PART-IV

Antimicrobial activities of Coumaran-3-ones, 3-Ary-[1]-Benzofurano [3,2-C]-1-Substituted Pyrazoles and 3-(Chloroaryl)-5-Aryl-1-Substituted Pyrazoles

<table>
<thead>
<tr>
<th>Chapter</th>
<th>Title</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Introduction</td>
<td>197</td>
</tr>
<tr>
<td>II</td>
<td>Origin of the problem</td>
<td>210</td>
</tr>
<tr>
<td>III</td>
<td>Summary of the Work</td>
<td>213</td>
</tr>
<tr>
<td>IV</td>
<td>Experimental and Discussion of the Result</td>
<td>220</td>
</tr>
</tbody>
</table>
Chapter 1
INTRODUCTION

Antimicrobial activities of 2-arylcoumaran-3-ones, 3-aryl-[1]
Benzofurano[3, 2-c]-1-substituted pyrazoles and 3-(Chloroaryl)-
5-aryl-1-substituted pyrazoles:

Heterocyclic compounds have gained immense importance in
human life because of their variety of applications, particularly these
compounds acquired medicinal importance and have been
successfully tested against several diseases.

The history of drugs is almost as old as the history of man, but
medicine really began with the Greeks who were well aware of the
value of Opium and Hippocrates (400 B.C.) has justly been termed
‘The Father of Medicine’. It is interesting that even primitive people
could discover relationship between drug and disease. The use of drugs
has been so prevalent throughout history that Sir William Osler stated
(1894) with some justification that, ‘Man has an inborn craving for
medicine’.

When a living system is in imbalance through distortion of
normal bodily processes a pathologic condition is present, it is then
that the chemical stimulus, the pharmacologic agent, the drug, is
administered to attempt to re-establish the organism to a normal
physiologic and chemical balance. A drug is administered to alleviate
symptoms and possibly cure the diseased condition.

Modern medicine is considered since ‘Hippocrates’ a Greek
Physician (400 B.C.) who introduced for the first time, the concept
of disease as a pathologic process and organized the science of medicine on the basis of observations, analysis and deduction.

Chemical agents not only provide the structural basis and energy supply of living organisms but also regulate their functional activities. The interaction between potent chemicals and living system contribute to the understanding of life processes and provide effective methods for the treatment, prevention and diagnosis of many diseases. Chemical compounds used for this purpose are called 'drug' and their action on living system are referred to as 'drug effect'. The subject of drug is as old as disease. Fighting diseases with drug is the timeless struggle. Man’s survival on this planet is dependent upon it’s success. Sickness has been man’s heritage from the beginning of his existence and search for remedies to contact is perhaps equally old.

The action of drug gives an insight on living organisms and isolated tissues. Knowledge of the mode of action of drugs, it’s effects on various body systems and the probable adverse effect is important. From early times, antibacterial Agents have been used as remedies. The earliest source of medicine came from ‘Egypt’ and two kingdoms of ‘Assyria’ and ‘Babylonia’. ‘Papyri’ was the first written account of medical experiences from Egypt and date back to 1900 B.C. Papyrus was discovered by Elber in 1872 and was prepared in 1500 B.C. and mentions about 700 herbal medicines including ‘Opium’.

Till the beginning of 19th century the treatment of disease consisted of obnoxious remedies such as flesh, excreta and metallic
and plant preparations. James Gregory (1753-1821) was responsible for popularizing heroic symptomatic treatment consisting of blood letting, large doses of emetics and drastic purgatives often with disastrous results. Such type of treatment without any rational basis was called 'Allopathy'.

In the early 19th century the concept of 'Homeopathy' was first introduced by 'Hanneman' and thought that 'Like Cures Like' and dilution potentiates the action of drugs. Homeopathy outlines the therapy for various ailments with drugs in very high dilution. Treatment of diseases with chemical substances has been known since the 1500's. The chemical substances used for the treatment of infectious diseases and diseases caused by the proliferation of malignant cells are called as chemotherapeutic agents.

It is very important to know specific mechanism by which chemotherapeutic agents inhibit or kills micro-organisms. This information has wide applications. It is conductive to intelligent use of drug. It may suggest some chemical entity as a superior drug, e.g. similar compound but with some modification in it’s configuration, it provides a better understanding of the cells. Chemotherapeutic agents are intensively investigated in order to establish their mode of action.

Pasteur and Koch established that, micro-organisms were the cause of infectious diseases. First time Paul Ehrlich was proposed that infectious disease may be cured by using chemical that kill the
infecting agent but do not harm the host at the concentration employed. He discovered 'Salvarson' which was active against the causative organism of 'Syphilis'. He used the term 'Chemotherapy'. According to him, cells possess chemical receptors to which the drug binds. He recognized the importance of quantitative measurement to determine the drug dose, which would be effective against the causative agent and would not have toxic effect on the host. He gave a number of methods for screening a large number of compounds for biological activity in relation to chemical structure. Different drugs were then synthesized and tested to see whether they have improved antibacterial activity and reduced toxicity.

Any chemical substance inhibiting the growth or causing the death of micro-organisms is known as antibacterial agents. Although a wide range of chemicals have these properties. It is possible to subdivide antimicrobial agents into the various groups according to the action and purposes for which they are employed. Agents acting on the bacteria are called bacteriostatic or bacteriocidal, those acting on fungi are called fungistatic or fungicidal. Antibacterial agents include disinfectants and antimicrobial drugs.

With the development of a wide variety of antibiotic active against the whole spectrum of pathogenic bacteria and effective vaccines against most viral diseases, expectations were raised about the eventual elimination of all infectious disease. However, such hopes were dampened when new infectious diseases began to appear
caused by ditherto unknown micro-organisms or by known microbes producing novel manifestation. In 1981 when AIDS was identified in USA and began it’s pandemic spread. Unceasing vigilance appears essential to protect man from microbes.

The world’s oldest pharmacological writing came from India and China. The great herbal of Chinese ‘Material Medica’, ‘Pan Tsao’ was probably written in 2735 B.C. The earliest Indian records are ‘Vedas’. Although there are medical descriptions in ‘Rigveda’ (2500-3000 B.C.), it was charak renowned ancient Indian physician and later Sushruta and Vagbhatt, who discovered various medicines, included in ‘Ayurveda’ the science of life. Initially it consisted mostly nonpoisonous vegetable drugs and minerals. Charak described about 300 vegetable drugs and classified them according to their effects, mostly on symptoms into fifty groups.

Twort (1915) and d’Herelle (1917) independently discovered a lytic phenomenon in bacterial cultures. The agents responsible were termed bacteriophages—virus that attack bacteria. Early hopes that bacteriophages may have the therapeutic applications have to be abundones, but these virus have paid unexpected scientific dividends. The essential part of viruses is their core of nucleic acid which acts as the carrier of genetic information in the same manner as a higher organisms. The discipline of molecular biology owes it’s origin largely for studies on the genetics of bacteriophages and bacteria.
The development of bacteriology has scientific dates from Louis Pasteur (1822-95). His studies of fermentation led him to take an interest in micro-organism. He established that fermentation was the result of microbial activity and that different types of fermentations were associated with the activity of different kinds of microorganisms (1857).

Development of modern pharmacology as a science is fairly recent and probably started taking shape following the introduction of experimental procedure by Francois Magendie (1783-1855) and Claude Bernard (1813-1878). Spectacular developments in physiology, biochemistry and organic chemistry during the recent years have greatly accelerated the advances in pharmacology.

The disc diffusion methods provides a simple convenient and reliable test specially applicable in routine clinical bacteriology laboratory. According to literature much work has been done on heterocyclic compounds for their antibacterial activities on gram positive and gram negative bacteria.

Chalcones and their derivatives are reported to have antibacterial, antifungal, antiparasitic, antitubercular, antiinflammatory$^1$ and insect repellent properties$^2$.

---

Sharma et al.\textsuperscript{3}, have reported the antimicrobial activities of hydroxyaryl pyrazoles (1).\[ \text{R}_1 \text{N} = \text{N} \backslash \text{R}_2 \text{OH} \]

Mittal et al.\textsuperscript{4}, have reported antimicrobial activities of substituted pyrazoles (2).\[ \text{R}_4 \text{N} = \text{N} \backslash \text{R}_3 \text{N} = \text{N} \backslash \text{R}_2 \text{OCH}_3 \]

Anderson and Paolella\textsuperscript{5} synthesized 1-phenyl-pyrazole derivatives (3) and reported as effective antidiabetics.\[ \text{R} = \text{Me, CF}_3, \text{NH}_2 \]

\[ n = 1, 2 \]


Micetich et al.⁶, have reported that trifluoromethyl-1-aryl pyrazole as analgesic, antipyretic and antiinflammatory agents.

Alkyl pyrazole derivatives (4) have also been reported as hypolidermic agents⁷.

![Chemical structure of (4)](image)

Giri et al.⁸, studied antifungal nature of 3-(2-hydroxyphenyl)-5-(4-nitrophenyl) pyrazoles.

Basu et al.⁹, have been reported the synthesis of pyrazoles (5) as useful intermediates for pesticides and anticonvulsants.

![Chemical structure of (5)](image)

3, 5-Diphenyl-1H-pyrazole derivatives¹⁰(6) and (7) showed sedative, platelet antiaggregating, anaesthetic, analgesic and anti-inflammatory activities.

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3, 5-Diaryl substituted pyrazoles were reported by Chandara et al.\textsuperscript{11}, as antibacterial agents.

Parmar et al.\textsuperscript{12}, have reported that 3, 5-disubstituted pyrazoles (8) found to possess strong anti-invasive activity against human breast carcinoma cells.

Series of novel 1, 5-diarylpyrazole derivatives was synthesized and tested for anti-inflammatory and analgesic activity have been reported\textsuperscript{13}.

---


Kidwai et al.\textsuperscript{14}, have reported the antifungal activity of substituted pyrazole derivatives (9).

![Chemical structure of substituted pyrazole derivatives](image)

Tayade et al.\textsuperscript{15}, have reported the antibacterial activities of 3,5-diaryl pyrazoles.

Synthesis of some novel pyrazole derivatives as potential anti-inflammatory agents with minimum ulcerogenic activity have been reported\textsuperscript{16}.

Palkar et al.\textsuperscript{17}, have been reported the antibacterial activity of 3,5-diaryl pyrazoles (10) and (11).

![Chemical structures of pyrazole derivatives](image)

\begin{itemize}
\end{itemize}
Shingare et al.\textsuperscript{18}, have reported the antimicrobial activity of some new coumarino pyrazoles (12) against \textit{E. coli}, \textit{Lactobacillus}, \textit{aurens}, \textit{Ps. aeruginosa}.

\begin{center}
\includegraphics{image1}
\end{center}

Kidwai et al.\textsuperscript{19}, have reported the antifungal activity of substituted pyrazoles (13).

\begin{center}
\includegraphics{image2}
\end{center}

Mulwad and Pawar\textsuperscript{20} have reported antimicrobial activity of 4-hydroxy-3-[3',5'-substituted) pyrazoles benzopyran-2-ones (14).

\begin{center}
\includegraphics{image3}
\end{center}

Nagar et al.\textsuperscript{21}, have reported antimicrobial activity of 2-amino-3-cyano-4,5-dimethyl-7-N-phenyl-4-(p-arylaminophenyl)-pyrano-[3,2-d] pyrazoles (15).

![Chemical Structure](chemical_structure_15.png)

\textsuperscript{(15)}

Thakare\textsuperscript{22} reported the antimicrobial activity of 1-substituted-3,5-diaryl pyrazoles (16).

![Chemical Structure](chemical_structure_16.png)

\textsuperscript{(16)}

\begin{itemize}
\end{itemize}
Antimicrobial activity of 3,5-diaryl-4-aryloyl-1-substituted pyrazoles (17) have been reported\textsuperscript{23}.

\[ \text{Diagram of 3,5-diaryl-4-aryloyl-1-substituted pyrazoles (17)} \]

Chandak\textsuperscript{24} reported the antimicrobial activity of 1-substituted-3-(4-benzenesulphonamidophenyl)-5-aryl pyrazoles (18).

\[ \text{Diagram of 1-substituted-3-(4-benzenesulphonamidophenyl)-5-aryl pyrazoles (18)} \]

Thakare\textsuperscript{25} reported the antimicrobial activity of 1-substituted-3-(4-methyl-7-hydroxy-8-coumarinyl)-5-aryl pyrazoles (19).

\[ \text{Diagram of 1-substituted-3-(4-methyl-7-hydroxy-8-coumarinyl)-5-aryl pyrazoles (19)} \]

\begin{itemize}
\end{itemize}
Chapter 2
ORIGIN OF THE PROBLEM

The literature survey reveals that, much work has been done over many years for the study of antimicrobial activities of heterocyclic compounds on gram positive and gram negative micro-organism.

Many synthetic flavones are known to possess various physiological activities\(^{26-29}\). In many flavanoids, a large group of plant products are known to possess bactericidal and anti-inflammatory and analgesic activities\(^{30}\).

On the other hand pyrazoles are found to possess uretic\(^{31}\), antihelminthetic\(^{32}\) activities in addition to fungicidal activities\(^{33}\). Some substitutted pyrazoles are found to have a antimicrobial activity\(^{34}\).

26. Murti, V. V. S., Rao, N. V.S. and Sheshadri, T. R.,

27. Mertzer, C.,

28. Dimaggio, G.,

29. Griffith, J. Krewson, C. F. and Maghashi, J.,

30. Lesphghol, A., Hesagnol, C.,
Lesieur, D., Cazin, J. C., Lazin and Ramond, C.,

31. Garg, H. G.,

32. Garg, H. G. and Kaur, N.,


34. Ahluwalia, V. K., Dutta, U. and Sharma, H. R.,


Chem. Abstr., 78(1973), 52753C.
Dhiman\textsuperscript{35} reported the antimicrobial and MIC study of some synthesized heterocycles.

Flavanones and its related compounds are known to possess antimicrobial activity\textsuperscript{36}.

Thus literature survey reveals that, antimicrobial activities and MIC of 2-arylcoumaran-3-ones (4a-f), 3-aryl-[1] benzofurano [3,2-c]-1-substituted pyrazoles (5a-f/6a-f/7a-f) and 3-(chloroaryl)-5-aryl-1-substituted pyrazoles (10a-i/11a-i/12a-i). (Preparation described in chapter 4 of Part I and II) have not yet been studied.

Hence it was thought of interest to synthesize these compounds and study their antimicrobial activities and their minimum inhibitory concentrations against gram positive and gram negative microorganisms with the help of disc diffusion method.

\textsuperscript{35} Dhiman, A. M.,

\textsuperscript{36} Desai, Jigar and Nair, K. B.,


PROBLEM

This part of the thesis deals with the study of antimicrobial activities and MIC of newly synthesized coumaran-3-ones, and pyrazoles against pathogenic micro-organisms. Screening of the following compounds were carried out against the microbes Staphylococcus aureus, Escherichia coli, Proteus vulgaris, Pseudomonas aeruginosa, Enterobacter aerogen and Salmoella typhi.

1) 2-Aroyl-coumaran-3-ones (4a-f).

![2-Aroyl-coumaran-3-ones (4a-f)](image)

2) 3-Aryl-[1] benzofurano [3,2-c]-1-substituted pyrazoles (5a-f / 6a-f / 7a-f).

![3-Aryl-[1] benzofurano [3,2-c]-1-substituted pyrazoles (5a-f / 6a-f / 7a-f)](image)

3) 3-(Chloroaryl)-5-aryl-1-substituted pyrazoles (10a-i / 11a-i / 12a-i).

![3-(Chloroaryl)-5-aryl-1-substituted pyrazoles (10a-i / 11a-i / 12a-i)](image)
Chapter 3
SUMMARY OF THE WORK

In the present work, following compounds were synthesized and tested against micro-organisms (Staphylococcus aureus, Escherichia coli, Proteus vulgaris, Pseudomonas aeruginosa, Enterobacter aerogen and Salmonella typhi) for the study of their antimicrobial activity and the minimum inhibitory concentrations.

2-Aroyl-coumaran-3-ones (4a-f):
1. (4a) 2-Benzoyl-5-chloro-coumaran-3-one.
2. (4b) 2-Benzoyl-5-chloro-7-bromo-coumaran-3-one.
3. (4c) 2-Benzoyl-5-chloro-7-nitro-coumaran-3-one.
4. (4d) 2-(4-Methoxy benzoyl)-5-chloro-coumaran-3-one.
5. (4e) 2-(4-Methoxy benzoyl)-5-chloro-7-bromo-coumaran-3-one.
6. (4f) 2-(4-Methoxy benzoyl)-5-chloro-7-nitro-coumaran-3-one.

3-Aryl-[1] benzofurano [3,2-c]-1-substituted pyrazoles (5a-f/6a-f/7a-f):
1. (5a) 3-Phenyl-[1]-5-chloro-benzofurano [3, 2-c]-1-isonicotinoyl pyrazole.
2. (5b) 3-Phenyl-[1]-5-chloro-7-bromo-benzofurano [3,2-c]-1-isonicotinoyl pyrazole.
3. (5c) 3-Phenyl-[1]-5-chloro-7-nitro-benzofurano [3,2-c]-1-isonicotinoyl pyrazole.
4. (5d) 3-(4-Methoxy phenyl-[1]-5-chloro-benzofurano [3,2-c]-1-isonicotinoyl pyrazole.

5. (5e) 3-(4-Methoxy phenyl-[1]-5-chloro-7-bromo-benzofurano [3,2-c]-1-isonicotinoyl pyrazole.

6. (5f) 3-(4-Methoxy phenyl-[1]-5-chloro-7-nitro-benzofurano [3,2-c]-1-isonicotinoyl pyrazole.

10. (6a) 3-Phenyl-[1]-5-chloro benzofurano [3,2-c]-1-carboxamido pyrazole.

11. (6b) 3-Phenyl-[1]-5-chloro-7-bromo-benzofurano [3,2-c]-1-carboxamido pyrazole.

12. (6c) 3-Phenyl-[1]-5-chloro-7-nitro-benzofurano [3,2-c]-1-carboxamido pyrazole.

13. (6d) 3-(4-Methoxy phenyl)-[1]-5-chloro benzofurano [3,2-c]-1-carboxamido pyrazole.

14. (6e) 3-(4-Methoxy phenyl)-[1]-5-chloro-7-bromo-benzofurano [3,2-c]-1-carboxamido pyrazole.

15. (6f) 3-(4-Methoxy phenyl)-[1]-5-chloro-7-nitro-benzofurano [3,2-c]-1-carboxamido pyrazole.

10. (7a) 3-Phenyl-[1]-5-chloro benzofurano [3,2-c]-1-thiocarboxamido pyrazole.

11. (7b) 3-Phenyl-[1]-5-chloro-7-bromo-benzofurano [3,2-c]-1-thiocarboxamido pyrazole.

12. (7c) 3-Phenyl-[1]-5-chloro-7-nitro-benzofurano [3,2-c]-1-thiocarboxamido pyrazole.
13. (7d) 3-(4-Methoxy phenyl)-[1]-5-chloro benzofurano [3,2-c]-1-thiocarboxamido pyrazole.

14. (7e) 3-(4-Methoxy phenyl)-[1]-5-chloro-7-bromo- benzofurano [3,2-c]-1-thiocarboxamido pyrazole.

15. (7f) 3-(4-Methoxy phenyl)-[1]-5-chloro-7-nitro-benzofurano [3,2-c]-1-thiocarboxamido pyrazole.

3-(Chloroaryl)-5-aryl-1-substituted pyrazoles (10a-i/11a-i/12a-i):

1. (10a) 3-(2-hydroxy-5-chlorophenyl)-5-(4-methoxy phenyl)-1-isonicotinoyl pyrazole.

2. (10b) 3-(2-hydroxy-3-bromo-5-chlorophenyl)-5-(4-methoxy phenyl)-1-isonicotinoyl pyrazole.

3. (10c) 3-(2-hydroxy-3-nitro-5-chlorophenyl)-5-(4-methoxy phenyl)-1-isonicotinoyl pyrazole.

4. (10d) 3-(2-hydroxy-5-chloro phenyl)-5-phenyl-1-isonicotinoyl pyrazole.

5. (10e) 3-(2-hydroxy-3-bromo-5-chloro phenyl)-5-phenyl-1-isonicotinoyl pyrazole.

6. (10f) 3-(2-hydroxy-3-nitro-5-chloro phenyl)-5-phenyl-1-isonicotinoyl pyrazole.

7. (10g) 3-(2-hydroxy-5-chloro phenyl)-5-(3-chloro phenyl)-1-isonicotinoyl pyrazole.

8. (10h) 3-(2-hydroxy-3-bromo-5-chloro phenyl)-5-(3-chloro phenyl)-1-isonicotinoyl pyrazole.
9. (10i) 3-(2-hydroxy-3-nitro-5-chlorophenyl)-5-(3-chlorophenyl)-1-isonicotinoyl pyrazole.

10. (11a) 3-(2-hydroxy-5-chlorophenyl)-5-(4-methoxy phenyl)-1-carboxamido pyrazole.

11. (11b) 3-(2-hydroxy-3-bromo-5-chlorophenyl)-5-(4-methoxy phenyl)-1-carboxamido pyrazole.

12. (11c) 3-(2-hydroxy-3-nitro-5-chlorophenyl)-5-(4-methoxy phenyl)-1-carboxamido pyrazole.

13. (11d) 3-(2-hydroxy-5-chlorophenyl)-5-phenyl-1-carboxamido pyrazole.

14. (11e) 3-(2-hydroxy-3-bromo-5-chlorophenyl)-5-phenyl-1-carboxamido pyrazole.

15. (11f) 3-(2-hydroxy-3-nitro-5-chlorophenyl)-5-phenyl-1-carboxamido pyrazole.

16. (11g) 3-(2-hydroxy-5-chlorophenyl)-5-(3-chlorophenyl)-1-carboxamido pyrazole.

17. (11h) 3-(2-hydroxy-3-bromo-5-chlorophenyl)-5-(3-chlorophenyl)-1-carboxamido pyrazole.

18. (11i) 3-(2-hydroxy-3-nitro-5-chlorophenyl)-5-(3-chlorophenyl)-1-carboxamido pyrazole.

19. (12a) 3-(2-hydroxy-5-chlorophenyl)-5-(4-methoxy phenyl)-1-thiocarboxamido pyrazole.

20. (12b) 3-(2-hydroxy-3-bromo-5-chlorophenyl)-5-(4-methoxy phenyl)-1-thiocarboxamido pyrazole.
21. (12c) 3-(2-hydroxy-3-nitro-5-chlorophenyl)-5-(4-methoxyphenyl)-1-thiocarboxamido pyrazole.

22. (12d) 3-(2-hydroxy-5-chlorophenyl)-5-phenyl-1-thiocarboxamido pyrazole.

23. (12e) 3-(2-hydroxy-3-bromo-5-chlorophenyl)-5-phenyl-1-thiocarboxamido pyrazole.

24. (12f) 3-(2-hydroxy-3-nitro-5-chlorophenyl)-5-phenyl-1-thiocarboxamido pyrazole.

25. (12g) 3-(2-hydroxy-5-chlorophenyl)-5-(3-chlorophenyl)-1-thiocarboxamido pyrazole.

26. (12h) 3-(2-hydroxy-3-bromo-5-chlorophenyl)-5-(3-chlorophenyl)-1-thiocarboxamido pyrazole.

27. (12i) 3-(2-hydroxy-3-nitro-5-chlorophenyl)-5-(3-chlorophenyl)-1-thiocarboxamido pyrazole.

All the compounds were synthesized and experimental details have been described in chapter 4 (Part I and II) of the thesis. Structures of all the above compounds were confirmed on the basis of chemical properties, elemental analysis and spectral data.

These compounds were tested against following six pathogens for their antimicrobial activities using paper-disc method.

1) Staphylococcus aureus:

Staphylococcus are gram +ve, non-motile cocci arranged in groups. They are parasites occurring in the skin and mucous membranes of human and animals.
2) **Escherichia coli:**

These are gram -ve bacillus. *E. coli* occurs in the lower part of intestine of humans and animals, where it is a part of the normal flora. Some strains can cause gastroenteritis, others can cause urinary tract infections.

3) **Proteus vulgaris:**

These are gram -ve, non-sporeforming, motile, rod shaped bacteria. They are found in soil, sewage and decaying matter. They can produce $\text{H}_2\text{S}$ gas from sulphur containing amino acids, but do not ferment lactose. They may inhibit intestinal tract of man and animals and are responsible for infection of urinary tract, septicemia etc.

4) **Pseudomonas aeruginosa:**

These are gram -ve bacillus. They are widely distributed in soil and water. *P. aeruginosa* produces a water soluble blue pigment, pyocyanin and a water soluble fluorescent pigment, pyoverdin. The organisms are also frequently opportunistic pathogen and can often be isolated from wounds, burns and urinary tract infections.

5) **Enterobacter aerogen:**

These are gram -ve bacteria. They are widely distributed in soil and water. The organisms are frequently opportunistic pathogen and can often be isolated from wounds, burns and urinary tract infections and is a enteric pathogens.
6) Salmoella typhi:

These are gram-ve bacillus. They are pathogenic for humans, causing typhoid, gastroenteritis and septicemia, many strains also infect a variety of animals, over 2000 different types of salmonellae occur.
Chapter 4

EXPERIMENTAL AND DISCUSSION OF THE RESULTS

The compounds have been characterized on the basis of chemical properties, elemental analysis and spectral analysis (IR and \(^1\)HNMR). The melting points were recorded on open capillary-silicon oil bath and are uncorrected. The purity of the compounds were checked by TLC on silicon gel-G plates. The procedure for the synthesis of coumaran-3-ones (4a-f), 3-aryl-1 benzofurano [3,2-c]-1-substituted pyrazoles (5a-f/6a-f/7a-f) and 3-(chlooraryl)-5-aryl-1-substituted pyrazoles (10a-i/11a-i/12a-i) is described in chapter 4 (Part I and II) of the thesis.

MATERIALS AND METHOD

The compounds mentioned above were tested against pathogenic bacteria for their antibacterial activities using paper disc method and their minimum inhibitory concentrations (MICs) were determined using broth macrodilution method. The organisms used for both these methods were Staphylococcus aureus, Escherichia coli, Proteus vulgaris, Pseudomonas aeruginosa, Enterobacter aerogen and Salmoella typhi.

PREPARATION OF WET DISC FOR ANTIBIOTIC SENSITIVITY TEST

Method:

Discs (10 mm) in diameter from Whatman filter paper were punched and batches of 100 in screw-capped bottles were dispersed and sterilized by dry heat at 140°C for 60 minutes. The solution of the compound in DMSO solvent was prepared so that 1 ml contains 100
times the amount of the compound required in the disc. 1 ml solution of the compound was added to each bottle of 100 discs and as the whole of this volume is absorbed. It was assumed that each disc contains approximately 0.01 ml of media. Discs were stored in wet conditions.

**Culture Medium:**

The medium used throughout the experiment was HI-media (Indian make) nutrient agar and having following composition:

**Composition of nutrient agar**

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peptone</td>
<td>5.0 g/litre</td>
</tr>
<tr>
<td>Beef extract</td>
<td>5.0 g/litre</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>5.0 g/litre</td>
</tr>
<tr>
<td>Yeast extract</td>
<td>15 g/litre</td>
</tr>
<tr>
<td>Agar</td>
<td>15 g/litre</td>
</tr>
<tr>
<td>pH (Approx.)</td>
<td>7.4 ± 0.2</td>
</tr>
</tbody>
</table>

The medium was prepared by suspending 28 gm ingredients in 1000 ml distilled water. It was boiled to dissolve the medium completely and was sterilized by autoclave at 15 lbs/inch² pressure at 121°C temperature. It was cooled to about 50°C and poured into sterile petriplates and allowed to solidify.

**Medium used**

<table>
<thead>
<tr>
<th>Nutrient Agar Medium</th>
</tr>
</thead>
<tbody>
<tr>
<td>Size</td>
</tr>
<tr>
<td>Depth of agar</td>
</tr>
<tr>
<td>Distance between 2 discs</td>
</tr>
<tr>
<td>Diameter of the antibiotic discs</td>
</tr>
</tbody>
</table>

(Not more than five discs per plate were used)
Test Procedure:

The culture material was inoculated in a nutrient broth and kept at 37°C for 24 hours incubation. The culture plate nutrient agar was dried until its surface was free from visible moisture. The inoculating material was then flooded on the surface of nutrient agar uniformly taking all antiseptic precautions. The plate was dried again for up to 30 minutes without further delay and the compound disc were applied at adequate spacing (2 cm or more apart) to the surface of the plate with sterile fine-pointed forceps and gently pressed to ensure full contact with the medium and moistening of the disc. Control was run using plane DMSO solvent for aseptic conditions. The plates were incubated at 37°C for 18-24 hours. After incubation degree of sensitivity to drugs is determined by measuring the visible clear areas of growth of free zones (zones of inhibition) produced by diffusion of antibiotics into the media from the discs.

DETERMINATION OF MINIMUM INHIBITORY CONCENTRATION (MIC)

Broth Macrodilution Method:

A series of test tubes (12 x 75 mm) containing 0.5 ml nutrient broth each were prepared and sterilized in autoclave. After sterilization the tubes were cooled to room temperature. To the first tube 0.5 ml of stock solution of the test compound (1000 µg/ml) was added so that concentration of compound in first tube will be 500 µg/ml. Then it was mixed and 0.5 ml was transferred to the second tube, mixed thoroughly (conc. 250 µg/ml) and again 0.5 ml of it was transferred to third tube (conc. 125 µg/ml). In this way
concentration up to 15 \( \mu g/ml \) were prepared, 0.5 ml suspension from the last tube was discarded.

The compound disc were applied to the surface of the plate as described in test procedure. All the plates were incubated at 37 ± 2°C for 24 hours.

**MIC reading:**

The lowest concentration of the compound that inhibited the growth of organisms after incubation was taken as minimum inhibitory concentration (MIC).

Width of the zone of inhibition depends on:

1) Size of inoculum.
2) Nature of culture medium.
3) Presence of inhibitors.
4) Concentration of agar in the medium.
5) Thickness of the medium in the plate.
6) Condition and time of incubation.
7) Composition of antibiotic disc.

Zones of inhibition are measured and reported. The results are cited in Table No. 1, 2, 3, 4, 5, 6, 7 and 8.
Zone of inhibition against the compounds

Plate No. 1

Plate No. 2
Table No. 1:  
Antimicrobial activity of 2-aroyl-coumaran-3-ones (4a-f).

Table No. 2:  
Antimicrobial activity of 3-aryl-[1]-benzofurano [3,2-c]-1-isonicotinoyl pyrazoles (5a-f).

Table No. 3:  
Antimicrobial activity of 3-aryl-[1]-benzofuran [3,2-c]-1-carboxamidopyrazoles (6a-f).

Table No. 4:  
Antimicrobial activity of 3-aryl-[1] benzofurano [3,2-c]-1-thiocarboxamidopyrazoles (7a-f).

Table No. 5:  
Antimicrobial activity of 3-(chloroaryl)-5-aryl-1-isonicotinoyl pyrazoles (10a-i).

Table No. 6:  
Antimicrobial activity of 3-(chloroaryl)-5-aryl-1-carboxamidopyrazoles (11a-i).

Table No. 7:  
Antimicrobial activity of 3-(chloroaryl)-5-aryl-1-thiocarboxamidopyrazoles (12a-i).

Table No. 8:  
Minimum inhibitory concentration of coumaran-3-ones (4a) and 1-substituted pyrazoles (5a, 6a, 7a) and (10a, 11a, 12a).
Table No. 1
Antimicrobial activity of 2-arylcoumaran-3-ones (4a-f).

<table>
<thead>
<tr>
<th>Organism</th>
<th>4a</th>
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<th>4c</th>
<th>4d</th>
<th>4e</th>
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<tbody>
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<td>++</td>
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<td>-</td>
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<tr>
<td>E. coli</td>
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<td>-</td>
<td>++</td>
<td>+++</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Pr. vulgaris</td>
<td>++</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>++</td>
<td>+++</td>
</tr>
<tr>
<td>Ps. aeroginosa</td>
<td>+++</td>
<td>+++</td>
<td>++</td>
<td>++</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>Et. aerogen</td>
<td>+++</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>S. typhi</td>
<td>++</td>
<td>+</td>
<td>-</td>
<td>++</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

Table No. 2
Antimicrobial activity of 3-aryl-[1]-benzofurano [3,2-c]-1-isonicotinoylpyrazoles (5a-f).

<table>
<thead>
<tr>
<th>Organism</th>
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<th>5d</th>
<th>5e</th>
<th>5f</th>
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</thead>
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<td>++</td>
<td>++</td>
</tr>
<tr>
<td>E. coli</td>
<td>-</td>
<td>++</td>
<td>++</td>
<td>+++</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Pr. vulgaris</td>
<td>+++</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Ps. aeroginosa</td>
<td>++</td>
<td>+++</td>
<td>++</td>
<td>+++</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Et. aerogen</td>
<td>++</td>
<td>-</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
<td>++</td>
</tr>
<tr>
<td>S. typhi</td>
<td>-</td>
<td>-</td>
<td>+++</td>
<td>++</td>
<td>+++</td>
<td>++</td>
</tr>
</tbody>
</table>

N.B.:  
+++ : Strongly active, range > 15 mm
++  : Moderately active, range 12-15 mm
+   : Weakly active, range < 12 mm
-   : Inactive
### Table No. 3
Antimicrobial activity of 3-aryl-[1]-benzofurano [3,2-c]-1-carboxamido pyrazoles (6a-f).

<table>
<thead>
<tr>
<th>Organism</th>
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<th>6d</th>
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</tr>
<tr>
<td>E. coli</td>
<td>-</td>
<td>++</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Pr. vulgaris</td>
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<td>-</td>
<td>-</td>
<td>++</td>
<td>+++</td>
<td>++</td>
</tr>
<tr>
<td>Ps. aeruginosa</td>
<td>+</td>
<td>-</td>
<td>++</td>
<td>++</td>
<td>-</td>
<td>++</td>
</tr>
<tr>
<td>Et. aerogen</td>
<td>++</td>
<td>-</td>
<td>++</td>
<td>++</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>S. typhi</td>
<td>++</td>
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<td>+</td>
<td>+</td>
<td>+</td>
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</tr>
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</table>

### Table No. 4
Antimicrobial activity of 3-aryl-[1] benzofurano [3,2-c]-1-thiocarboxamido pyrazoles (7a-f).

<table>
<thead>
<tr>
<th>Organism</th>
<th>7a</th>
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<th>7c</th>
<th>7d</th>
<th>7e</th>
<th>7f</th>
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<td>++</td>
<td>-</td>
<td>-</td>
<td>++</td>
</tr>
<tr>
<td>E. coli</td>
<td>+</td>
<td>-</td>
<td>++</td>
<td>-</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>Pr. vulgaris</td>
<td>-</td>
<td>-</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>Ps. aeruginosa</td>
<td>-</td>
<td>+++</td>
<td>-</td>
<td>++</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Et. aerogen</td>
<td>++</td>
<td>+++</td>
<td>+++</td>
<td>++</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>S. typhi</td>
<td>++</td>
<td>-</td>
<td>+++</td>
<td>-</td>
<td>-</td>
<td>++</td>
</tr>
</tbody>
</table>

**N.B.:**
- **+++**: Strongly active, range > 15 mm
- **++**: Moderately active, range 12-15 mm
- **+**: Weakly active, range < 12 mm
- **-**: Inactive
### Table No. 5

**Antimicrobial activity of 3-(chboroaryl)-5-aryl-1-isonicotinoyl pyrazoles (10a–i).**

<table>
<thead>
<tr>
<th>Organism</th>
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<th>10b</th>
<th>10c</th>
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<tr>
<td>S. aureus</td>
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<td>++</td>
<td>++</td>
<td>−</td>
<td>++</td>
<td>++</td>
<td>++</td>
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</tr>
<tr>
<td>E. coli</td>
<td>−</td>
<td>−</td>
<td>++</td>
<td>++</td>
<td>−</td>
<td>+</td>
<td>−</td>
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<td>++</td>
</tr>
<tr>
<td>Pr. vulgaris</td>
<td>++</td>
<td>−</td>
<td>−</td>
<td>++</td>
<td>+</td>
<td>−</td>
<td>+++</td>
<td>++</td>
<td></td>
</tr>
<tr>
<td>Ps. aeruginosa</td>
<td>++</td>
<td>++</td>
<td>−</td>
<td>−</td>
<td>+++</td>
<td>++</td>
<td>−</td>
<td>+++</td>
<td>++</td>
</tr>
<tr>
<td>Et. aerogen</td>
<td>−</td>
<td>+</td>
<td>−</td>
<td>+++</td>
<td>+</td>
<td>+</td>
<td>−</td>
<td>−</td>
<td>−</td>
</tr>
<tr>
<td>S. typhi</td>
<td>++</td>
<td>+</td>
<td>−</td>
<td>+++</td>
<td>++</td>
<td>−</td>
<td>+</td>
<td>−</td>
<td>−</td>
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</tbody>
</table>

### Table No. 6

**Antimicrobial activity of 3-(chloroaryl)-5-aryl-1-carboxamido pyrazoles (11a–i).**

<table>
<thead>
<tr>
<th>Organism</th>
<th>11a</th>
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<th>11e</th>
<th>11f</th>
<th>11g</th>
<th>11h</th>
<th>11i</th>
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<td>−</td>
<td>++</td>
<td>+++</td>
<td>−</td>
<td>+++</td>
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<td>+++</td>
</tr>
<tr>
<td>E. coli</td>
<td>−</td>
<td>+</td>
<td>−</td>
<td>+++</td>
<td>++</td>
<td>+</td>
<td>−</td>
<td>−</td>
<td>−</td>
</tr>
<tr>
<td>Pr. vulgaris</td>
<td>−</td>
<td>−</td>
<td>++</td>
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<td>−</td>
<td>−</td>
<td>+++</td>
</tr>
<tr>
<td>Ps. aeruginosa</td>
<td>++</td>
<td>−</td>
<td>++</td>
<td>−</td>
<td>−</td>
<td>+</td>
<td>−</td>
<td>−</td>
<td>−</td>
</tr>
<tr>
<td>Et. aerogen</td>
<td>+++</td>
<td>−</td>
<td>++</td>
<td>−</td>
<td>+</td>
<td>+</td>
<td>++</td>
<td>−</td>
<td>−</td>
</tr>
<tr>
<td>S. typhi</td>
<td>+++</td>
<td>++</td>
<td>−</td>
<td>−</td>
<td>+++</td>
<td>−</td>
<td>+++</td>
<td>++</td>
<td>−</td>
</tr>
</tbody>
</table>

**N.B.:**

+++ : Strongly active, range > 15 mm  
++ : Moderately active, range 12–15 mm  
+ : Weakly active, range < 12 mm  
− : Inactive
### Table No. 7

**Antimicrobial activity of 3-(chloroaryl)-5-aryl-1-thiocarboxamido pyrazoles (12a-i).**

<table>
<thead>
<tr>
<th>Organism</th>
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<th>12c</th>
<th>12d</th>
<th>12e</th>
<th>12f</th>
<th>12g</th>
<th>12h</th>
<th>12i</th>
</tr>
</thead>
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<tr>
<td>S. aureus</td>
<td>-</td>
<td>+</td>
<td>++</td>
<td>-</td>
<td>-</td>
<td>++</td>
<td>+++</td>
<td>++</td>
<td>+++</td>
</tr>
<tr>
<td>E. coli</td>
<td>-</td>
<td>+</td>
<td>++</td>
<td>++</td>
<td>-</td>
<td>-</td>
<td>++</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Pr. vulgaris</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>++</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Ps. aeroginosa</td>
<td>++</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>++</td>
<td>++</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Et. aerogen</td>
<td>+++</td>
<td>++</td>
<td>++</td>
<td>-</td>
<td>++</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>S. typhi</td>
<td>++</td>
<td>++</td>
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<td>-</td>
<td>++</td>
<td>-</td>
<td>++</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

**N.B.:**
- +++: Strongly active, range > 15 mm
- ++ : Moderately active, range 12-15 mm
- +  : Weakly active, range < 12 mm
- -  : Inactive

### Table No. 8

**Minimum inhibitory concentration of coumaran-3-ones (4a) and 1-substituted pyrazoles (5a, 6a, 7a) and (10a, 11a, 12a).**

<table>
<thead>
<tr>
<th>Organism</th>
<th>4a</th>
<th>5a</th>
<th>6a</th>
<th>7a</th>
<th>10a</th>
<th>11a</th>
<th>12a</th>
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<td>250</td>
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<td>500</td>
<td>62</td>
<td>125</td>
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<tr>
<td>E. coli</td>
<td>30</td>
<td>30</td>
<td>30</td>
<td>125</td>
<td>125</td>
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<tr>
<td>Pr. vulgaris</td>
<td>500</td>
<td>30</td>
<td>30</td>
<td>30</td>
<td>62</td>
<td>30</td>
<td>125</td>
</tr>
<tr>
<td>Ps. aeroginosa</td>
<td>62</td>
<td>250</td>
<td>62</td>
<td>500</td>
<td>125</td>
<td>62</td>
<td>30</td>
</tr>
<tr>
<td>Et. aerogen</td>
<td>62</td>
<td>125</td>
<td>125</td>
<td>125</td>
<td>62</td>
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<td>125</td>
</tr>
<tr>
<td>S. typhi</td>
<td>250</td>
<td>125</td>
<td>30</td>
<td>62</td>
<td>30</td>
<td>30</td>
<td>125</td>
</tr>
</tbody>
</table>
DISCUSSION OF THE RESULTS

I] Antimicrobial activity of 2-arylcoumaran-3-ones (4a-f):

From Table No. 1, the compound (4a) was inactive against S. aureus and E. coli, moderately active towards Pr. vulgaris and S. typhi and was found strongly active towards Ps. aerogenosa and Et. aerogen.

The compound (4b) was moderately active towards S. aureus, Et. aerogen and S. typhi, inactive against E. coli and Pr. vulgaris and was found strongly active towards Ps. aeroginosa.

The compound (4c) was found moderately active towards S. aureus, E. coli and Ps. aeroginosa and was found inactive against Pr. vulgaris, Et. aerogen and S. typhi.

The compound (4d) was found strongly active towards S. aureus and E. coli, moderately active towards Ps. aeroginosa and S. typhi and was found inactive against Pr. vulgaris, Et. aerogen.

The compound (4e) was weakly active against S. aureus and Et. aerogen, moderately active towards E. coli and Pr. vulgaris and was found inactive against Ps. aeroginosa, S. typhi.

The compound (4f) was inactive against S. aureus and S. typhi, moderately active towards E. coli and Ps. aeroginosa and Et. aerogen was found strongly active towards Pr. vulgaris.
II] Antimicrobial activity of 3-aryl-[1]-benzofurano [3,2-c]-1-isonicotinoyl pyrazoles (5a-f):

From Table No. 2, the compound (5a) was found weakly active towards *S. aureus*, inactive against *E. coli* and *S. typhi*, moderately active towards *Ps. aeroginosa* and *Et. aerogen* and strongly active towards *Pr. vulgaris*.

The compound (5b) was found moderately active towards *S. aureus*, *E. coli* and *Pr. vulgaris*, strongly active towards *Ps. aeroginosa* and was inactive against *Et. aerogen* and *S. typhi*.

The compound (5c) was found moderately active towards *S. aureus*, *E. coli* and *Ps. aeroginosa*, strongly active towards *Et. aerogen* and *S. typhi* and was inactive against *Pr. vulgaris*.

The compound (5d) was inactive against *S. aureus* and *Pr. vulgaris*, strongly active towards *E. coli*, *Ps. aeroginosa* and *Et. aerogen* and was found moderately active towards *S. typhi*.

The compound (5e) was found moderately active towards *S. aureus* and *Ps. aeroginosa*, was inactive against *E. coli* and *Pr. vulgaris* and strongly active towards *Et. aerogen* and *S. typhi*.

The compound (5f) was found moderately active towards *S. aureus*, *Et. aerogen* and *S. typhi*, and was inactive against *E. coli*, *Pr. vulgaris* and *Ps. aeroginosa*. 
Antimicrobial activity of 3-aryl-[1]-benzofurano [3,2-c]-1-carboxamido pyrazoles (6a-f):

From Table No. 3, the compound (6a) was inactive against *S. aureus* and *E. coli*, weakly active towards *P. vulgaris* and *P. aeruginosa* and was found moderately active towards *E. aerogen* and *S. typhi*.

The compound (6b) was found moderately active towards *S. aureus* and *E. coli*, inactive against *P. vulgaris*, *P. aeruginosa* and *E. aerogen* and was strongly active towards *S. typhi*.

The compound (6c) was found weakly active towards *S. aureus* and *S. typhi*, was inactive towards *E. coli* and *P. vulgaris* and was found moderately active towards *P. aeruginosa* and *E. aerogen*.

The compound (6d) was inactive against *S. aureus* and *E. coli*, was found moderately active towards *P. vulgaris*, *P. aeruginosa* and *E. aerogen* and was weakly active towards *S. typhi*.

The compound (6e) was inactive against *E. coli*, *P. aeruginosa*, was found moderately active towards *S. aureus* and was strongly active towards *P. vulgaris* and *S. typhi*.

The compound (6f) was inactive against *S. aureus*, *E. coli* and *E. aerogen*, was moderately active towards *P. vulgaris* and *P. aeruginosa* and was strongly active towards *S. typhi*.
IV] Antimicrobial activity of 3-aryl-[1]-benzofurano [3,2-c]-1-thiocarboxamido pyrazoles (7a-f):

From Table No. 4, the compound (7a) was inactive against Pr. vulgaris and Ps. aeroginosa, was found weakly active towards S. aureus and E. coli and was moderately active towards Et. aerogen and S. typhi.

The compound (7b) was inactive against E. coli, Pr. vulgaris and S. typhi and moderately active towards S. aureus but was found strongly active towards Et. aerogen and S. typhi.

The compound (7c) was found moderately active towards S. aureus, E. coli and Pr. vulgaris and inactive against Ps. aeroginosa but found strongly active towards Et. aerogen and S. typhi.

The compound (7d) was inactive against S. aureus, Ps. aeroginosa, Et. aerogen and S. typhi and was found moderately active towards Pr. vulgaris, Ps. aeroginosa and Et. aerogen.

The compound (7e) was inactive against S. aureus, Ps. aeroginosa, Et. aerogen and S. typhi and moderately active towards E. coli and Pr. vulgaris.

The compound (7f) was moderately active towards S. aureus, E. coli, Pr. vulgaris and S. typhi but was inactive against Ps. aeroginosa and Et. aerogen.
V] Antimicrobial activity of 3-(chloroaryl)-5-aryl-1-isonicotinoyl pyrazoles (10a-i):

From Table No. 5, the compound (10a) was inactive against \( \text{S. aureus} \), \( \text{E. coli} \) and \( \text{Et. aerogen} \) and was found moderately active towards \( \text{Pr. vulgaris} \), \( \text{Ps. aeroginosa} \) and \( \text{S. typhi} \).

The compound (10b) was inactive against \( \text{Ps. aeroginosa} \) and \( \text{Et. aerogen} \) and moderately active towards \( \text{S. aureus} \) and \( \text{Ps. aeroginosa} \) but was found weakly active towards \( \text{Et. aerogen} \) and \( \text{S. typhi} \).

The compound (10c) was found moderately active towards \( \text{S. aureus} \) and \( \text{E. coli} \) and was found inactive against \( \text{Pr. vulgaris} \), \( \text{Ps. aeroginosa} \), \( \text{Et. aerogen} \) and \( \text{S. typhi} \).

The compound (10d) was inactive against \( \text{S. aureus} \), \( \text{Ps. aeroginosa} \) and \( \text{Et. aerogen} \) and was found moderately active towards \( \text{E. coli} \), \( \text{Pr. vulgaris} \) and \( \text{S. typhi} \).

The compound (10e) was inactive against \( \text{E. coli} \) and \( \text{Pr. vulgaris} \), and moderately active towards \( \text{S. aureus} \) and \( \text{S. typhi} \) but was found strongly active towards \( \text{Ps. aeroginosa} \) and \( \text{Et. aerogen} \).

The compound (10f) was weakly active towards \( \text{E. coli} \), \( \text{Pr. vulgaris} \) and \( \text{Et. aerogen} \) and was found moderately active towards \( \text{S. aureus} \), \( \text{Ps. aeroginosa} \) and \( \text{S. typhi} \).
The compound (10g) was inactive against E. coli, Pr. vulgaris, Ps. aeroginosa and S. typhi and was found moderately active towards S. aureus and Et. aerogen.

The compound (10h) was inactive against S. aureus and E. coli, and strongly active towards Pr. vulgaris and Ps. aeroginosa but found weakly active towards Et. aerogen and S. typhi.

The compound (10i) was inactive against S. aureus, Et. aerogen and S. typhi and was found moderately active towards E. coli, Pr. vulgaris and Ps. aeroginosa.

VI | Antimicrobial activity of 3-(chloroaryl)-5-aryl-1-carboxamido pyrazoles (11a-i):

From Table No. 6, the compound (11a) was inactive against S. aureus, E. coli and Pr. vulgaris and moderately active towards Ps. aeroginosa but found strongly active towards Et. aerogen and S. typhi.

The compound (11b) was found moderately active towards S. aureus and S. typhi and weakly active towards E. coli and was found inactive against Pr. vulgaris, Ps. aeroginosa and Et. aerogen.

The compound (11c) was inactive against S. aureus, E. coli and S. typhi and was found moderately active towards Pr. vulgaris, Ps. aeroginosa and Et. aerogen.
The compound (11d) was moderately active towards S. aureus and strongly active towards E. coli and Pr. vulgaris but was found inactive against Ps. aeroginosa, Et. aerogen and S. typhi.

The compound (11e) was strongly active towards S. aureus and S. typhi, and moderately active towards E. coli and Pr. vulgaris but was found inactive against Ps. aeroginosa and Et. aerogen.

The compound (11f) was inactive against S. aureus, Ps. aeroginosa and S. typhi and was found weakly active towards E. coli, Pr. vulgaris and Et. aerogen.

The compound (11g) was inactive against E. coli and Pr. vulgaris and weakly active towards Ps. aeroginosa and Et. aerogen but was found strongly active towards S. aureus and S. typhi.

The compound (11h) was inactive against E. coli and Pr. vulgaris and Ps. aeroginosa and moderately active towards S. aureus, Et. aerogen and S. typhi.

The compound (11i) was found strongly active towards S. aureus and Pr. vulgaris and inactive towards E. coli, Ps. aeroginosa, Et. aerogen and S. typhi.

VIII Antimicrobial activity of 3-(chloroaryl)-5-aryl-1-thiocarboxamido pyrazoles (12a-i):

From Table No. 7, the compound (12a) was inactive against S. aureus, E. coli and Pr. vulgaris, moderately active towards Ps. aeroginosa and S. typhi and was found strongly active towards Et. aerogen.
The compound (12b) was weakly active towards S. aureus and E. coli and found inactive against Pr. vulgaris and Ps. aeroginosa but was found moderately active towards Et. aerogen.

The compound (12c) was found moderately active towards S. aureus, E. coli and Et. aerogen and inactive against Pr. vulgaris and Ps. aeroginosa but was strongly active towards S. typhi.

The compound (12d) was inactive against S. aureus, Ps. aeroginosa, Et. aerogen and S. typhi and was found moderately active towards E. coli and Pr. vulgaris.

The compound (12e) was inactive against S. aureus, E. coli and Pr. vulgaris and was found moderately active towards Ps. aeroginosa, Et. aerogen and S. typhi.

The compound (12f) was inactive against E. coli, Pr. vulgaris and S. typhi and moderately active towards S. aureus and Ps. aeroginosa but was found strongly active towards Et. aerogen.

The compound (12g) was strongly active towards S. aureus and moderately active towards E. coli, Ps. aeroginosa and S. typhi but was found inactive against Pr. vulgaris and Et. aerogen.
The compound (12h) was found inactive against E. coli, Pr. vulgaris and Et. aerogen and was moderately active towards S. aureus, Ps. aeroginosa and S. typhi.

The compound (12i) was inactive against E. coli, Ps. aeroginosa and Et. aerogen and strongly active towards S. aureus but was found moderately active towards Pr. vulgaris, and S. typhi.

VIII] Minimum inhibitory concentration of coumaran-3-ones (4a) and 1-substituted pyrazoles (5a, 6a, 7a) and (10a, 11a, 12a).

From Table No. 8, the MIC results indicates that, the compound (4a) have inhibited the growth of E. coli at 30 μg/ml while for other bacteria the MIC range was 500-62 μg/ml.

The compound (5a) was found strongly active in inhibiting the growth of E. coli and Pr. vulgaris at MIC range 30 μg/ml while for other bacteria the MIC range was 250-62 μg/ml.

The compound (6a) was found to inhibited the growth of E. coli, Pr. vulgaris and S. typhi at 30 μg/ml while for other bacteria the MIC range was found to be at 250-62 μg/ml.

The compound (7a) inhibited the growth of Pr. vulgaris at 30 μg/ml while the other bacteria were found to inhibited the growth in the MIC range 500-62 μg/ml.

The compound (10a) inhibited the growth of S. typhi in the MIC range 30 μg/ml while other bacteria found to shown the MIC range 500-62 μg/ml.
The compound (11a) was found strongly active and inhibiting the growth of *E. coli*, *P. vulgaris*, *P. aeruginosa* and *S. typhi* at 30 μg/ml while other bacteria showed the MIC range 62 μg/ml.

The compound (12a) inhibited the growth of *E. coli* and *E. aerogen* at 30 μg/ml while other bacteria showed the MIC range 125 μg/ml.