due to the wide range of chemical and biological characteristics imparted by their unusual structural framework. Compounds containing thiazole rings and aromatic benzene systems have a broad range of pharmacological effects due to the many interactions they may form with biological macromolecules.

Benzothiazole derivatives have a well-established therapeutic use. The capacity of these chemicals to bind to key cellular components, such as nucleic acids, enzymes, and receptors, has led to their impressive antibacterial, anticancer, anti-inflammatory, antioxidant, antiviral, and antitubercular effects. Because of the concerning increase in antibiotic resistance among harmful bacteria, their

IJAMSR

International Journal of Advanced Multidisciplinary Scientific Research (IJAMSR) ISSN:2581-4281

antimicrobial properties have attracted a lot of interest. One potential strategy for fighting MDR bacteria is benzothiazole derivatives, which may hinder bacterial growth by interacting with processes involved in cell wall formation, protein biosynthesis, and nucleic acid activity. Their potential as medicinal agents has been expanded by investigations into their antifungal properties against many clinically significant fungi.

Benzothiazole derivatives have shown strong anticancer activity against many cancer cell lines in oncology. Their anticancer actions are mediated by a wide variety of molecular mechanisms, including apoptosis induction, cell cycle arrest, tubulin polymerization inhibition, and suppression of important signaling pathways including PI3K/Akt and MAPK. A number of these derivatives have shown promising anticancer drug development lead compounds due to their specific cytotoxicity against cancer cells with no effects on normal cells. The fact that benzothiazoles may block topoisomerase and serve as DNA intercalators further adds to their importance in cancer treatment.

Benzothiazole derivatives have also been the subject of much research into their antioxidant and anti-inflammatory properties. Inflammation and oxidative stress have important roles in the development of many long-term illnesses, such as diabetes, cardiovascular disease, and neurodegenerative disorders. Due to their heterocyclic and aromatic electron-rich structure, benzothiazole derivatives have the ability to scavenge reactive oxygen species (ROS) and suppress pro-inflammatory mediators. These chemicals have the potential to manage illnesses associated with oxidative stress because of their characteristics. The neuroprotective characteristics of some benzothiazole derivatives suggest they might be useful in the fight against neurodegenerative disorders like Parkinson's and Alzheimer's.

The antiviral and antitubercular actions of benzothiazole derivatives further demonstrate their flexibility. The need to find novel treatment agents has been driven by the recent rise of resistant virus strains and Mycobacterium tuberculosis. There is evidence that benzothiazoles block viral enzymes including HIV-1 reverse transcriptase and hepatitis C virus NS5B polymerase, in addition to important enzymes of Mycobacterium tuberculosis. They are promising scaffolds for the development of next-generation antiviral and antitubercular medicines due to their multifunctional biological profile and simplicity of structural alteration.

Scientists may now create a wide variety of analogs with improved biological characteristics thanks to developments in the chemical synthesis of benzothiazole derivatives. Typical procedures include subjecting 2-aminothiophenol to acidic or oxidative conditions in order to cyclize it with different carboxylic acids, aldehydes, or their derivatives. Green chemistry, microwave-assisted synthesis, and metal-catalyzed reactions are some of the modern synthetic techniques that endeavor to increase selectivity, decrease environmental impact, and enhance reaction efficiency. The therapeutic potential of these derivatives may be enhanced by fine-tuning their physicochemical and pharmacological characteristics by introducing various substituents at key places of the benzothiazole ring.

The structure-activity relationship (SAR) of benzothiazole derivatives has been extensively studied

IJAMSR

International Journal of Advanced Multidisciplinary Scientific Research (IJAMSR) ISSN:2581-4281

using computational methods, such as molecular docking and quantitative structure-activity relationship (QSAR) research. Predicting binding affinities with target enzymes and receptors, identifying critical pharmacophores, and rationally designing novel analogs with increased effectiveness are all made possible by these in silico approaches. Researchers have shown that combining computational modeling with synthetic chemistry speeds up drug development, lowers experimental costs, and shortens the time it takes to bring potential medication ideas to market.

II. Review of Literature

Sallal, Zainab. (2019) To create 2-bromophenylimidazo (1, 2-a) benzothiazole, researchers in this work reacted bromophenyl phencyl bromide with 2-amino-benzothiazole [1]. After that, combine 2-bromophenyl imidazo (1,2-a) benzothiazol [1] with POCl3, DMF, and CH3Cl to get 2-bromophenyl imidazo (1,2-a) benzothiazol-3-carbaldyhade [2]. 2-Bromo phenyl imidazo (1, 2-a) benzothiazole-3-carpaldehyde [2] reacted with various primary aromatic amines to produce Schiff base derivatives [A1-A3]. A series of new oxazepine derivatives (B1–B3) were synthesized by reacting amino acids (A1–A3) with malic anhydride. New beta-lactam compounds [C1-C3] were prepared by reacting the amino acids [A1-A3] with phenyliso cyanide. Finally, different Schiff bases [A1-A3] were reduced to create novel imidazo (1, 2-a) benzothiazole amino derivatives [D1-D3]. The compounds that were produced were evaluated using FT-IR and melting point analysis. A few of them have been annotated with 1H-NMR and 13C-NMR spectra. Researchers also tested the antimicrobial effects of many produced derivatives using a variety of microorganisms.

Redayan, Muayed et al., (2017) Synthesis of novel Schiff base derivatives containing benzimidazole rings is the focus of the current investigation. A series of benzimidazole derivatives bearing free -NH2 groups were produced by reacting o-pheneylenediamine with a number of amino acids, including glycine, alanine, phenyl alanine, and tyrosine, in the presence of 6N HCl. This reaction produced compounds 1(a-d). Afterwards, by reacting with a variety of aromatic aldehydes, these compounds might be used to create numerous Schiff bases. The chemical composition of the produced substances was validated by the use of FTIR, ´H, ´^{3}C-NMR, and ´^{3}C-NMR dept135 spectroscopy. The in vitro antibacterial activity of a few substances was tested against two kinds of Gram-positive bacteria (Staphylococcus aureus and Bacillus subtilis) and two kinds of Gram-negative bacteria (Pseudomonas aeruginosa and Escherichia coli). When tested against the gold standard antibiotics ampicillin and ciprofloxacin, the majority of these compounds showed promising antibacterial activity.

Baluja, S et al., (2017) Using infrared, nuclear magnetic resonance, and mass spectral data, many new Schiff bases and thiazolidinones were characterized after being synthesized from 6-methoxy 2-amino benzothiazole. Both dimethyl sulfoxide (DMF) and dimethyl ether (DMSO) were used for the antibacterial screening of these newly produced chemicals. The chemicals' inhibitory effects varied depending on the solvent (DMF vs. DMSO), which in turn affected the levels of inhibition in various strains. It seems that the anti-microbial action of a chemical is affected by its molecular structure, the solvent utilized, and the specific strain being studied.

Bansod, Nitin et al., (2013) To test for antibacterial activity, this work prepared and tested five novel

IJAMSR

International Journal of Advanced Multidisciplinary Scientific Research (IJAMSR) ISSN:2581-4281

metal complexes of the ligand 2-amino acetate, 6-chloro benzothiazole with various metal ions, including Ni(II), Cu(II), Zn(II), Cd(II), and Sn(II). In order to characterize the produced complexes, we measured their conductivity, electronic spectra, infrared spectra, magnetic moments, and elemental analyses. The complexes' monomer structures were suggested based on spectral measurements. The copper complex was suggested to have a square planar shape. It was thought that the other complexes would have a tetrahedral shape. Use of the disc diffusion approach allowed for the investigation of antimicrobial activity against both Gram-positive and Gram-negative harmful bacteria. and take pleasure in the activities that were noted.

Alang, G. et al., (2010) The antimicrobial activity of seven newly synthesized benzothiazole derivatives (R1–R7) was examined in this work. 2-Benzothiazolamines were produced by the reaction of p-toluidine with ammonium thiocynate; hydrazino derivatives were produced by the reaction of hydrazine hydrate with these compounds. The hydrazine derivative was reactated with several acetophenones, including 2'-fluoroacetophenone, 4'-fluoroacetophenone, 2'-chloroacetophenone, 4'-chloroacetophenone, p-hydroxyacetophenone, 2'-hydroxyacetophenone, and 2, 5-dihydroxyacetophenone, to produce compounds R1–R7. By using IR and NMR, we were able to identify each of the synthetic substances. Researchers looked into the antimicrobial properties and found that they worked.

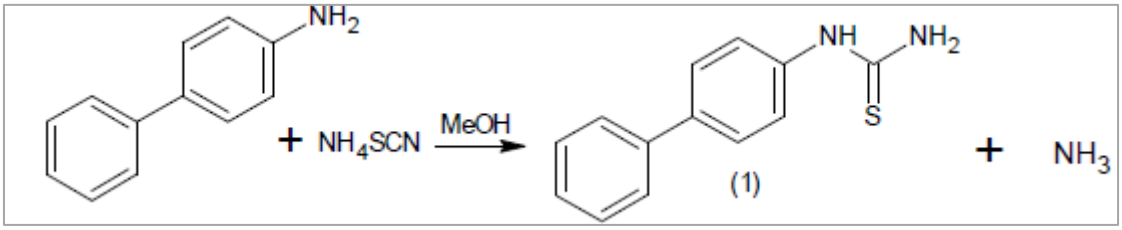
III. Material and Methods

Chemicals and Reagents

All chemicals and reagents used in the present investigation were of analytical grade and were employed without further purification. Solvents were obtained from standard commercial suppliers and used as received. Silica gel (mesh size 120–160) was used for column chromatography, while silica gel-G served as the stationary phase for thin layer chromatography (TLC).

Synthesis of Compounds

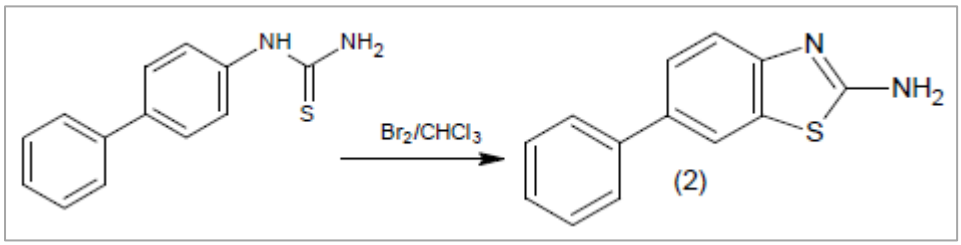
Two compounds, 2-amino-6-phenylbenzothiazole (2) and N-(biphenyl-4-yl)thiourea (1), were synthesized using established procedures. At normal temperature, N-(biphenyl-4-yl)thiourea (1) was produced by reacting biphenylamine with ammonium thiocyanate. A cream-colored crystalline solid known as 2-amino-6-phenylbenzothiazole (2) was created by cyclization of the synthetic thiourea with a recently obtained bromine in chloroform solution.



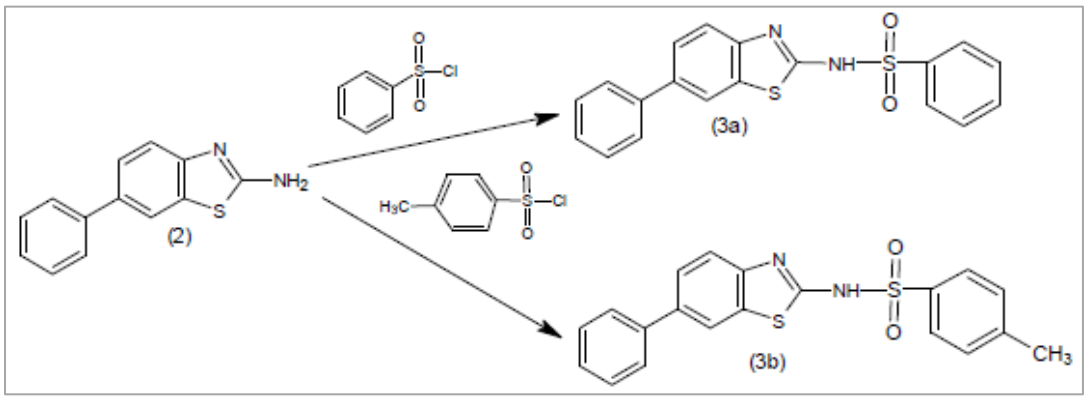
Scheme 1: Synthesis of N-biphenyl-4-yl thiourea (1)

IJAMSR

International Journal of Advanced Multidisciplinary Scientific Research (IJAMSR) ISSN:2581-4281



Scheme 2: Synthesis of 2-Amino-6-phenylbenzothiazole (2)



Scheme 3: Synthesis of benzothiazole derivatives of sulphonamide (3a & 3b)

To further modify compound (2), it was reacted with benzenesulphonyl chloride to produce N-(6-phenyl-1,3-benzothiazole-2-yl) benzenesulphonamide (3a) and with p-toluenesulphonyl chloride to produce N-(6-phenyl-1,3-benzothiazole-2-yl)-4-methylbenzenesulphonamide (3b), respectively. Before the compounds were characterized and tested biologically, they underwent purification using column chromatography.

Physical Measurements and Instrumentation

Using an open capillary melting point equipment, the synthesized compounds' melting points were determined and are presented as uncorrected values. Using TLC, we were able to track the start and end of reactions. The National Research Institute for Chemical Technology (NARICT) in Zaria followed normal protocols to record infrared (FT-IR) spectra using KBr pellets on a Shimadzu FT-IR 8400S spectrophotometer in the range of 4000-400 cm^{-1}.

^{1}H and ^{13}C nuclear magnetic resonance (NMR) spectra of compounds 1 and 2 were recorded in $CDCl_3$ using an Agilent FT-NMR spectrometer. Compounds 3a and 3b's ´H and ´^{3}C NMR spectra were also acquired in $CDCl_3$ using a Bruker WM 300 FT-NMR spectrometer.

Antibacterial Activity Assay

We used the agar well diffusion technique to test the synthetic compounds' antibacterial properties. The Gram-positive Staphylococcus aureus and Gram-negative Escherichia coli bacteria were used to test the antibacterial activity. Disperse 0.1 mL of each standardized bacterial culture equally over sterile agar plates. The bacterial cultures were standardized to around 10^{8} CFU/mL. The agar was aseptically drilled with a 6-millimeter diameter hole after it had dried.

IJAMSR

International Journal of Advanced Multidisciplinary Scientific Research (IJAMSR) ISSN:2581-4281

The solution of the test chemical, which was produced at a concentration of 20 mg/mL, was 200 µL thick and added to each well. As a reference medication, ampicillin was utilized. Zones of inhibition were determined in millimeters after incubating the inoculation plates at 37 °C for 24 hours.

Determination of Minimum Inhibitory Concentration (MIC)

The compounds that were produced had their minimum inhibitory concentration (MIC) determined by means of the broth dilution procedure. The concentrations of the test solutions were varied between 12.5 and 200 µg/mL. A compound's minimum inhibitory concentration (MIC) was defined as the concentration at which observable bacterial growth was utterly suppressed.

IV. Results and Discussion

Spectral Characterization of Synthesized Compounds

In order to validate their structural properties, the produced compounds underwent spectroscopy using FT-IR, ^{1}H NMR, and ^{13}C NMR. The FT-IR spectra displayed distinct absorption bands between 2040 and 2220 cm^{-1}, which correspond to the -N-C=S / -N-S-C-functional group. The distinct absorption bands seen in the 1345–1445 cm^{-1} range in compounds 3a and 3b are thought to be caused by the symmetric and asymmetric stretching vibrations of the sulphonyl (–SO_2-) group. The compounds' aromatic character was validated by the presence of C=C stretching vibrations detected between 1595 and 1685 cm^{-1}.

Typical signals for the protons of the benzothiazole ring were observed in the range of δ 3.60-3.85 ppm in the ^{13}C NMR spectra of the benzothiazole derivatives. The presence of electron-withdrawing sulphonyl groups and conjugation in compounds 3a and 3b caused the aromatic protons of the phenyl substituents linked to the benzothiazole moiety to resonate in the downfield area of δ 7.40-8.35 ppm, suggesting deshielding effects.

Signals related to benzothiazole carbons in the area of δ 38.50-41.20 ppm were seen in the ^{13}C NMR spectra of compound 2. These signals moved downfield to δ 45.10-74.80 ppm after sulphonylation to produce compounds 3a and 3b, indicating the electronic impact of the sulphonamide substituents. Because the sulphonyl functionality strongly withdraws electrons, the carbon atoms immediately bound to the -SO_2- group were seen farther downfield at δ 168.90-172.40 ppm. The structural integrity and effective synthesis of the target compounds are confirmed by these spectrum data.

Antibacterial Activity

Table 1: Zone of Inhibition of Compounds (mm)

Organism	Compd 1	Compd 2	Compd 3a	Compd 3b	Ampicillin
Staphylococcus aureus	15.5	18.0	27.5	21.0	33.0
Escherichia coli	9.0	–	18.5	22.0	26.0

Table 1 shows that in terms of antibacterial activity against Staphylococcus aureus, compound 3a outperformed all of the manufactured compounds. It produced a zone of inhibition of 27.5 mm, followed by compound 3b at 21.0 mm, compound 2 at 18.0 mm, and compound 1 at 15.5 mm. Compound 3a's significant inhibitory impact implies considerable antibacterial potential, even

IJAMSR

International Journal of Advanced Multidisciplinary Scientific Research (IJAMSR) ISSN:2581-4281

though the biggest zone of inhibition (33.0 mm) was observed with the conventional antibiotic ampicillin. In terms of activity against Escherichia coli, compound 3b (22.0 mm), compound 3a (18.5 mm), and compound 1 (9.0 mm) were the most effective, although compound 2 did not demonstrate any discernible action against this Gram-negative bacteria.

Table 2: Minimum Inhibitory Concentration (MIC) of Compounds (µg/mL)

Organism	Compd 1	Compd 2	Compd 3a	Compd 3b	Ampicillin
Staphylococcus aureus	>90	<80	60	<60	27.0
Escherichia coli	>120	–	>60	<60	15.4

The results of the zone of inhibition are further supported by the MIC values that are shown in Table 2. Compound 3b showed the most effective antibacterial action against Staphylococcus aureus with the lowest minimum inhibitory concentration (MIC) value (<60 µg/mL), followed by compound 3a (60 µg/mL) and compound 2 (<80 µg/mL). With a MIC value higher than 90 µg/mL, Compound 1 exhibited relatively lower action. Ampicillin proved to be more effective than the synthetic drugs, since it had the lowest MIC (27.0 µg/mL). Regarding Escherichia coli, a same pattern was noted; compound 3b outperformed the other produced compounds with a MIC value of less than 60 µg/mL, while compound 3a shown moderate activity (>60 µg/mL). Compound 2 showed no effect against this organism, while compound 1 had low activity (>120 µg/mL). With a MIC of 15.4 µg/mL, the conventional medicine ampicillin once again shown its higher potency.

V. Conclusion

The present investigation involved the effective synthesis and structural confirmation by FT-IR, ^{1}H NMR, and ^{13}C NMR spectroscopy of a series of thiourea and sulphonamide derivatives based on benzothiazole. The chemical structures and functional groups that were intended to be formed were confirmed by the spectrum data. The produced compounds showed strong antibacterial activity in biological testing, with the sulphonamide derivatives (3a and 3b) showing more activity than the thiourea and benzothiazole predecessors. Larger zones of inhibition and lower minimum inhibitory concentration values demonstrated that 3b had the most effective antibacterial action among the chemicals tested, especially against Staphylococcus aureus. Compound effectiveness is affected by bacterial cell wall structure and efflux mechanisms, as shown by the lower activity against Escherichia coli. The results highlight the potential of sulphonamide scaffolds based on benzothiazole as prospective leads for the creation of new antibacterial medicines, even if the synthesized derivatives were not as effective as the conventional antibiotic ampicillin. Their medicinal potential can be further enhanced with more structural optimization and comprehensive pharmacological research.

References

1. A. Mubarik, S. Mahmood, N. Rasool, M. A. Hashmi, M. Ammar, S. Mutahir, K. Ali, M. Bilal, M. Akhtar, and G. Ashraf, “Computational study of benzothiazole derivatives for conformational, thermodynamic and spectroscopic features and their potential to act as antibacterials,” *Crystals*, vol. 12, no. 7, pp. 1–16, 2022.

IJAMSR

International Journal of Advanced Multidisciplinary Scientific Research (IJAMSR) ISSN:2581-4281

2. D. Shaheed, J. Alshams, I. Alrubaie, H. Abbas, and A. J. Radhi, "Synthesis and studying biological activity of new benzothiazole derivatives," *Journal of Pharmaceutical Negative Results*, vol. 13, no. 4, pp. 573–578, 2022.
3. M. Khalil and Q. Khalal, "Synthesis and characterization of new compounds derived from 2-hydrazinobenzothiazole and evaluated their antibacterial activity," *Journal of Physics: Conference Series*, vol. 1853, Art. no. 012007, 2021, doi: 10.1088/1742-6596/1853/1/012007.
4. M. Elamin, A. Elaziz, and E. Abdallah, "Benzothiazole moieties and their derivatives as antimicrobial and antiviral agents: A mini-review," *International Journal of Research in Pharmaceutical Sciences*, vol. 11, no. 3, pp. 3309–3315, 2020.
5. Z. Sallal, "Antibacterial activity of new synthesized derivatives 3-substituted imidazo benzothiazole from 2-amino-benzothiazole," *Journal of Global Pharma Technology*, vol. 11, no. 3, pp. 433–438, 2019.
6. S. Choudhary, N. Kalra, and J. Govindasamy, "Synthesis, characterization and pharmacological evaluation of some newer benzothiazole derivatives," *Indian Journal of Pharmaceutical and Biological Research*, vol. 6, no. 2, pp. 31–36, 2018.
7. M. Redayan, M. Hussein, and A. Lafta, "Synthesis, spectroscopic characterization, and antibacterial evaluation of new Schiff bases bearing benzimidazole moiety," *Journal of Physics: Conference Series*, vol. 4, no. 11, pp. 1–8, 2017.
8. S. Baluja, S. Chanda, and A. Kulshrestha, "Synthesis, physical characterization and antibacterial activity of some derivatives of 2-amino benzothiazoles," *International Journal of Pharmaceutical Sciences Review and Research*, vol. 7, no. 4, pp. 1–13, 2017.
9. K. Kumar, N. Karthik, P. Begum, and C. Rao, "Synthesis, characterization and biological evaluation of benzothiazole derivatives as potential antimicrobial and analgesic agents," *Asian Journal of Research in Pharmaceutical Science*, vol. 7, no. 2, pp. 1–5, 2017.
10. S. Gupta, D. Agarwal, and A. Garg, "Synthesis, characterization and evaluation of antimicrobial activity of novel hybrid moieties—benzothiazoles and azetidinones," *International Journal of Pharmaceutical Sciences Review and Research*, vol. 6, no. 2, pp. 99–108, 2016.
11. M. Bhoi, M. Borad, H. Parmar, and H. Patel, "Synthesis and characterization of novel N-(benzo[d]thiazol-2-yl)-2-(2-(6-chloroquinolin-4-yl)hydrazinyl)acetamide derivatives containing quinoline linkage as potent antibacterial agents," *International Letters of Chemistry, Physics and Astronomy*, vol. 53, no. 1, pp. 114–121, 2015.
12. A. Salman, A. Abdel-Aziem, and M. Alkubbat, "Synthesis, spectroscopic characterization and antimicrobial activity of some new 2-substituted imidazole derivatives," *International Journal of Organic Chemistry*, vol. 5, no. 2, pp. 15–28, 2015.
13. M. Singh, S. K. Singh, M. Gangwar, G. Nath, and S. Singh, "Design, synthesis and mode of action of some benzothiazole derivatives bearing an amide moiety as antibacterial agents," *RSC Advances*, vol. 4, no. 36, pp. 19013–19023, 2014.
14. N. Bansod, G. Chaudhari, M. S. Thihale, and S. Patil, "Synthesis and antibacterial activity of new complexes of benzothiazole derivatives," *Der Pharma Chemica*, vol. 5, no. 2, pp. 144–148, 2013.
15. G. Alang, R. Kaur, G. Kaur, A. Singh, and P. Singla, "Synthesis and antibacterial activity of some new benzothiazole derivatives," *Acta Pharmaceutica Sciencia*, vol. 52, no. 2, pp. 213–218, 2010.