PREFACE

A heterocyclic compound represents an important class of biologically active molecules. This is reflected by the voluminous data available in the literature on heterocyclic chemistry. Many useful drugs indeed have emerged from such investigations which strengthens the trend. Spectacular advancement have been made in this field to furtherance the knowledge of relationship between chemical structure and biological activity. Thus, the successful application of this class of compounds in various fields ensures a limitless scope for the development of structurally novel heterocycles with a wide range of psycho-chemical and biological properties.

Amongst different heterocyclic systems, the chemistry of five membered heterocycles with more than one heteroatom has gained importance because many of them exhibit pronounced bioactive nature. One such type of compounds includes pyrazoles and pyrazolines. Hence, any attempt to study their detailed chemistry would add new dimensions to the existing knowledge. Pyrazolones, pyrazoles and related heterocycles possess various types of biological activities. A good deal of importance is given to pyrazolone derivatives. It is due to their wide use in medicinal chemistry.

Specifically, those containing the pyrazolone nucleus have been shown to possess biological activities such as tranquilizing, muscle relaxant, psycho analeptic, anticonvulsant, antihypertensive, antidepressant activities. The derivatives of pyrazolone are important
class of antipyretic and analgesic Compounds. Besides antituberculosis antineoplastic, antidiadetic, anti fertility and antithyroid activity. In this perspective, a study on Synthesis, characterization, biological evaluation and electro chemical behavior of hetero cyclic compounds have been taken up and incorporated in the thesis.

CHAPTER – I: Brief review on chemistry of pyrazole, azetidones and thiazolidinone derivatives

A five membered cyclic diene containing three carbons and two nitrogen’s is called a diazole. If two nitrogen atoms are adjacent, it is known as a pyrazole. If one double bond is present, it is a pyrazoline and two double bonds are present it is a pyrazole. If a five-membered ring containing two adjacent nitrogen atoms and ketonic group in the same molecule, it is called pyrazolone. Several types are possible, depending on the substituents to pyrazoline ring.

Amongst the all types of pyrazolines the chemistry of pyrazolone 2 has received much attention in the recent past due to their industrial and pharmacological applications. Pyrazolones,
pyrazoles and related heterocycles possess various types of biological activities.

A good deal of importance is given to pyrazoline-5-one derivatives. It is due to their wide use in medicinal chemistry and some of them possess antituberculosis, antineoplastic, antidiabetic, antifertility and anti hydhydrhyroid activity.

In this chapter the general methods of synthesis of pyrazoles and Pyrazoline derivatives are presented three different headings.

1. Synthesis of Pyrazolines by hydrazine based reactions.
2. Synthesis of aryl azo heterocyclic Pyrazolines.

This chapter deals with a review of literature pertaining to previous synthetic approaches, and biological activity of azetidine-2-ones and thiazolidi-4-ones. The importance of azetidine-2-ones and thiazolidi-4-ones have been outlined.

CHAPTER – II: Brief literature survey

This chapter deals with introduction, previous synthetic approaches of novel manich bases,1,3,4 oxadiazole derivatives, sulphonamide derivatives, phosphonate derivatives, containing pyrazoline moity and phosphonates bearing carboxamide derivatives.
CHAPTER – III: Theoretical analysis

This chapter discusses the details of antimicrobial activity and their functioning, apparatus used, discuss about the preparation of solutions used, description of the instruments employed, general polarographic procedures and the basic principles involved in polarographic techniques are described and describes the basic principles involved in cyclic voltammetric technique and general cyclic voltammetric on experimental procedures.

CHAPTER-4: Experimental investigation

This chapter discuss with the experimental section of all synthetic compounds and their analytical data.

CHAPTER-5 – Results and discussion

Chapter-5.1: Synthesis of novel aryl hydrazono pyrazolin-5-one-isatin-mannich bases.

A review of literature concerning to the synthesis and biological activity of Mannich bases is depicted in the preceding pages of this chapter. The need for the present study has been outlined. The antibacterial activity of mannich bases has been well established. In view of these observations, it appeared of interest to synthesize some novel mannich bases bearing Aryl hydrazono pyrazole-5-one-indole moieties. In this chapter, we reported the synthesis and characterization of
1. Synthesis of ethyl 4, 4, 4-trifluoro-3-oxo-2-(4-phenylhydrazono) butanoate 3.

2. Ethyl 4-(5-oxo -4-(4-phenylhydrazono)-3-(trifluoromethyl)-4, 5-dihydro-1H-pyrazol-1-yl)benzoate 5

3. 4-(5-oxo-4-(4-phenylhydrazono)-3-(trifluoromethyl)-4, 5-dihydro-1H-pyrazol-1-yl)benzoic acid 6


5. (S)-methyl 2-(2-(4-(5-oxo-3-(trifluoromethyl)phenyl)hydrazono)-4,5-dihydro-1H-pyrazol-1-yl)benzamido) acetamido)propanoate 8


7. indoline-2, 3-dione 10

8. N-(2-oxo-2-(((S)-1-oxo-1-((Z)-2-((2-oxoindolin-3-ylidene)hydrazinyl)propan-2-yl)amino)ethyl)-4-(5-oxo-3-(trifluoromethyl)-4-(2-(4-(trifluoromethyl)phenyl)hydrazono)-4,5-dihydro-1H-pyrazol-1-yl)benzamide 11

CHAPTER 5.2: Synthesis of triazaspiro pyrazoline-5-one derivatives bearing 1,3,4-Oxadiazole and 4-oxoazetidine-1-yl moieties

In this chapter, the synthesis of 1,3,4-Oxadiazole pyrazoline-3-one bearing oxoazetidine were presented. In the preceding pages of this chapter, the general methods of synthesis and biological importance of 1,3,4-Oxadiazoles were briefly summarized. It is due to their wide use in medicinal chemistry and some of them possess antituberculosis, antineoplastic, antidiabetic, antifertility and antihydrhyroid activity. The need of the present work has been outlined. In view of these observations, it appeared of interest to synthesize some novel 1,3,4-oxadiazole system bearing aryl hydrazono pyrazole moiety possessing oxoazetidine ring. In this chapter, we described the synthesis and characterization of

1. synthesis of Ethyl 4-(3-chloro-2,5-dioxo-8-(trifluoromethyl)-1-((4- (trifluoromethyl) phenyl) amino)-1,6.7-triazaspiro[3,4]oct-7-en-6- yl) benzoate (2).

2. synthesis of 4-(3-chloro-2,5-dioxo-8-(trifluoromethyl)-1-((4- (trifluoromethyl) phenyl) amino) -1,6.7-triazaspiro[3,4]oct-7-en-6- yl) benzohydrazide (3)

3. synthesis of 4-(3-chloro-2,5-dioxo-8-(trifluoromethyl)-1-(4- (trifluoromethyl) phenyl amino)-1,6.7-triazaspiro[3,4]oct-7-en-6- yl)-N'-(3-methyl-5-oxo-4,5-dihydro-1H-pyrazol-4-yl)methylene) benzohydrazide (5)
4. synthesis of 4-(3-chloro-2,5-dioxo-8-(trifluoromethyl)-1-((4-(trifluoromethyl) phenyl)amino)-1,6,7-triazaspiro[3,4]oct-7-en-6-yl)-N-(3-chloro-2-(3-methyl-5-oxo-4,5-dihydro-1H-pyrazol-4-yl)-4-oxazetidin-1-yl)benzamide(6)

5. synthesis of ethyl 2-(4-(3-chloro-2,5-dioxo-8-(trifluoromethyl)-1-((4-(trifluoromethyl) phenyl)amino)-1,6,7-triazaspiro[3,4]oct-7-en-6-yl)benzamido)-4-oxazetidin-2-yl)-3-methyl-5-oxo-4,5-dihydro-1H-pyrazol-1-yl)acetate(7)

6. synthesis of 4-(3-chloro-2,5-dioxo-8-(trifluoromethyl)-1-((4-(trifluoromethyl) phenyl)amino)-1,6,7-triazaspiro[3,4]oct-7-en-6-yl)-N-(3-chloro-2-(1-(2-hydrazinyl-2-oxoethyl)-3-methyl-5-oxo-4,5-dihydro-1H-pyrazol-4-yl)-4-oxazetidin-1-yl)benzamide(8)

7. synthesis of 4-(3-chloro-2,5-dioxo-8-(trifluoromethyl)-1-((4-(trifluoromethyl) phenyl)amino)-1,6,7-triazaspiro[3,4]oct-7-en-6-yl)-N-(3-chloro-2-(1-((5-(4-substituted phenyl)-1,3,4-oxadiazol-2-yl)methyl)-3-methyl-5-oxo-4,5-dihydro-1H-pyrazol-4-yl)-4-oxazetidin-1-yl)benzamide(9)

CHAPTER 5.3: Triazaspiro-pyrazoline-5-one derivatives containing 1,3,4-thiadiazole sulphonamides

Synthesis, and characterization of Aza-spiro pyrazoline-5-ones bearing thiazolidines were incorporated in this chapter.

In this chapter, we have reported the synthesis and characterization of N-(5-((4-(3,6-dioxo-9-(trifluoromethyl)-4-(trifluoromethyl)phenyl)amino)-1-thia-4,7,8-triazaspiro[4.4]non-8-en-
The synthetic route was depicted in the scheme 5.1. We have reported the synthesis and characterization of

1 Synthesis of ethyl 4,4,4-trifluoro-3-oxo-2-(2-(4-(trifluoromethyl)phenyl)hydrazono) butanoate ethyl 2-(4-(5-oxo-3-(trifluoromethyl)-4-(2-(4-(trifluoromethyl)phenyl)hydrazono)-4,5-dihydro-1H-pyrazol-1-yl)phenoxy)acetate(3)

2 Synthesis of ethyl 2-(4-(5-oxo-3-(trifluoromethyl)-4-(2-(4-(trifluoromethyl)phenyl)hydrazono)-4,5-dihydro-1H-pyrazol-1-yl)phenoxy)acetate(5)

3 Synthesis of 1-(4-hydroxyphenyl)-3-(trifluoromethyl)-4-(2-(4-(trifluoromethyl)phenyl)hydrazono)-1H-pyrazol-5(4H)-one(6)

4 Synthesis of ethyl 2-(4-(3,6-dioxo-9-(trifluoromethyl)-4-((4-(trifluoromethyl)phenyl)amino)-1-thia-4,7,8-triazaspiro[4.4]non-8-en-7-yl)phenoxy)acetate(7)

5 Synthesis of 2-(4-(3,6-dioxo-9-(trifluoromethyl)-4-((4-(trifluoromethyl)phenyl)amino)-1-thia-4,7,8-triazaspiro[4.4]non-8-en-7-yl)phenoxy)acetic acid(8)

6 Synthesis of ethyl 2-(4-(3,6-dioxo-9-(trifluoromethyl)-4-((4-(trifluoromethyl)phenyl)amino)-1-thia-4,7,8-triazaspiro[4.4]non-8-en-7-yl)phenoxy)acetate(8)

7 Synthesis of 7-(4-((5-amino-2,5-dihydro-1,3,4-thiadiazol-2-yl)methoxy)phenyl)-9-(trifluoromethyl)-4-((4-
(trifluoromethyl)phenyl)amino)-1-thia-4,7,8-triazaspiro[4.4]non-8-ene-3,6-dione (9)

8 Synthesis of (5-((4-(3,6-dioxo-9-(trifluoromethyl)-4-((4-(trifluoromethyl) phenyl)amino)-1-thia-4,7,8-triazaspiro[4.4]non-8-en-7-yl)phenoxy)methyl)-2,5-dihydro-1,3,4-thiadiazol-2-yl)sulfamoyl chloride (10)

9 Synthesis of N-(5-((4-(3,6-dioxo-9-(trifluoromethyl)-4-((4-(trifluoromethyl) phenyl)amino)-1-thia-4,7,8-triazaspiro[4.4]non-8-en-7-yl)phenoxy)methyl)-2,5-dihydro-1,3,4-thiadiazol-2-yl)piperidine-1-sulfonamide (12a-f)

CHAPTER 5.4: Synthesis of Pyrazoline-5-one phoshonate derivatives bearing pyrazoline-5-one moiety

In this chapter, the synthesis of phoshonate derivatives bearing pyrazolone moiety are presented. A review of literature concerning to the synthesis and biological activity of phoshonate is depicted in the preceding pages of this chapter. The need for the present study has been outlined. This chapter consists of synthesis and characterization of

1. Ethyl 2-(3-methyl-5-oxo-4-((4'-substituted phenylimino)methyl)-4,5-dihydro-1H-pyrazol-1-yl)acetate (4a-d)

2. 2-(3-methyl-5-oxo-4-((4'-substituted phenyl imino)methyl)-4,5-dihydro -1H-pyrazol-1-yl)acetohydrazide (5a-d)
3. 3-methyl-1-(2-oxo-2-(5-oxo-3-(trifluoromethyl)-4,5-dihydro-1H-pyrazol-1-yl)ethyl)-4-((4′-substituted phenyl imino)methyl)-1H-pyrazol-5(4H)-one (6a-d).

Diethyl ((3-methyl-5-oxo-1-(2-oxo-2-(5-oxo-3-(trifluoromethyl)-4,5-dihydro-1H-pyrazol-1-yl) ethyl) 4,5-dihydro-1H-pyrazol-4-yl)(4′-substituted phenyl amino)methyl)phoshonate (7a-d).

CHAPTER 5.5: Studies towards the Synthesis of Pyrazoline-5-onephosfonates derivatives of carboxamides

In this chapter, the synthesis and characterization of phoshonate derivatives containing pyrazolone moiety bearing piperidine/ morpholine/thiomorpholine/ N-methyl piperizine/ aniline were presented. A review of literature concerning to the synthesis and biological activity of carbamido derivatives were depicted in the preceding pages of this chapter. The need for the present study has been outlined. This chapter consists of synthesis and characterization of

1. Ethyl2-(3-methyl-5-oxo-4-(((4-(trifluoromethyl)phenyl)imino)methyl) -4,5-dihydro-1H-pyrazol-1yl)acetate(4)
2. 2-(3-methyl-5-oxo-4-(((4-(trifluoromethyl)phenyl)imino)methyl)-4,5-dihydro -1H-pyrazol-1-yl) acetic acid (5)
3. 2-(3-methyl-5-oxo-4-(((4-(trifluoromethyl)phenyl)imino)methyl)-4,5-dihydro -1H-pyrazol-1-yl) acetyl azide (6).
4. N-(3-methyl-5-oxo-4-(((4-(trifluoromethyl)phenyl)imino)methyl)-4,5-dihydro -1H-pyrazol-1-yl)
methyl)piperidine/morpholine/thiomorpholine/N-methyl piperizine/aniline-1-carboxamide(8a-e).

5. Ethyl methyl((3-methyl-5-oxo-1-
((piperidine/morpholine/thiomorpholine/N-methyl piperizine/aniline -1-carboxamido)methyl)-4,5-dihydro -1H-
pyrazol-4-yl) ((4-(trifluoromethyl)phenyl)amino)methyl) phosphonate(9a-e).

CHAPTER-5.6: Antimicrobial studies of novel tri-substituted pyrazoline-5-ones

All the synthesized compounds were screened for antimicrobial studies against antibacterial and antifungal activity by disc diffusion method and MIC by serial dilution method. For the prepared compounds using amoxicillin and cefaclor as references were subjected to preliminary antibacterial screening by disc diffusion method against Staphylococcus aureus NCCS 2079, Bacillus cereus NCCS2106 (gram positive) and Escherichia coli NCCS2065(gram negative). The antifungal studies were carried out against Aspergillus niger 1196, Candida albicans NCCS2106.

In chapter 5.1, We have reported the synthesis of N-(2-((R)-1-((Z)-2-(1-
((4-methylpiperazin-1-yl)methyl)-2-oxoindolin-3-ylidene)hydrazinyl)-1-
oxopropan-2-ylamino)-2-oxoethyl)-4-(5-oxo -4-(4-phenylhydrazono)-3-
(trifluoromethyl)-4, 5-dihydro-1H -pyrazol-1-yl)benzamide12a-h exhibit moderate anti-bacterial activity against the tested organisms at the concentration of 250µg/ml. The pyrazoline-5-one manich bases containing nitro (12f,12h), trifluoromethyl(12b,12g) showed
more activity against tested organismsthan other substituted compounds. similar trends are also observed in anti-fungal studies.

In chapter 5.2, Pyrazole-5-one system bearing 1,3,4-oxadiazole and oxoazetidine (9a-h) synthesis were reported in chapter 5.2. The pyrazoline-5-one heterocycles containing nitro (9g),CF₃(9h), Chloro(9e), and Bromo(9f) showed moderate activity against *Staphylococcus aureus* NCCS 2079, *Bacillus cereus* NCCS 2106 and *Escherichia coli* NCCS 2065 at the concentration of 250µg/ml. Similar trends were also noticed against fungal activity of *Aspergillus Niger* NCCS 1196 ,*Candida albicans* NCCS 2106.

In chapter 5.3,Azaspiro-oxoazetidine-pyrazoline-5-one containing sulphonyl carboxamido1,2,4-thiadiazoles (12a-f) reported in the chapter-5.3 exhibit good anti-bacterial activity against *Staphylococcus aureus* NCCS 2079,*Bacillus cereus* NCCS 2106(gram positive) and *Escherichia coli* NCCS 2065 at the concentration of 250µg/ml. The system containing thiomorpholino(12c), morpholino(12h) and nitroimidazole(12f) exhibit more conductivity than other substituted compounds. The similar observations were also noticed in Azaspiro-thioazotidinone-pyrazoline-5-one bearing sulphonyl carboxamido 1,2,4-thiadiazoles (12a-f)

In chapter 5.4, Diethyl phosphonates bearing pyrazoline-5-ones(7a-d) synthesis reported in chapter 5.4 were subjected to antimicrobial profile. The organo phosphorous compounds(7a-f) exhibit good antimicrobial activity against *Staphylococcus aureus* NCCS 2079,*Bacillus cereus* NCCS 2106(gram positive) and *Escherichia*
coli NCCS 2065 at the concentration of 250µg/ml. The diethylphosphonates containing nitro(7c), trifluoromethyl(7d) and chloro(7b) exhibit more activity than other substituted compounds. Similar trends were reported in antifungal activity.

In chapter 5.5, Diethyl phosphonates bearing pyrazoline-5-one-carboxamido derivatives(9a-e) synthesis presented in chapter 5.5 exhibit moderate antimicrobial activity against *Staphylococcus aureus* NCCS 2079, *Bacillus cereus* NCCS 2106 (gram positive) and *Escherichia coli* NCCS 2065 at the concentration of 250µg/ml. The diethylphosphonates carboxamido derivatives processing thiomorpholino(7c), morphilino(7d) and pyrrolidine(7b) exhibit more activity than other carboxamido derivatives. Similar trends were also noticed in antifungal activities.

**Chapter 5.7:** Polarographic behaviour of 3-methyl-1-(morpholinomethyl)-4-(2-(4’-substituted aryl hydrazono)-1H-pyrazol-5(4H)-one(4a-e)

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<tr>
<td>I. 4a</td>
<td>H</td>
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<tr>
<td>II. 4b</td>
<td>4'1-CH₃</td>
</tr>
<tr>
<td>III. 4c</td>
<td>4'1-OCH₃</td>
</tr>
<tr>
<td>VI. 4d</td>
<td>4'1-Cl</td>
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<tr>
<td>V. 4e</td>
<td>4'1-Br</td>
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Polarographic behavior of 4a-e

The Polarographic studies of 4a-e were carried out in Britton-Robinson buffer solution of pH 2.10-10.10 Containing 40% aqueous dimethyl formamide. The 3-methyl-1-(morpholinomethyl)-1H-pyrazol-5(4H)-one(A) was synthesized and characterized by spectral studies and elemental analysis. The pyrazoline-5-one(A) polarographic behavior was studied in Britton Robinson buffer solution of pH 2.10-10.10 Containing 40% aqueous dimethyl formamide. The polarographic behavior of pyrazoline-5-one(A) reveal that it fails to exhibit polarographic reduction wave under the experimental conditions studied. This is probably due to the stabilization of the pyrazoline-5-one ring by keto enol tautomerism.

This observation therefore, unequivocally suggests that polarographic reduction waves noticed for 4a-e under experimental conditions are due to electron reduction of exo-cyclic hydrogeno group(-NH-N=) in keto form of 4a-e.

The arylhydrazonopyrazoline-5-ones 4a-e exhibit one polarographic wave in acidic buffer solutions of pH 2.10-6.10 besides only one wave is noticed in buffer solution of pH 7.10. In alkaline buffer solutions of pH 8.10-10.10 (pH >PKa) the compounds 4a-e exhibit two waves.
The half wave potentials(-E$_{1/2}$) of 4a-e increases with increases in the pH of the solution. This observation is attributed to involvement of protons in electro-reduction process. However, the half wave potential(-E$_{1/2}$) remain constant in alkalin buffer solutions.

The limiting current of the wave decreases with increase in pH, while the half-wave potential shifts to more negative values with increase in pH of the medium. The diffusion current shows a linear relationship with the concentration of the deplolariser. Semi-logarithmic analysis of the wave reveals that the electrochemical reduction process is irreversible. The irreversible nature of the wave is also confirmed by the low values of the heterogenous formal rate constant ($K_{0h}$) and the high values of activation free energy ($\Delta G^*$).

The milli-coulumetric studies proposed by devrier-kroon is carried out on 1-(morpholinomethyl)-3-methyl-4-arylhydrazono-pyrazoline-5-one (4a) is carried in britton robinson buffer solutions of pH 4.10 containing 40% aqueous dimethyl formamide.

The milli-coulumetric data of 4a revealed that four electrons are involved in electro reduction of polarographic wave noticed in the acidic buffer solution of pH 2.10-10.10. However, the experimental results reveal that in alkaline solution 4-electrons and 2-electrons are involved in the electro reduction of first and second polarographic waves respectively.

The controlled potential electrolysis is carried out with 1-(morpholinomethyl)-3-methyl-4-arylhydrazono-pyrazoline-5-one (4a) is carried in britton robinson buffer solutions of pH 4.10 and pH 8.10
containing 40% aqueous dimethyl formamide at an applied voltage of 1.25V and 1.70V respectively.

The electrode-products have been characterized by spectral analysis (IR, \(^1\)H-NMR, and \(^{13}\)C-NMR) and elemental analysis, they are identified as (a) Aniline(III) (b) \(1\)-\((\text{morpholinomethyl})\)-3-\((\text{methyl})\)-4-\((\text{arylhydrazono})\)-pyrazoline-5-one(IV) (c) Phenylhydrazine and (d) \(1\)-\((\text{morpholinomethyl})\)-4-\((\text{hydroxypyrazoline})\)-5-one(V). Based on the polarographic, milli-coulumetric and controlled potential studies, aplausible mechanism has been suggested for electro reduction of 4a-e.

**Chapter 5.8: Cyclic voltammetric behavior of 3-\((\text{methyl})\)-1-\((\text{morpholinomethyl})\)-4-\((2\text{-}4\text{'-substituted aryl hydrazono})\)-1\((\text{H})\)-pyrazol-5(4\((\text{H})\))-one 4a-e with handing mercury drop electrode (HMDE).**

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<tr>
<td>I. 4a</td>
<td>H</td>
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<tr>
<td>II. 4b</td>
<td>4(^1)-(\text{CH}_3)</td>
</tr>
<tr>
<td>III. 4c</td>
<td>4(^1)-(\text{OCH}_3)</td>
</tr>
<tr>
<td>VI. 4d</td>
<td>4(^1)-(\text{Cl})</td>
</tr>
<tr>
<td>V. 4e</td>
<td>4(^1)-(\text{Br})</td>
</tr>
</tbody>
</table>
Cyclic voltammetric studies of 4a – e were carried out in Britton-Robinson buffer solution of \( pH \) 2.10-10.10 Containing 40% aqueous dimethyl formamide. The arylhydrazono pyrazoline-5-ones 4a-e exhibit two cyclic voltammograms in acidic buffer solution of \( pH \) 2.10-10.10 in the place of single polarographic reduction wave noticed in DC-polarography. However, it exhibit two cyclic voltammograms in alkaline buffer solution of \( pH \) 8.10-10.10 in the place of single polarographic reduction wave noticed in DC-polarography.

The compounds fail to exhibit the anodic peak in buffer solutions of \( pH \) 2.10-10.10. But at the same time the pyrazoline-5-ones 4a-e exhibit an inverted peak in acidic buffer solutions of \( pH \) 2.10-6.10. In general, the results of cyclic voltometric studies substantiate the results observed in DC-polarographic studies. The CVM- technique is useful technique in the present studies to detect the short-lived intermediate formed in electro-reduction of 4a-e in acidic buffersolutions.

**CHAPTER-6: SUMMARY AND CONCLUSIONS**

The important conclusions drawn from the research investigation have been incorporated under the separating heading Summary and Conclusions.