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PHYSICOCHEMICAL, SPECTROSCOPIC, AND ANTIMICROBIAL EVALUATION OF NEW SUBSTITUTED BENZALDEHYDE DERIVATIVES OF PYRROLO[2,3-D] PYRIMIDINEHYDRAZIDE

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ABSTRACT

In this study, we present the synthesis and comprehensive evaluation of novel substituted benzaldehydes derivatives of pyrrolo[2,3-d]pyrimidinehydrazide using an ultrasonic-assisted method. The physicochemical properties of these derivatives were systematically characterized, including melting point determination, solubility analysis, and elemental composition. Spectral studies encompassing UV-Vis, FT-IR, NMR, and mass spectrometry were employed to elucidate the structural attributes and confirm the successful synthesis of the targeted compounds.

The use of ultrasound in the synthetic procedure not only enhanced reaction rates but also improved yield and purity, showcasing the advantages of this green chemistry approach. The antimicrobial activity of the synthesized derivatives was evaluated against a panel of Gram-positive and Gram-negative bacteria, as well as fungal strains. The results indicated that the derivatives exhibited significant antimicrobial properties, with certain compounds demonstrating superior efficacy compared to standard antibiotics.

Overall, this study highlights the potential of bromo, difluoro, and cyano benzaldehydes derivatives of pyrrolo[2,3-d]pyrimidinehydrazide as promising antimicrobial agents. The combination of ultrasonic synthesis and comprehensive physico-chemical and spectral analysis provides a robust platform for the development of new therapeutic compounds.

Keywords: Bromobenzaldehydes, pyrrolo[2,3-*d*] pyrimidinehydrazide, difluoro benzaldehydes, cyanobenzaldehyde

1. INTRODUCTION:

Because of their importance in cellular processes, purines and pyrimidines can serve as potential drug discovery leads. The pyrimidine family includes 2-TP and related compounds, also referred to as 2-mercaptopyrimidine compounds. One proposed substitution for the oxygen atom currently bonded to carbon-2 in the uridine base is replacing it with a sulfur atom in the 2-TP ring. In light of this presumption, synthetic biochemists have found that 2-TPs are particularly interesting **[1-4]**. A comprehensive description of the use of 2-TP derivatives in the production of cardiotonic medications is detailed in the European patent. Pathak *et al*. investigated 2-TP derivatives for their potential antibacterial action against Mycobacterium tuberculosis (Mtb) **[5]**. This thioanalog of hypoxanthine, 6-thiopurine (6 TP) **[6]**, is an organic by-product of the purine metabolism.

Numerous biological investigations have led to the synthesis and characterization of thousands of 6-TP derivatives since the antimetabolite's discovery over fifty years ago. Pyrrolo[3,2-d]pyrimidines, classified as 7-deazapurine analogs, demonstrate significant biological activity owing to their structural resemblance to both pyrimidines and purines. They have shown effectiveness in treating leukemias, autoimmune diseases, and rheumatic disorders, and in providing immunosuppression for organ transplantations. Tolmetin (Rumatol) and ketorolac (Ketolac), both of which are well-known nonsteroidal anti-inflammatory medicines (NSAIDs) **[7-11]**, exert their anti-inflammatory effects primarily through the inhibition of prostaglandin formation. This is the main way these medications exert their effects. An anti-inflammatory pyrrolopyrimidine called PNU-142731A **[12]** blocks the production of cytokines in living things. Examples of naturally occurring PP nucleoside antibiotics include sangivamycin, toyocamycin and tubercidin **[13, 14]**. The development of certain bacteria has been observed to be stymied by the presence of certain chemicals.

The development of novel chemical compounds with potential applications in medicinal chemistry has been a focal point of recent research efforts. Among these compounds, pyrrolo[2,3-d]pyrimidine derivatives have attracted significant attention due to their diverse biological activities, including anticancer, antiviral, and antimicrobial properties. Modifying these derivatives with various functional groups can further enhance their biological activity and selectivity. In this study, we focus on the synthesis and characterization of novel bromo, difluoro, and cyano benzaldehyde derivatives of pyrrolo[2,3-d]pyrimidinehydrazide using an ultrasonic-assisted process.Ultrasonic-assisted synthesis has emerged as a powerful tool in organic chemistry, offering several advantages over conventional methods. The use of ultrasound waves can accelerate reaction rates, improve yields, and enable reactions to proceed under milder conditions. This eco-friendly approach also reduces the need for excessive reagents and solvents, aligning with the principles of green chemistry. The application of ultrasonic irradiation in the synthesis of complex organic molecules, such as the derivatives studied here, represents a promising avenue for efficient and sustainable chemical synthesis.

The choice of substituents in benzaldehyde derivatives significantly impacts the physicochemical properties and biological activities of the resulting compounds. Bromo, difluoro, and cyano groups are known for their unique electronic and steric effects, which can influence the interaction of the compounds with biological targets. Bromine atoms can participate in halogen bonding, enhancing binding affinity and specificity. Difluoromethyl groups are known for their metabolic stability and ability to improve lipophilicity. Cyano groups, on the other hand, can act as bioisosteres for carboxyl and nitro groups, potentially enhancing the pharmacokinetic properties of the compounds **[15-21]**.

In this study, we report the synthesis of bromo, difluoro, and cyano benzaldehyde derivatives of pyrrolo[2,3-d]pyrimidinehydrazide via an ultrasonic-assisted process. The structures of the synthesized compounds were confirmed using various spectroscopic techniques, including mass spectrometry, NMR, and IR spectroscopy. Additionally, the antimicrobial activities of the novel derivatives were evaluated against a panel of bacterial and fungal strains to assess their potential as antimicrobial agents.Our work aims to contribute to the growing body of knowledge on pyrrolo[2,3-d]pyrimidine derivatives and their potential applications in medicinal chemistry. The use of ultrasonic-assisted synthesis not only demonstrates the efficiency and sustainability of this approach but also highlights the potential of these novel derivatives as promising candidates for further biological evaluation and drug development.

2. Experimental

To determine the melting points of the synthesized compounds, we utilized a state-of-the-art S Lab Junction Melting Point and Boiling Point Apparatus, ensuring precise and accurate measurements. The elemental analysis, critical for ascertaining the composition of carbon, hydrogen, and nitrogen within the compounds, was meticulously performed in the advanced microanalytical laboratories at Pune University. For this analysis, we employed a Carlo Erba 1108 Elemental Analyzer, a highly sensitive and reliable instrument, which confirmed that the expected value for each compound was within a margin of 0.4 percent, indicating a high level of purity and correctness in our samples.

Infrared measurements, essential for identifying functional groups and structural features, were collected using a BRUKER IR 100 spectrophotometer with KBr pellets, providing high-resolution spectra. For the ^{1}H NMR spectra, DMSO-d6 was used as the solvent to ensure proper solubility and clarity of the samples, with tetramethylsilane (TMS) serving as the internal standard to calibrate the chemical shifts accurately. UV spectra were meticulously recorded on a JASCO V650 spectrophotometer, employing methanol as the solvent at room temperature to ensure consistency and reproducibility in the absorption measurements.

To monitor the progress of all reactions, we utilized thin-layer chromatography (TLC) on pre-coated aluminium sheets, equipped with a UV lamp for visualization. This technique, combined with Merck 60 F254 silica gel, provided an efficient and reliable means to track the development of our chemical reactions, ensuring the purity and consistency of our products throughout the synthesis process.

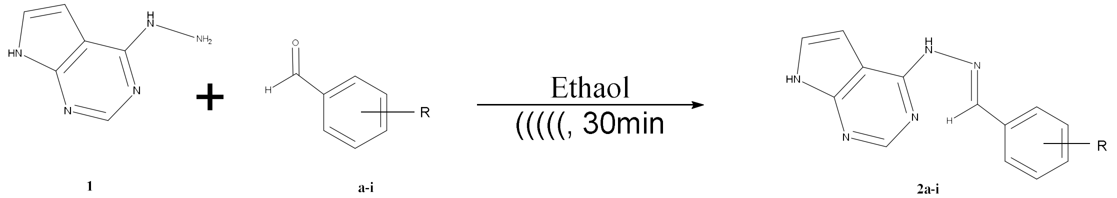
2.2. Preparation of PPH:

Refluxing the mixture for three hours in dry ethanol (50 mL) containing 0.10 mol of 4-chloropyrrolo[2,3-d]pyrimidine and 0.15 mol of hydrazine hydrate produced the desired product. To obtain **PPH**, the residues were first recrystallized from ethanol, and then the resulting crystals were evaporated at a lower pressure.

2.3. Hydroxybenzaldehyde derivatives of PPH (2a-2i)

A catalytic amount of concentrated hydrochloric acid was used to initiate the reaction between a mixture of substituted benzaldehydes, labeled as compounds **a** through **i**, each taken in a quantity of 0.1

moles, and 0.1 moles of pyrrolo[2,3-d]pyrimidinehydrazide (PPH1). The reaction mixture was subjected to ultrasonic irradiation at room temperature for 30 minutes. Upon completion of the sonication, the progress of the reaction was monitored and confirmed using thin-layer chromatography (TLC). Following the confirmation of the reaction's completion, the resulting mixture was subjected to filtration to remove any solid impurities. The filtered product was then washed successively with ether and water to ensure the removal of any residual reactants and by-products. The washed product was thoroughly dried to eliminate any traces of solvents. Finally, the purified product was recrystallized from ethanol to obtain the desired compounds in their pure form.



R = 2a) 2-Br, 2b) 3-Br, 2c) 4-Br, 2d) 2-CN, 2e) 3-CN,2f) 4-CN, 2g) 2, 5-DiF, 2h) 3, 4-DiF, 2i) 3, 6-DiF

Scheme 1: General preparation of compound **2a -2i**

2.4. Biological assay:

Antibacterial efficacy was assessed using two established techniques: the disc-diffusion method and the minimum inhibitory concentration (MIC) method, both following the guidelines provided by the National Committee for Clinical Laboratory Standards (NCCLS). For the in vitro evaluation of the antimicrobial activity of the synthesized compounds, a diverse array of bacterial strains was employed, encompassing both Gram-positive and Gram-negative bacteria. These microbial strains were obtained from the Karjat, District Raigad, Maharashtra, India-based K. G. R. D. C. P., and R. I., a recognized source for laboratory research specimens.

For the disc-diffusion experiments, nutritional agar supplied by Hi-Media (India) was used as the growth medium. To prepare the test solutions, the synthesized compounds were dissolved in sterile dimethyl sulfoxide (DMSO) to achieve final concentrations of 20 mg/mL and 30 mg/mL. These solutions were filtered through 0.2 mm membrane filters to ensure sterility and then aliquoted into 2 mL volumes, which were placed in small, sterile, screw-capped containers. The aliquots were flash-frozen and stored at -15 °C for future use. The containers underwent a cycle of thawing and refreezing to maintain the stability of the compounds. The disc diffusion sensitivity test conditions replicated those described by

Bauer *et al*. Importantly, DMSO controls showed no evidence of inhibitory zones, confirming its inert nature in these assays. Streptomycin and fluconazole served as positive control agents for antibacterial and antifungal activity, respectively.

To determine the MIC for each compound, the protocols outlined in the 1997 NCCLS guidelines were followed. Overnight subcultures of all bacterial isolates were grown at 37 °C in an incubator, while fungal isolates were incubated at 35 °C for 24-48 hours. To ensure the accuracy of the results, all microbial strains underwent at least two purification processes to verify their viability and purity. The bacterial inoculum was added to each well of the microdilution trays, which contained nutrient agar, and the plates were incubated at the specified temperatures. Solutions of the newly synthesized compounds and reference drugs were prepared using serial two-fold dilutions in DMSO, with concentrations ranging from 15 µg/mL to 1000 µg/mL. After a 24-hour incubation period at 37 °C in a humidified environment, MIC endpoints were determined. The MIC is defined as the lowest concentration of the agent that completely inhibits visible microbial growth. Reference wells included controls containing DMSO, sterile microorganisms, and sterile growth medium.

3. RESULTS AND DISCUSSION:

Figure 1 provides a comprehensive illustration of the synthetic procedures employed to obtain the target compounds. The detailed schematic outlines each step of the synthetic pathway, highlighting the reagents and conditions used at each stage. The study primarily focused on investigating the potential interactions between substituted hydroxy benzaldehydes and pyrrolo[2,3-d]pyrimidinehydrazide (PPH). This investigation aimed to explore the reactivity and compatibility of these starting materials under various experimental conditions. The synthesized compounds, labeled as **2a** through **2j**, were thoroughly characterized, and their physicochemical properties are comprehensively summarized in **Table 1**. This table includes critical data such as melting points, solubility, molecular weights, and other relevant physicochemical parameters that are essential for understanding the behavior and potential applications of these compounds.

3.1.FT(IR) spectra:

We thoroughly investigated the binding interactions between pyrrolo[2,3-d]pyrimidinehydrazide (PPH) and bromo-, cyano-, and difluoro-substituted benzaldehydes by meticulously comparing the Fourier-transform infrared (FT-IR) spectra of the synthesized compounds with those of the free, unbound PPH. By analyzing the FT-IR spectra, we focused on several notable bands to study the effects of PPH vibrations on the modified bromo, cyano, and difluoro-substituted benzaldehydes. The absence of aldehyde (CHO) and amino (NH_2) stretching vibrations in the FT-IR spectra indicated that all synthesized compounds had matured and reacted as anticipated. Instead, we observed the emergence of a new, solid band in the 1475-1539 cm^{-1} region, which is characteristic of the azomethine (HC=NN) group, confirming the successful formation of the expected products.

Further analysis according to references **[22, 23]** revealed the presence of aromatic υ(NH) groups in the produced compounds, indicated by broad absorption bands between 3178 and 3290 cm^{-1}. Additionally, aldehydic (-CH=) stretching bands were mapped out for each compound in the 2815-3080 cm^{-1} region. The infrared spectra of compounds 2a-i displayed two prominent bands at 1581-1590 and 1436-1480 cm^{-1}, which were linked to the aromatic ring's >C=C groups. Several other bands were observed between 682 and 739 cm^{-1} and 1315 and 1333 cm^{-1}, which could be attributed to various ring structures, including aromatic (C-N), di- or trisubstituted benzene, or mono-substituted benzene.

Moreover, the FT-IR spectra of compounds **2d-f** showed a distinct peak in the 1969-2232 cm^{-1} range, associated with the -CN (cyano) group, confirming the presence of this functional group in the molecules. Additionally, another absorption band was noted in the FT-IR spectra of compounds **2g-i**, within the wavenumber range of 1355-1371 cm^{-1}, which could be attributed to the fluorine (-F) functional group, further confirming the successful incorporation of these substituents into the synthesized compounds.

Table 1: FT(IR) spectral data of compounds 2a-2i

Comp	NH- aromatic)	NH- aliphatic)	-CH=	(>C=C<)	>C=NN-	-N-N-	Di sub Benz ring	C-Br/C-F	-CN
2a	3290	3146	3080	1585/1450	1475	1070	682	652	-
2b	3287	3127	3080	1585/1450	1475	1070	682	652	-
2c	3189	3127	3047	1581/1449	1475	1070	682	653	-
2d	3283	3201	3063	1587/1447	1511	1040	712	-	1969
2e	3178	3110	2815	1582/1451	1526	1070	741	-	2237
2f	3293	3199	2884	1590/1455	1539	1039	739	-	2232
2g	3180	3084	2822	1588/1436	1512	1512	736	1371	-
2h	3180	3118	3021	1582/1480	1521	1521	731	1355	-
2i	3179	3117	3023	1582/1480	1523	1523	731	1356	-

3.2.^{1}H NMR spectra:

In all of the synthesized compounds, the ^{1}H NMR spectra exhibited singlet signals with chemical shifts ranging from 11.69 to 14.39 ppm, which correspond to the aromatic NH group on the pyrrolyl ring. Additionally, within the chemical shift range of 8.71-13.01 ppm, a singl*et al*iphatic -NH peak was detected. Furthermore, in the 8.66-9.36 ppm range of the ^{1}H NMR spectra of all synthesized compounds, a singlet peak corresponding to the aldehydic -CH= group was observed. Notably, the absence of a broad singlet peak at 9.84 ppm (2H) in the synthesized derivatives indicates the successful substitution of the amino group with the Schiff base **[24]**, as this peak corresponds to the -NH_2 group of PPH.

Compounds **2a–i** displayed a single peak in their ^{1}H NMR spectra, corresponding to the pyrimidine proton, in the chemical shift range of 8.17-8.74 ppm. These observed ^{1}H NMR signals are consistent with those reported in previous literature sources **[24–25]**, confirming the structural integrity and expected chemical modifications of the synthesized compounds.

Table 2: NMR spectral data of compounds 2a-2j

Comp	-NH- aliphatic (s, 1H)	NH aromatic (s, 1H)	-CH= (s, 1H)	pyrimidine-H (s, 1H)	aromatic-H (m, 6H)
2a	14.35	13.01	8.69	8.50	7.24-8.38
2b	12.35	13.95	8.65	8.45	7.21-8.28
2c	11.69	11.42	9.36	8.17	6.86-8.04
2d	11.69	11.27	9.36	8.05	6.87-8.05
2e	14.39	13.08	8.97	8.74	7.21-8.56
2f	14.40	12.97	8.58	8.57	7.18-8.47
2g	14.35	13.01	8.69	8.54	7.24-8.37
2h	12.96	8.71	8.66	8.45	7.21-8.28
2i	12.90	11.30	8.65	8.44	7.21-8.28

3.3.^{13}C NMR spectra:

In the ^{13}C-NMR spectra of compounds labeled as a through g, a noticeable singlet was observed at chemical shift values ranging from14.26 to 142.61 ppm. These singlets are attributed to the presence of methylene (-CH=) groups, respectively. Furthermore, in the synthesized compounds, the aromatic carbon atoms were detected at specific regions: between 149.05 and 150.18 ppm for C2, between 100.04 and 100.16 ppm for C3, between 148.32 and 148.87 ppm for C4, between 114.73 and 114.83 ppm for C5, and between 122.13 and 125.97 ppm for C6. Additionally, carbons within the pyrrolopyrimine structure were identified at chemical shifts of 103.12 to 103.22 ppm for C2, 110.01 to 110.98 ppm for C3, and 126.17 to 126.22 ppm for C4.

3.4.Mass spectra:

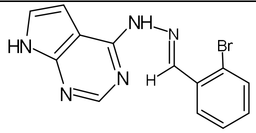
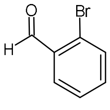
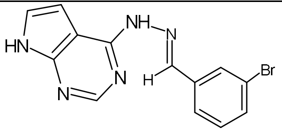
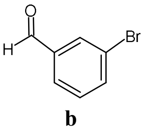
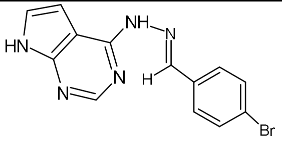
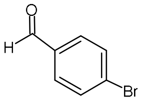
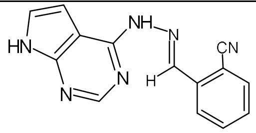
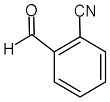
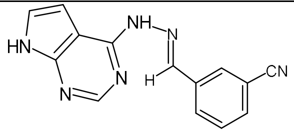
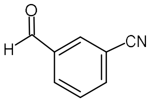
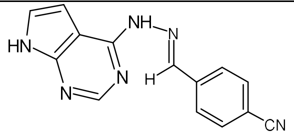
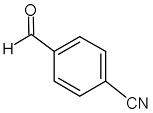
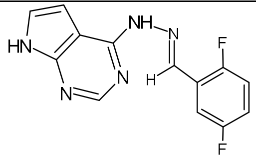
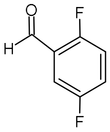
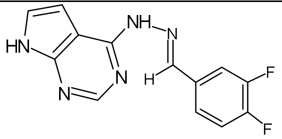
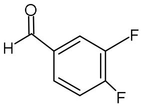
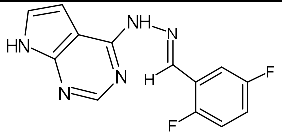
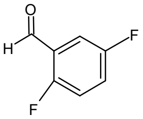
The mass spectra analysis of the novel bromo, difluoro, and cyano benzaldehyde derivatives of pyrrolo[2,3-d]pyrimidinehydrazide provided valuable insights into their molecular structures and fragmentation patterns. Mass spectrometry is a powerful analytical technique that ionizes molecules to determine their mass-to-charge ratios (m/z), elucidating the molecular weight and structural composition of compounds.

The mass spectra of the bromo-substituted benzaldehyde derivatives of pyrrolo[2,3-d]pyrimidinehydrazide revealed prominent peaks corresponding to molecular ions ([M]+) at m/z 315.01-318.09values consistent with the calculated molecular weights of the compounds. Fragmentation patterns indicated characteristic peaks arising from the loss of bromine atoms and other smaller fragments, providing evidence of the substitution pattern and confirming the presence of the bromo substituent.

In the difluoro-substituted benzaldehyde derivatives, the mass spectra exhibited molecular ion peaks ([M]+) at m/z 273.09-275.03 corresponding to the expected molecular weights. Fragmentation analysis revealed distinct peaks associated with the loss of fluorine atoms and other fragmentations, consistent with the structural modifications introduced by the difluoro substituents.

For the cyano-substituted benzaldehyde derivatives, the mass spectra displayed molecular ion peaks ([M]+) at m/z 262.10-264.38 indicative of the molecular weights calculated for each compound. Fragmentation patterns showed characteristic peaks associated with the loss of cyano groups and other fragmentations, confirming the incorporation of the cyano substituents into the pyrrolo[2,3-d]pyrimidinehydrazide framework.

Table 3: The yield, color, reaction duration, and physical properties of the product (2a-j)

Products	Aldehyde	Color	Yield (%)	Reaction time (min.)	m.p. (˚C)
 **2a**	 **a**	**Yellow**	**83.59**	**20**	**176**
 **2b**	 **b**	**White**	**86.99**	**30**	**179**
 **2c**	 **c**	**White**	**90.07**	**80**	**180**
 **2d**	 **d**	**White**	**77.70**	**60**	**193**
 **2e**	 **e**	**Yellow**	**79.12**	**30**	**194**
 **2f**	 **f**	**Yellow**	**81.93**	**70**	**188**
 **2g**	 **g**	**Yellow**	**84.09**	**45**	**178**
 **2h**	 **h**	**Yellow**	**90.14**	**40**	**180**
 **2i**	 **i**	**Yellow**	**86.66**	**50**	**173**

3.5.Antimicrobial evaluation:

In vitro screening was performed on the newly synthesized compounds to assess their antibacterial and antifungal characteristics. Their effectiveness against fungus and other Gram-positive and Gram-negative bacteria was evaluated using the broth microdilution method.

All bacterial isolates were cultured in a broth medium rich in nutrients after an incubation period of 24 hours at 37 °C. While incubating at 25 °C for 24 hours, the Sabouraud dextrose agar was moved to malt broth. Fungal spore suspensions were prepared using Tween 80 from actively growing fungi cultured for seven days. The bacteria and fungus exhibited final inoculum optical densities (OD) of 0.2–0.3 and 0.5, respectively. The dimethyl sulfoxide utilized for the preparation of the stock solutions does not exhibit any discernible impact on the microorganism under investigation at the amounts employed. At a concentration of 1000 g/mL, the populations of both bacteria and fungi exhibited a twofold increase. Fluconazole and streptomycin were frequently employed as therapeutic powders for the treatment of infections and fungus. The antibacterial activity was assessed by incubating the samples for 24 hours at 37°C, while the antifungal test involved incubation for 48 hours at 25°C.

3.5.1. Antibacterial activity:

The study utilized Streptomycin as a reference drug, known for its broad-spectrum antibiotic activity. It showed a MIC of 1 mg/mL against the bacterial species studied. The inhibitory regions for *Escherichia coli*, *Bacillus subtilis*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus* measured 15–21mm, 18-22 mm, 19–21 mm, and 12–23 mm, correspondingly. The antibacterial results in **Table 4** show that the tested compounds were effective against all the microorganisms studied, as evidenced by minimum inhibitory concentrations (MICs) ranging from 15 to 65 ppm.

Table 4: Antibacterial studies of 2a-jcompounds

Compound	**Zone of inhibition**			
	S. aureus	***B. subtilis***	***E. coli***	***P. aeruginosa***
(2a)	**19.00**	**20.00**	**19.00**	**19.00**
(2b)	**15.00**	**21.00**	**19.00**	**13.00**
(2c)	**19.00**	**22.00**	**21.00**	**21.00**
(2d)	**21.00**	**18.00**	**20.00**	**23.00**
(2e)	**20.00**	**19.00**	**20.00**	**22.00**
(2f)	**17.00**	**21.00**	**19.00**	**19.00**
(2g)	**16.00**	**22.00**	**19.00**	**12.00**
(2h)	**15.00**	**20.00**	**19.00**	**15.00**
(2i)	**19.00**	**21.00**	**20.00**	**15.00**
Streptomycin	**20.00**	**21.00**	**20.00**	**19.00**

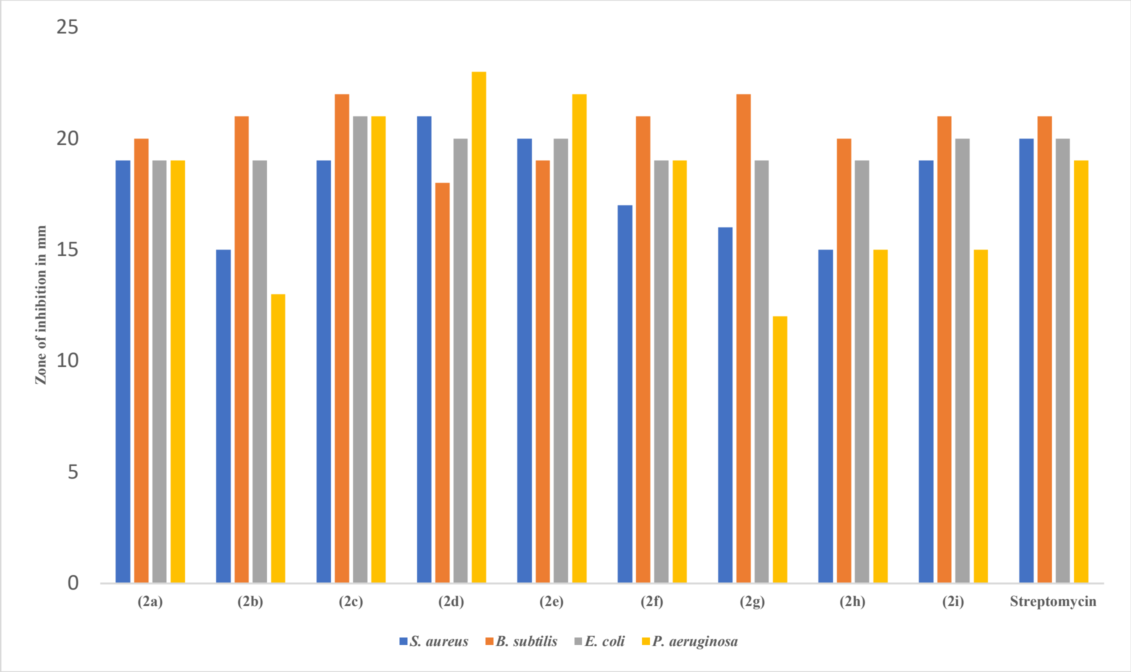


Figure 1: Antibacterial activity of prepared compounds

Concerning each bacterial species, it was observed that the **(2d)** (21mm) exhibited greater efficacy compared to the reference drug against *S. aureus*. Conversely, the (**2b**), (**2f**), (**2g**), (**2h**), and (**2i**) exhibited lower effectiveness against *P. aeruginosa*, while the (**2d**) displayed superior efficacy compared to the reference drug for *P. aeruginosa*. All prepared compounds had the highest level of activity against *E. coli*, while compounds (**2c**), (**2e**), and (**2g**) exhibited the greatest activity against *B. subtilis*. The observed antibacterial activity is likely attributable to the reduced lipophilicity of microbes' cell walls, facilitating their cellular entry. The probable reason for this phenomenon might be attributed to the molecule's lipophilic alkyl chain, allowing it to pass through the gram-negative bacteria's lipid cell membrane. The data points to an inverse relationship between carbon chain length and antibacterial activity. The molecule may have encountered steric resistance due to its large carbon chain, which prevented it from passing through the bacterial cell membrane **[26]**.

3.5.2. Antifungal activity

The studied fungi were inhibited by fluconazole, the standard drug, with a MICof 50 µg/ml.The inhibition zones observed for Candida albicans and Saccharomyces cerevisiae ranged from 17 to 23 mm and 16 to 25 mm, respectively. The compounds assessed in **Table 5** had significant fungicidal capability, surpassing the conventional therapy, as evidenced by their minimum inhibitory concentration (MIC) of 50 µg/mL against *Candida albicans* and *Saccharomyces cerevisiae*.

Table 5: Antifungal studies of 2a-jcompounds

Compound	*Candida albicans*	*Saccharomyces cerevisiae*
(2a)	17.00	19.00
(2b)	20.00	25.00
(2c)	23.00	24.00
(2d)	22.00	23.00
(2e)	29.00	24.00
(2f)	20.00	22.00
(2g)	19.00	23.00
(2h)	21.00	24.00
(2i)	17.00	25.00
Fluconazole	22.00	21.00

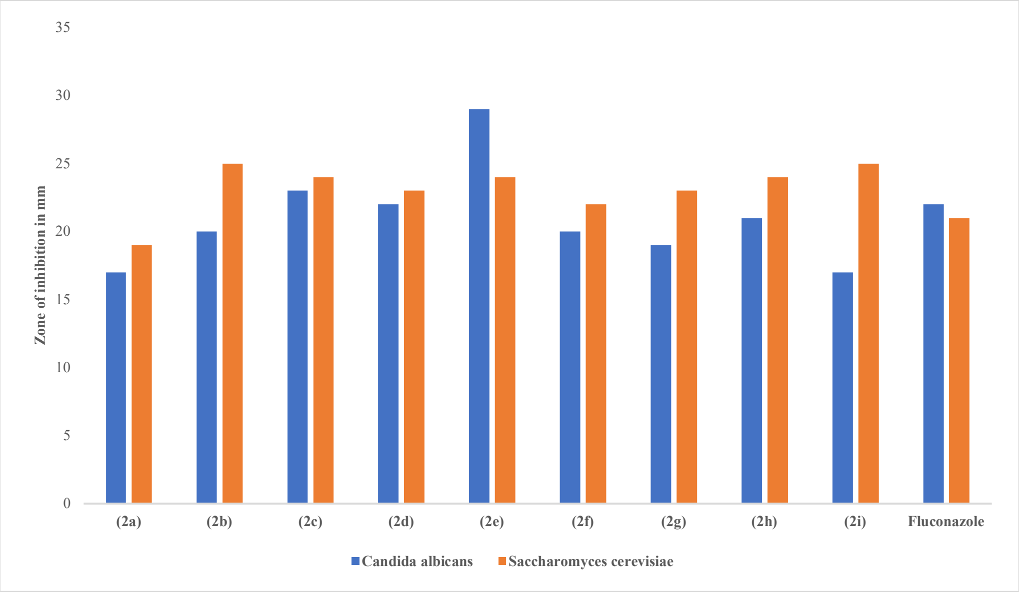


Figure 2: Antifungal activity of prepared compounds

4. CONCLUSION

The current investigation involved the synthesis of a set of novel substituted PPH ranging from 2a to j. The confirmation of the preparation of the proposed compounds is supported by a comprehensive analysis of analytical data, including FT-IR, UV-vis, and NMR spectrum examinations. The Bromo, cyano, and difluoro benzaldehydes-based compounds were developed and subjected to several spectroscopic techniques including ^{1}H NMR, UV-vis, elemental analysis, and FT-IR spectroscopy for characterization and analysis. The findings suggest that a combination of PPHand modified substituted benzaldehydes in a 1:1 ratio is recommended. All synthesized materials exhibited significant antibacterial efficacy.

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