

Synthesis, Characterization, and *in vitro* Antimicrobial Screening of Some "1H-Pyrazole's and 4H-Chromen-4-One's Derivatives with Penta- Fluoro-Benzoic Acid and 2,3,4,5-Tetra-Fluoro-Benzoic Acid"

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ABSTRACT

Background: The pyrazole nucleus has a five-membered heterocycle with two nitrogen atoms next to one another. The two nitrogen atoms are near in pyrazole nucleus heterocyclic compounds. Chromones are benzoannulated -pyrone-ringed heterocyclic compounds having a single oxygen ring. 4H-chromen-4-one **Materials and Methods:** All Chromones and Pyrazole derivatives were synthesized by conventional reflux method. 1-(4-chloro-2-hydroxy-5-methylphenyl)ethanone, penta fluoro benzoic acid, 2, 3, 4, 5-Tetrafluorobenzoic acid, Pyridine, Hydrazine Hydrate, Guanidine Hydrochloride, Ethanol, Con. Hydrochloric acid and Phosphorus oxychloride i.e. POCl₃ were used for the synthesis of Chromones and Pyrazole **Results:** Compared to Ciprofloxacin and Gentamycin, the anti-bacterial results for the substances or their derivatives like 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one (CC); 5-chloro-4-methyl-2-[5-(pentafluorophenyl)-1H-pyrazol-3-yl]phenol (CD); 5-chloro-2-[2-imino-6-(pentafluorophenyl)-1,2-dihydropyrimidin-4-yl]-4-methylphenol (CE); 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-(2,3,4,6-tetrafluorophenyl)propane-1,3-dione (CG); 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one (CH); 5-chloro-4-methyl-2-[5-(2,3,4,6-tetrafluorophenyl)-1H-pyrazol-3-yl]phenol(CI); 5-chloro-2-[2-imino-6-(2,3,4,6-tetrafluorophenyl)-1,2-dihydropyrimidin-4-yl]-4-methylphenol (CJ) against *S. aureus* and *Pseudomonas aeruginosa*. Moreover, the compounds code name like 2-acetyl-5-chloro-4-methylphenyl pentafluorobenzoate (CA); 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-(pentafluorophenyl) propane-1,3-dione: (CB); 2-acetyl-5-chloro-4-methylphenyl 2,3,4,6-tetrafluorobenzoate (CF); 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-(2,3,4,6-tetrafluorophenyl)propane-1,3-dione (CG) a derivatives gives potent anti-bacterial activity against *Escherichia coli*. **Conclusion:** The title compounds' and its derivatives' *in vitro* antibacterial activity against a few human pathogenic pathogens were investigated. Gram-positive Gram-negative *Pseudomonas aeruginosa* and *Escherichia coli*, along with *Staphylococcus aureus*, are the bacteria. Studies on the link between structure and activity have shown that compounds containing chromones and derivatives of the pyrazole had higher activity than those containing electron-donating groups.

Keywords: Ciprofloxacin and Gentamycin have antibacterial properties, As do 2,3,4,5-tetrafluorobenzoic acid and 1-(4-chloro-2-hydroxy-5-methylphenyl)ethanone.

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INTRODUCTION

In 1883, German scientist Ludwig Knorr made the pyrazole nucleus discovery. The nucleus and chromones were first discovered by Walther Flemming in 1882. The pyrazole nucleus has a five-membered heterocycle with two nitrogen atoms next to one another. Neighboring nitrogen atoms can be found

in heterocyclic compounds.^{1,2} Chromones are benzoannulated -pyrone-ringed heterocyclic compounds containing a single oxygen ring. 4H-chromen-4-one, also referred to as 4H-1-benzopyran-4-one, is the parent substance. Chromones and pyrazole include considerable amounts of heterocyclic nuclei because of the wide range of therapeutic applications for these elements. In Figure 1 below, chromones and pyrazole are shown. Today, chromones and pyrazole exhibit a variety of pharmacological properties.

Chromones have a molecular weight of 146.14 g/mol and the chemical formula C₉H₆O₂. Pyrazole has the chemical formula C₃H₄N₂.³ and a molecular weight of 68.07 g/mol. particularly,



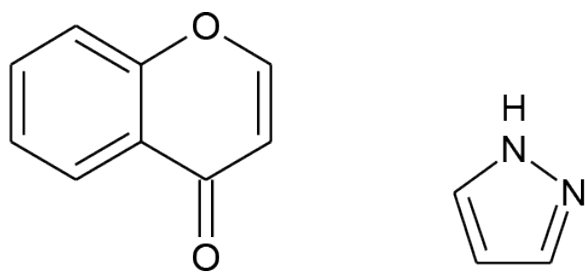
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4H-chromen-4-one **1H-pyrazole**

Figure 1: Chromones and Pyrazole heterocyclic nucleus.

the molecule with the chromones and pyrazole rings coupled to electron-withdrawing and electron-donating groups shown stronger inhibitory potential against bacterial and fungal strains than conventional medicine. Chromones and pyrazole both possess broad-spectrum anti-bacterial, anti-inflammatory, analgesic, anti-tubercular, antihypertensive, anticonvulsant, and antiviral activity, according to a conclusion drawn after reading multiple study and review articles.⁴ *Escherichia coli*, *Staphylococcus aureus*, and other parasitic or pathogenic micro-organisms have a marginally detrimental effect on human health. These microbial growths cause the host tissue to be destroyed and can cause fatal infections. Food poisoning, fever, and diarrhoea are symptoms of the bacterial parasites *Staphylococcus aureus* and *Escherichia coli*. Millions of people in poorer nations are also impacted. More than 80 million individuals worldwide are afflicted, and up to 1,30,000 people die from these illnesses each year. To treat these infections, a variety of medications, including Gentamycin, Amoxicillin, Norfloxacin, and Ciprofloxacin, are available on the market.^{5,6} They have substantial negative effects while being the most often recommended drugs for this bacterial disease. Based on these contributions, we will continue our drug research programme in order to produce interest in the synthesis of a new series of chromones and pyrazole to create more effective and toxic-free antimicrobial agents. This research will focus on the creation of new, safer, and more biologically active derivatives. Diverse biological processes have been associated with heterocyclic compounds comprising nitrogen and oxygen atoms. Chromones and pyrazole derivatives have a considerable impact on the medical industry thanks to their wide range of pharmacological actions.⁹⁻¹¹ such as their anti-bacterial, anti-viral, anti-diabetic, and anti-cancer activity. The information that implies that the substitution at the 2 and 5 positions considerably alters the pharmacological properties of these compounds has piqued the interest of numerous research teams in chromones and pyrazole. Particularly, compounds having both electron-withdrawing groups, such as chloro and fluoro connected with the Chromones ring and Pyrazole, showed more inhibitory ability against bacterial and fungal strains than conventional medicine. As a result, various research groups have worked to find novel antimicrobial medications and the WHO and new scientists around the world have made significant efforts to treat these illnesses. However,

pharmacologically, chromones and pyrazole, together with their derivatives, comprise the most significant class of organic heterocyclic compounds having anti-microbial capabilities, such as anti-bacterial, anti-fungal, herbicidal, and anti-viral activity.

MATERIALS AND METHODS

Materials

Chromones and pyrazole were synthesized by using phosphorus oxychloride, 1-(4-chloro-2-hydroxy-5-methylphenyl)ethanone, penta fluoro benzoic acid, 2, 3, 4, 5-Tetrafluorobenzoic acid, Pyridine, Hydrazine Hydrate, Guanidine Hydrochloride, Ethanol, Con. Hydrochloric acid. Chemicals of analytical grade were employed throughout. All chemicals were purchased from Modern Chemicals in Nashik and Atmaja Chemicals in Aurangabad.

Methods

All the chromones and pyrazole derivatives were prepared by the conventional reflux method. The melting points were calculated using the open tube capillary technique. Thin Layer Chromatography (TLC) plates were used to employ silica gel G in various solvent systems, such as chloroform: ethanol (6:4) and chloroform: methanol (8:2), and to test the purity of the chemicals. Using UV light and iodine vapors, the spots were located. Schemes 1 and 2 show the synthesis of these chromones and pyrazole derivatives. Figures 2 and 3.

EXPERIMENTAL WORK (Scheme I)

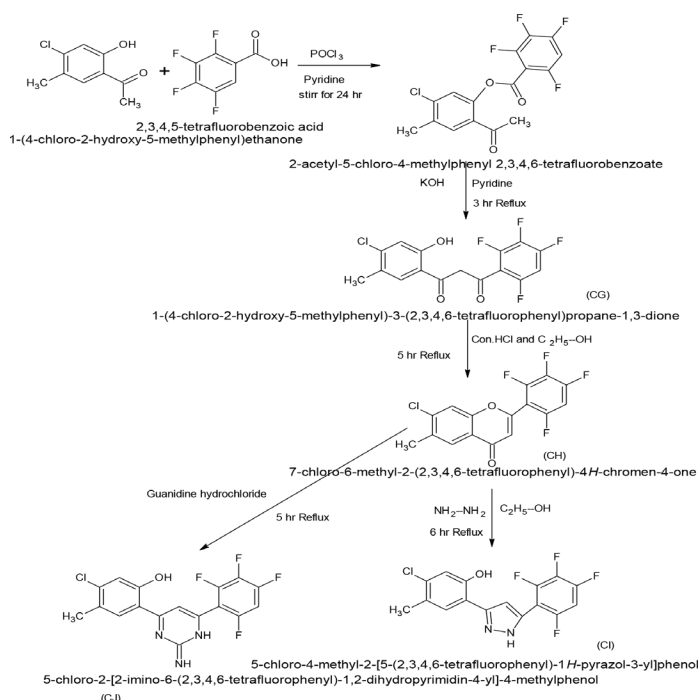


Figure 2: Scheme 1: Synthesis of 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one derivative's.

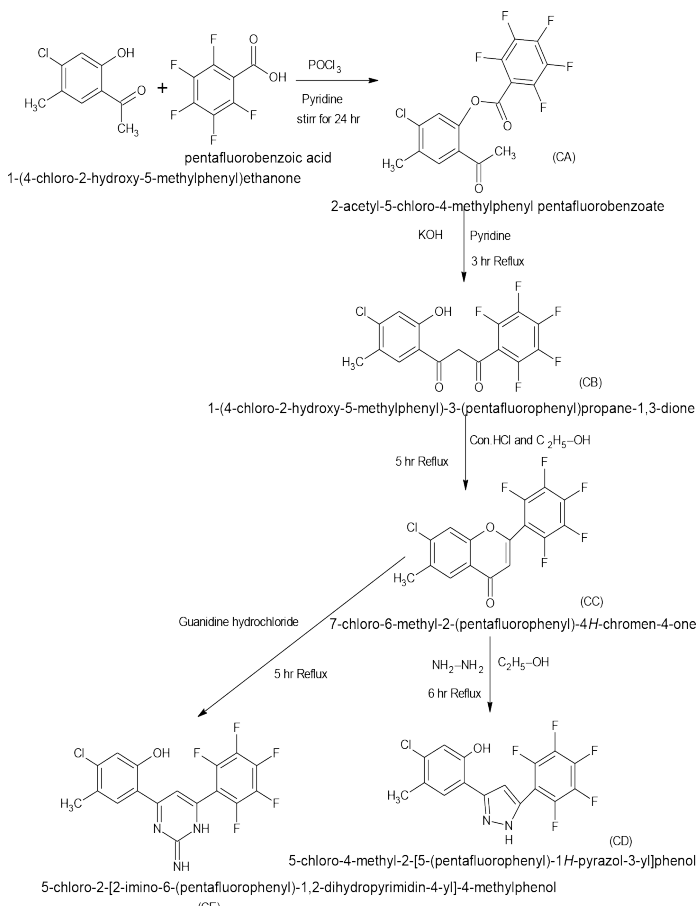
(Scheme II)

Figure 3: Scheme 2: 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one derivatives.

PROCEDURE**Synthesis of 2-acetyl-5-chloro-4-methylphenyl pentafluorobenzoate (CA): (Scheme 2)**

In the presence of POCl₃ (5 mL) and pyridine (15 mL), Pentafluorobenzoic acid (0.5 g) and 1-(4-chloro-2-hydroxy-5-methylphenyl)ethanone (0.5 g) interact with one another. After 20 to 24 hr of stirring on a magnetic stirrer, the reaction produces 2-acetyl-5-chloro-4-methylphenyl pentafluorobenzoate (CA), which solidifies when ice-cold water is added.

Synthesis of 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-(pentafluorophenyl) propane-1,3-dione (CB): (Scheme 2)

After 3 hr of refluxing a mixture of 2-acetyl-5-chloro-4-methylphenyl pentafluorobenzoate (CA), potassium hydroxide (0.5 g), and pyridine (5 mL), the reaction was finished. The completeness of the reaction was verified by TLC. The mixture was cooled by the ice addition. The precipitate formed when ethanol was re-crystallized and washed with water yields 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-(pentafluorophenyl) propane-1,3-dione (CB).

Synthesis of 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one (CC): (Scheme 2)

Using ordinary hydrochloric acid (5 mL), ethanol (5 mL), and 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-(pentafluorophenyl) propane-1,3-dione (CB), the reaction is carried out and left to reflux for 5 hr. TLC demonstrated that the reaction was over. The mixture was cooled by the ice addition. The precipitate that had developed became 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one (CC) after being washed with water and crystallised from ethanol.

Synthesis of 5-chloro-4-methyl-2-[5-(pentafluorophenyl)-1H-pyrazol-3-yl]phenol (CD): (Scheme 2)

The completion of the reaction was verified by TLC after a solution of 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one (CC) reacted with hydrazine hydrate (5 mL) and ethanol (10 mL) and refluxed for 6 hr. The addition of ice chilled the mixture. The result of recrystallizing the precipitate from ethanol after it had been rinsed with water is 5-chloro-4-methyl-2-[5-(pentafluorophenyl)-1H-pyrazol-3-yl] phenol (CD).

Synthesis of 5-chloro-2-[2-imino-6-(pentafluorophenyl)-1,2-dihydropyrimidin-4-yl]-4-methylphenol (CE): (Scheme 2)

A solution of 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one (CC) react with guanidine hydrochloride (5 mL) in the presences of acid and it was reflux for 5 hr and then Completion of the reaction were confirmed by TLC. The addition of ice chilled the mixture. After being recrystallized from ethanol, the precipitate was washed with water and produced 5-chloro-2-[2-imino-6-(pentafluorophenyl)-1,2-dihydropyrimidin-4-yl]-4-methylphenol.

Synthesis of 2-acetyl-5-chloro-4-methylphenyl 2,3,4,6-tetrafluorobenzoate (CF): (Scheme 1)

In the presence of POCl₃ (5 mL) and pyridine (15 mL), a mixture of 1-(4-chloro-2-hydroxy-5-methylphenyl)ethanone and 2,3,4,5-tetrafluorobenzoic acid reacts to form 2-acetyl-5-chloro-4-methylphenyl 2,3,4,6-tetrafluorobenzoate (CF), which is a solid after the addition of ice cold water.

Synthesis of 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-(2,3,4,6-tetrafluorophenyl)propane-1,3-dione (CG): (Scheme 1)

The completion of the reaction was verified by TLC after a solution of 2-acetyl-5-chloro-4-methylphenyl 2,3,4,6-tetrafluorobenzoate (CF) react with potassium hydroxide (0.5 g) and pyridine (5 mL) and reflux for 5 h. The addition of ice chilled the mixture. The result of recrystallizing

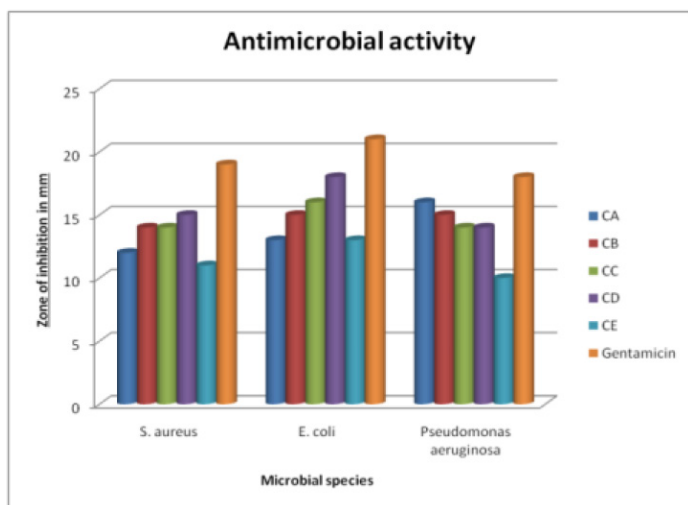


Figure 4: Antibacterial Activity of CA to CE With Gentamicin.

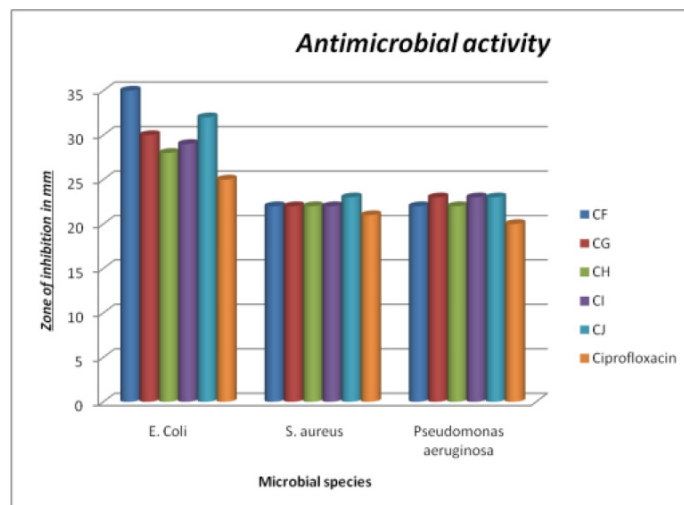


Figure 7: Antibacterial Activity of CF to CJ With Ciprofloxacin.

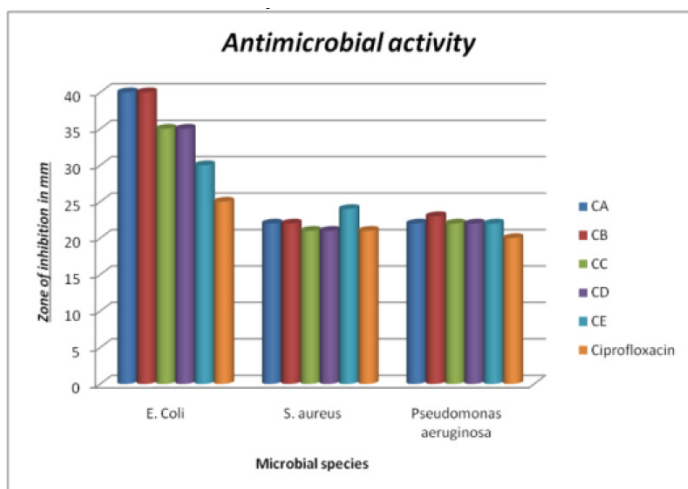


Figure 5: Antibacterial Activity of CA to CE With Ciprofloxacin.

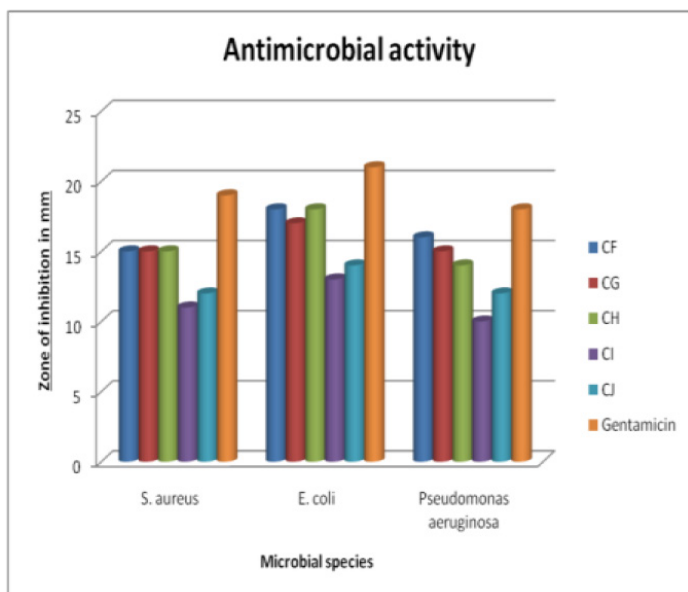


Figure 6: Antibacterial Activity of CF to CJ With Gentamicin.

the precipitate from ethanol after it had been rinsed with water is 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-(2,3,4,6-tetrafluorophenyl)propane-1,3-dione (CG).

Synthesis of 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one (CH): (Scheme 1)

The completion of the reaction was verified by TLC after a solution of 1-(5-chloro-2-hydroxy-4-methylphenyl)-3-(pentafluorophenyl)propane-1,3-dione (CB) was refluxed for 6 hr with con. hydrochloric acid (5 mL) and ethanol (5 mL). The addition of ice chilled the mixture. The ethanol was recrystallized from the precipitate after it had been washed with water, producing 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-Chromone-4H-one (CH).

Synthesis of 5-chloro-4-methyl-2-[5-(2,3,4,6-tetrafluorophenyl)-1H-pyrazol-3-yl]phenol (CI): (Scheme 1)

The completion of the reaction was verified by TLC after a solution of 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one (CH) reacted with hydrazine hydrate (5 mL) and ethanol (10 mL) and refluxed for 10 hr. The addition of ice chilled the mixture. When the precipitate is recrystallized from ethanol after being rinsed with water, it produces 5-chloro-4-methyl-2-[5-(2,3,4,6-tetrafluorophenyl)-1H-pyrazol-3-yl]phenol (CI).

Synthesis of 5-chloro-2-[2-imino-6-(2,3,4,6-tetrafluorophenyl)-1,2-dihydropyrimidin-4-yl]-4-methylphenol (CJ): (Scheme 1)

The completion of the reaction was verified by TLC after guanidine hydrochloride (5 mL) and a solution of 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one (CH) were refluxed for 10 hr. The addition of ice chilled the mixture. After being recrystallized from ethanol, the precipitate was

washed with water and produced 5-chloro-2-[2-imino-6-(2,3,4,6-tetrafluorophenyl)-1,2-dihydropyrimidin-4-yl]-4-methylphenol (CJ).

Characterization

We prepared 6 derivatives of 4H-chromen-4-one and 1H-pyrazol which were listed in below Table 1.

Physical characteristic like TLC, R_f value, M.P. and % yield of 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one derivatives and 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one derivative's were listed in above Table 2.

Spectral Data

Synthesis of 2-acetyl-5-chloro-4-methylphenyl pentafluorobenzoate (CA): (Scheme 2)

FTIR (KBr) ν cm^{-1} :3062 (Ar C-H stretching), 1641 (Ar C=C), 797 (Ar C=C stretching), 1172 (Ar C-F stretching), 796 (Ar C-Cl stretching),1730 (Ester C=O stretching), 1366 (C-O stretching); ^1H NMR (400 MHz CDCl_3 δ ppm): 2.34 (s, 3H, CH_3 , $J = 13.7$ Hz), 7.44-7.81 Ar C-H (m, 2H, $J = 15.9$ Hz), 2.50 (s, 3H, CH_3 , $J = 11.9$ Hz); JEOL GCMATE II MS (m/z): 365(M+), 366 (M++1) Mol. Wt.:366.

Synthesis of 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-(pentafluorophenyl) propane-1,3-dione (CB): (Scheme 2)

FTIR (KBr) ν cm^{-1} :3061 (Ar C-H stretching), 1568 (Ar C=C stretching), 884 (Ar C-H stretching), 1171 (Ar C-F stretching), 723 (Ar C-Cl stretching), 1641 (Aryl Ketone C=O stretching), 1250 (C-O stretching), 3573(O-H stretching); ^1H NMR (400 MHz CDCl_3 δ ppm): 3.81 C-H (s, 2H CH_2), 5.35 O-H (s, 1H, O-H), 2.34 (s, 3H, CH_3), 7.02-7.57 Ar C-H, (m, 3H Ar); JEOL GCMATE II GC-MS (m/z): 377(M+), 378 (M++1) Mol. Wt.:366.

Synthesis of 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one (CC): (Scheme 2)

FTIR (KBr) ν cm^{-1} :3028 (Ar C-H stretching), 1529 (Ar C=C stretching), 846 (Ar C-H stretching), 1108 (Ar C-F stretching), 769 (Ar C-Cl stretching), 1668 (Aryl Ketone C=O stretching), 1350 (C-O stretching); ^1H NMR(400 MHz CDCl_3 δ ppm):6.54 Ar C-H (s, 2H CH_2), 2.34 C-H (s, 3H, CH_3), 7.10-7.52 Ar C-H (m, 3H Ar); JEOL GCMATE II MS (m/z): 359(M+), 360 (M++1). Mol. Wt.:360.

Synthesis of 5-chloro-4-methyl-2-[5-(pentafluorophenyl)-1H-pyrazol-3-yl] phenol (CD): (Scheme 2)

FTIR (KBr) ν cm^{-1} 3025 (Ar C-H stretching), 1645 (Ar C=C stretching), 755 (Ar C-H stretching), 1273 (Ar C-F), 652 (Ar C-Cl

stretching),3535 (Ar O-H stretching), 1378 (C-O stretching), 3477 (Ar N-H stretching); ^1H NMR (400 MHz CDCl_3 δ ppm): 6.97-7.72 Ar C-H (m, 3H Ar), 2.34 C-H (s, 3H, CH_3), 5.35 O-H (s, 1H, O-H), 6.81 C-H (s, 2H CH_2),12.62 N-H (s, 1H); JEOL GCMATE II MS (m/z): 373(M+), 374 (M++1). Mol. Wt.:374.

Synthesis of 5-chloro-2-[2-imino-6-(pentafluorophenyl)-1,2-dihydropyrimidin-4-yl]-4-methylphenol (CE): (Scheme 2)

FTIR (KBr) ν cm^{-1} :3025 (Ar C-H stretching), 1631 (Ar C=C stretching), 817 (Ar C-H stretching), 1118 (Ar C-F stretching), 752 (Ar C-Cl stretching), 3385 (Ar O-H stretching), 1319 (C-O stretching), 3364 (N-H stretching); ^1H NMR (400 MHz CDCl_3 δ ppm): 6.92-7.62 (m, 2H Ar), 2.34 C-H (s, 3H, CH_3), 5.35 O-H (s, 1H, O-H), 6.31 C-H (s, 2H CH_2), 13.86 N-H (s, 1H), 13.76 N-H (s, 1H); JEOL GCMATE II GC-MS (m/z):394 (M+), 394 (M++1). Mol. W

Synthesis of 2-acetyl-5-chloro-4-methylphenyl 2,3,4,6-tetrafluorobenzoate (CF): (Scheme 1)

FTIR (KBr) ν cm^{-1} :3036 (Ar C-H), 1592 (Ar C=C stretching), 747 (Ar C-H stretching), 1115 (Ar C-F stretching), 796 (Ar C-Cl stretching), 1750 (Ester C=O stretching), 1334 (C-O stretching); ^1H NMR (400 MHz CDCl_3 δ ppm): 2.34 C-H (s, 3H, CH_3), 7.44-7.81 Ar C-H (m, 2H Ar), 2.50 C-H (s, 3H, CH_3), 7.6 Ar C-H (m, 1H Ar); JEOL GCMATE II GC-MS (m/z):359(M+), 360 (M++1) Mol. Wt.:360.

Synthesis of 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-(2,3,4,6-tetrafluorophenyl)propane-1,3-dione (CG): (Scheme 1)

FTIR (KBr) ν cm^{-1} :1568 (Ar C=C stretching), 884 (Ar C-H stretching), 1171(Ar C-F stretching), 723 (Ar C-Cl stretching), 1641 (Aryl Ketone C=O stretching), 1250 (C-O stretching), 3573 (Ar O-H stretching); ^1H NMR (400 MHz CDCl_3 δ ppm): 3.81 C-H (s, 2H CH_2), 5.35 O-H (s, 1H, O-H), 2.34 C-H (s, 3H, CH_3), 7.02-7.57 Ar C-H (m, 3H Ar), 7.4 Ar C-H (m, 1H Ar); JEOL GCMATE II GC-MS (m/z):358 (M+), 359 (M++1) Mol. Wt.:359.

Synthesis of 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one (CH): (Scheme 1)

FTIR (KBr) ν cm^{-1} :3029 (Ar C-H stretching), 1607 (Ar C=C stretching), 846 (Ar C-H stretching), 1158 (Ar C-F stretching), 688 (Ar C-Cl stretching), 1719 (Aryl Ketone C=O stretching), 1349 (C-O stretching); ^1H NMR (400 MHz CDCl_3 δ ppm):6.54 Ar C-H (s, 1H), 6.8 Ar C-H (m, 1H), 2.34 (s, 3H, CH_3), 7.10-7.52 Ar C-H (m, 2H); JEOL GCMATE II GC-MS (m/z):341(M+), 342 (M++1). Mol. Wt.:342.

Synthesis of 5-chloro-4-methyl-2-[5-(2,3,4,6-tetrafluorophenyl)-1H-pyrazol-3-yl]phenol (CI): (Scheme 1)

FTIR (KBr) ν cm^{-1} : 3073 (Ar C-H stretching), 1645 (Ar C=C stretching), 755 (Ar C-H stretching), 1312 (Ar C-F stretching), 688 (Ar C-Cl stretching), 3565 (Ar O-H stretching), 1378 (C-O stretching), 3383 (N-H stretching); ^1H NMR (400 MHz CDCl_3 δ ppm): 6.97-7.72 Ar C-H (m, 2H), 2.34 C-H (s, 3H, CH_3), 6.8 C-H (m, 1H Ar), 5.35 O-H (s, 1H, O-H), 6.81 C-H (s, 2H CH_2), 12.62 N-H (s, 1H); JEOL GCMATE II GC-MS (m/z): 355 (M+), 356 (M++1). Mol. Wt.: 356.

Synthesis of 5-chloro-2-[2-imino-6-(2,3,4,6-tetrafluorophenyl)-1,2-dihydropyrimidin-4-yl]-4-methylphenol (CJ): (Scheme 1)

FTIR (KBr) ν cm^{-1} : 3025 (Ar C-H stretching), 1631 (Ar C=C stretching), 817 (Ar C-H stretching), 1155 (Ar C-F stretching), 755 (Ar C-Cl stretching), 3591 (Ar O-H stretching), 1319 (C-O stretching), 3363 (N-H stretching); ^1H NMR (400 MHz CDCl_3 δ ppm): 6.92-7.62 Ar C-H (m, 2H), 2.34 C-H (s, 3H, CH_3), 6.8 Ar C-H (m, 1H Ar), 5.35 O-H (s, 1H, O-H), 6.31 C-H (s, 2H CH_2), 13.86 N-H (s, 1H), 13.76 N-H (s, 1H); JEOL GCMATE II GC-MS (m/z): 394 (M+), 395 (M++1). Mol. Wt.: 395

Biological evaluation

In vitro Antimicrobial Activity by Agar Well Diffusion Method

To evaluate the antibacterial activity of diverse drugs, the agar well diffusion method was employed. Mueller Hinton Agar (MHA) was prepared in accordance with the manufacturer's instructions, and 100 L of the bacterial solution were dispersed uniformly across the surface of the agar plates. The inhibitory zone's millimeter-scale diameter was measured, and the results were recorded. The Minimum Inhibitory Concentration (MIC) was determined using the agar dilution method.¹¹ The plates next underwent a 24-hr period of 37°C bacterial incubation. The lowest concentration of the test material that completely eliminated the microorganism's capacity to grow visibly was determined to be the MIC. Each experiment was conducted in an aseptic setting. The compounds CA to CE and CF to CJ's *in vitro* antibacterial effectiveness against a number of pathogens, including gram-positive *Staphylococcus aureus*, gram-negative *Escherichia coli* and *S* were examined *in vitro* using the disc diffusion method and Mueller Hinton Agar (M173) media. Tables 3, 4, 5 and 6 below indicate the *in vitro* antimicrobial activity of 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one (CH) and 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one (CC) derivatives against Gentamycin and Ciprofloxacin.

Table 1: Structure of 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one derivatives and 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one derivatives.

Label	Structure
CC	
CD	
CE	
CH	
CI	
CJ	

RESULTS AND DISCUSSION

The results showed that most synthetic compounds in varied degrees inhibited the studied bacteria. In general, compared to some gram-negative bacteria, some gram-positive bacteria investigated exhibit greater inhibition from inhibitory activity. In addition, substances having code names like 5-chloro-4-methyl-2-[5-(pentafluorophenyl)-1H-pyrazol-3-yl]; 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one (CC); 5-Chloro-2-[2-imino-6-(pentafluorophenyl)-1,2-dihydropyrimidin-4-yl]phenol (CD); 4-methylphenol (CE), 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-

Table 2: TLC, R_f value, M.P. and % yield of 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one derivatives and 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one derivative's







Comp. Name	TLC image	R _f value	M.P (°C)	% Yields	Comp. Name	TLC image	R _f value	M.P (°C)	% Yields
CC		0.93	188-192	78	CH		0.73	168-172	71
CD		0.85	187-191	69	CI		0.79	178-182	94
CE		0.78	183-186	81	CJ		0.68	176-179	86

Table 3: In vitro Antimicrobial Activity of 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one (CH) derivatives against Gentamycin.

Compound No.	Diameter of zone of inhibition (mm)		
	<i>Escherichia coli</i>	<i>Staphylococcus aureus</i>	<i>Pseudomonas aeruginosa</i>
	ATCC 25922	ATCC 25923	ATCC 27853
CF	12	17	09
CG	10	11	10
CH	13	20	20
CI	15	24	21
CJ	16	21	19
Gentamycin	20	36	28

Table 4: In vitro Antimicrobial Activity of 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one (CH) derivatives against Ciprofloxacin.

Compound No.	Diameter of zone of inhibition (mm)		
	<i>Escherichia coli</i>	<i>Staphylococcus aureus</i>	<i>Pseudomonas aeruginosa</i>
	ATCC 25922	ATCC 25923	ATCC 27853
CF	14	24	14
CG	12	21	17
CH	13	20	20
CI	15	24	21
CJ	16	21	17
Ciprofloxacin	20	36	28

Table 5: In vitro Antimicrobial Activity of 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one (CC) against Gentamycin

Compound No.	Diameter of zone of inhibition (mm)		
	<i>Escherichia coli</i>	<i>Staphylococcus aureus</i>	<i>Pseudomonas aeruginosa</i>
	ATCC 25922	ATCC 25923	ATCC 27853
CA	12	18	10
CB	08	15	09
CC	13	20	19
CD	14	22	20
CE	14	21	21
Gentamycin	20	36	28

Table 6: In vitro Antimicrobial Activity of 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one (CC) against Ciprofloxacin.

Compound No.	Diameter of zone of inhibition (mm)		
	<i>Escherichia coli</i>	<i>Staphylococcus aureus</i>	<i>Pseudomonas aeruginosa</i>
	ATCC 25922	ATCC 25923	ATCC 27853
CA	18	18	16
CB	16	15	24
CC	15	20	19
CD	14	22	20
CE	14	21	21
Ciprofloxacin	20	36	28

(2,3,4,6-tetrafluorophenyl)propane-1,3-dione (CG), 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one (CH), 5-chloro-4-methyl-2-[5-(2,3,4,6-te against *S. aureus* and *Pseudomonas aeruginosa* using 4-methylphenol (CJ). Moreover, the compounds code name like 2-acetyl-5-chloro-4-methylphenyl pentafluorobenzoate (CA); 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-(pentafluorophenyl)propane-1,3-dione: (CB); 2-acetyl-5-chloro-4-methylphenyl 2,3,4,6-tetrafluorobenzoate (CF); 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-(2,3,4,6-tetrafluorophenyl)propane-1,3-dione (CG) against *Escherichia coli*. The antibacterial activity was marginally boosted as a result of the oxygen atom being swapped out for a nitrogen atom. Our research revealed that all the compounds had a stronger antibacterial effect against Gram-positive bacteria than they did against Gram-negative bacteria. The novel synthesised compounds 5-chloro-4-methyl-2-[5-(pentafluorophenyl)-1H-pyrazol-3-yl] phenol (CD) and 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one (CC) both demonstrated good, considerable antimicrobial activity against Ciprofloxacin and Gentamycin. Tables 3 to 6 display the findings of preliminary antimicrobial testing of the produced compounds using common broad-spectrum antibacterial medications like Ciprofloxacin and Gentamycin. Zones of Inhibition of 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one (CC) and 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one (CC) were tested for the ability of the synthesized compounds to inhibit bacteria. The effects of -4H chromen-4-one (CH) against antibiotics like Ciprofloxacin and Gentamycin are depicted in Figures 4-7.

CONCLUSION

Ciprofloxacin and Gentamycin, the usual reference substance for this investigation, showed the best activity against all tested germs at all tested levels. At a dosage of 100 g/mL, both Ciprofloxacin and Gentamycin demonstrated moderate to strong activity against all three bacteria in addition to inhibitory zones. The activity of these drugs, however, is lower than that of the widely used reference substance, ciprofloxacin. More study is needed to comprehend the mechanism of action and potential clinical applications of these compounds. Furthermore, substances with code names like 7-chloro-6-methyl-2-(pentafluorophenyl)-4H-chromen-4-one (CC); 5-chloro-4-methyl-2-[5-(pentafluorophenyl)-1H-pyrazol-3-yl] phenol (CD); 5-chloro-2-[2-imino-6-(pentafluorophenyl)-1,2-dihydropyrimidin-4-yl]-4-methylphenol (CE); 1-(4-chloro-2-hydroxy-5-methylphenyl)-3-(2,3,4,6-tetrafluorophenyl)propane-1,3-dione (CG); 7-chloro-6-methyl-2-(2,3,4,6-tetrafluorophenyl)-4H-chromen-4-one (CH); 5-chloro-4-methyl-2-[5-(2,3,4,6-tetrafluorophenyl)-1H-pyrazol-3-yl]phenol (CI); 5-chloro-2-[2-imino-6-(2,3,4,6-tetrafluorophenyl)-1,2-dihydropyrimidin-4-yl]-4-methylphenol (CJ) against *S. aureus* and *Pseudomonas aeruginosa*.

uorophenyl)-1H-pyrazol-3-yl]phenol(CI); 5-chloro-2-[2-imino-6-(2,3,4,6-tetrafluorophenyl)-1,2-dihydropyrimidin-4-yl]-4-methylphenol (CJ) against *S. aureus* and *Pseudomonas aeruginosa*.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

ABBREVIATIONS

FTIR: Fourier transform infrared spectroscopy; **NMR spectroscopy:** Nuclear magnetic spectroscopy; **MS:** Mass spectroscopy; **KBr:** Potassium Bromide; **% yield:** Percentage yields; **M.P.:** Melting point; **mg/kg:** Milligram/ kilograms; **sec:** seconds; **δ:** Chemical shift; **Mol. Wt:** Molecular Weight; **gm:** Gram.

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