Summary

Results of explanatory research are recorded in this thesis. The thesis comprises of five following chapters:

Chapter 1: Synthesis of pyrazolones and phthalazindiones

Chapter 2: Synthesis of isoxazole and dihydroisoxazoles

Chapter 3: Synthesis of oxadiazothiones, triazothiones and triazolothiadiazines

Chapter 4: Synthesis of hydrazono-isatin and N-methyl isatin derivatives

Chapter 5: Synthesis of oxazolines and thiazolines

The research work carried out is based on the synthesis of heterocyclic derivatives of selected fatty acids. All the synthesized compounds were characterized by IR, ¹H NMR, ¹³C NMR and mass spectra (MS). Further, synthesized compounds were tested for their biological activities.

CHAPTER 1: SYNTHESIS OF PYRAZOLONES AND PHTHALAZINDIONES

The novel series of long alkyl, alkenyl and hydroxyalkenyl chain substituted derivatives of pyrazolones and phthalazindiones, (26-34) have been synthesized by using selected fatty acid hydrazides (21-25), (Scheme 1). Products obtained in good yield and purified by column chromatography.

All the compounds were screened for their \textit{in vitro} antimicrobial activity against the assortment of two gram-positive bacteria, \textit{Staphylococcus aureus} SA 22, \textit{Bacillus subtilis} MTCC 121; two gram-negative bacteria, \textit{Escherichia coli} K12, \textit{Pseudomonas aeruginosa} and fungal strain, \textit{Candida albicans} IOA 109. All compounds showed moderate to good inhibitory activity against the tested organisms but compounds (33 and 34) were found to be more potent antimicrobial agents.
Summary

Scheme 1: Synthesis of 1,3-disubstituted-1H-pyrazol-5(4H)-ones and 2-substituted-3H-1,4-phthalazindiones
CHAPTER 2: SYNTHESIS OF ISOXAZOLE AND DIHYDROISOXAZOLES

The novel 3,5-disubstituted isoxazole (69) has been synthesized by 1,3-dipolar cycloaddition of nitrile oxide to undec-10-ynoic acid (66), (Scheme 2). Nitrile oxide was generated \textit{in situ} from 1-nitrobutane (67) employing 1,4-phenylene diisocyanate (68) as dehydrating agent. Similarly, one-pot synthesis of 3,5-disubstituted-4,5-dihydroisoxazole (74) and 3,4,5-trisubstituted-4,5-dihydroisoxazoles (75-77) possessing ester moiety is reported, (Scheme 3) using \textit{in situ} generated nitrile oxide and fatty acid esters (70-73). All the newly synthesized compounds were purified by column chromatography and characterized by spectral data.

\[
\begin{array}{c}
\text{COOH} + \text{NO}_2 + \text{OCN} \\ \text{(66)} \quad \text{(67)} \quad \text{(68)} \\
\text{Et}_3\text{N, dry THF} \\
\downarrow \\
\text{COOH} \\
\text{(69)}
\end{array}
\]

Scheme 2: Synthesis of 3,5-disubstituted isoxazole

The synthesized compounds (69, 74-77) were screened for their \textit{in vitro} antibacterial activity against different (clinical isolates) bacterial strains by disc diffusion method.

The susceptibility was assessed on the basis of the diameter of zone of inhibition against gram-positive and gram-negative strains of bacteria. Inhibition zones were measured and compared with the controls.

Synthesized compounds (69, 74-77) were also screened for their \textit{in vitro} antifungal activity against a panel of standard and clinical species of \textit{Candida}. The results of
antimicrobial screening showed that compounds (76 and 77) exhibit excellent antimicrobial activity nearly equivalent to the control compounds.

Scheme 3: Synthesis of 3,5-disubstituted and 3,4,5-trisubstituted-4,5-dihydroisoxazoles
CHAPTER 3: SYNTHESIS OF OXADIAZOLTHIONES, TRIAZOLTHIONES AND TRIAZOLOTHIADIAZINES

This chapter deals with the synthesis of long alkenyl/hydroxyalkenyl chain substituted 1,3,4-oxadiazol-2-thione (108-111) and 1,2,4-triazol-3-thione (112-115) derivatives using long alkenyl/hydroxyalkenyl chain hydrazides (22-25) as cheap starting material, (Scheme 4). Products were purified by column chromatography and characterized by IR, $^1$H NMR, $^{13}$C NMR and mass spectral data.

Compounds (112-115) were transformed into 4-amino-5-long chain alkenyl/hydroxyalkenyl-6-phenyl-7H-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazines (116-119) derivatives by ring closure reaction using phenacyl bromide as reagent, (Scheme 5). Structure elucidation of these compounds is based on spectral data.

Scheme 4: Synthesis of 5-substituted-1,3-4-oxadiazol-2-thiones and 4-amino-5-substituted-1,2,4-triazol-3-thiones
Scheme 5: Synthesis of 3-substituted-6-phenyl-7H-1,2,4-triazolo[3,4-b]-1,3,4-thiadiazines

Furthermore, synthesized compounds were tested for in vitro anticancer activity against PBMCs (peripheral blood mononuclear cells) and three different human cancer cell lines by MTT [3-(4,5-dimethylthiazolyl-2)-2,5-diphenyltetrazolium bromide] assay. The results of these studies showed that compounds (118 and 119)
were found to be most promising anticancer agents among all the newly synthesized compounds.

**CHAPTER 4: SYNTHESIS OF HYDRAZONO-ISATIN AND N-METHYL ISATIN DERIVATIVES**

The novel long alkyl, alkenyl, hydroxyalkenyl chain substituted hydrazono-isatin (144-148) and N-methyl isatin derivatives (149-153) were achieved by dehydrative condensation reaction of isatin/N-methyl isatin with fatty acid hydrazides (21-25) using ethanol as solvent containing catalytic amount of glacial acetic acid, (Scheme 6). Products were purified by column chromatography. The structures of all the newly synthesized compounds were elucidated by spectral data.

Further, these compounds were evaluated for *in vitro* cytotoxicity against four different human cancer cell lines by MTT assay. Among all the tested compounds, compounds (148 and 153) were found to be most potential cytotoxic agents. Apoptosis induced by these two compounds was also proved by PARP [poly(ADP-ribose)polymerase] cleavage studies done by western blotting technique and FACS (fluorescence-activated cell sorting) assay results.

\[
\text{R-CONHNH}_2 \quad + \quad \text{(21-25)}
\]

\[
\text{EtOH, Cat. AcOH, } \Delta \quad \rightarrow \quad \text{R'}: \text{H, CH}_3
\]

\[
\text{(120, 143)}
\]

\[
\text{NNHCOR}
\]

\[
\text{N}
\]

\[
\text{(144-153)}
\]

\[
\text{(144-148), R'}: \text{H; (149-153), R'}: \text{CH}_3
\]
Summary

### Compound Codes

<table>
<thead>
<tr>
<th>Compound</th>
<th>Codes</th>
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<tr>
<td>21, 144, 149</td>
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<td>![Structure 2]</td>
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<td>![Structure 4]</td>
</tr>
<tr>
<td>25, 148, 153</td>
<td>![Structure 5]</td>
</tr>
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</table>

**Scheme 6: Synthesis of novel long chain hydrazono-isatin and N-methyl isatin derivatives**

### CHAPTER 5: SYNTHESIS OF OXAZOLINES AND THIAZOLINES

The long chain dibromo derivatives (179-182) of fatty acid esters were used as starting material and these on reaction with urea and thiourea gave their corresponding 2-amino-5-substituted-1,3-oxazoline (183), 2-amino-4,5-disubstituted-1,3-oxazolines (184-186) and 2-amino-5-substituted-1,3-thiazoline (187), 2-amino-4,5-disubstituted-1,3-thiazolines (188-190), respectively, (Scheme 7). Products were purified by column chromatography. The newly synthesized compounds were characterized on the basis of their spectral data.

All the newly synthesized compounds were tested for their antimicrobial activity by disk diffusion assay and minimum inhibitory concentration (MIC) by broth micro dilution method against the representative panel of gram-positive and gram-negative bacteria. Also, compounds were tested for their inhibitory action against different fungal strains. The investigation of antimicrobial screening revealed that compounds...
(185, 186, 189 and 190) were found to be better antimicrobial agents among all the synthesized compounds.

\[
\begin{align*}
\text{NH}_2\text{CONH}_2 & \quad \text{MeOH, reflux} \\
\text{MeOH, \ reflux} & \quad \text{NH}_2\text{CSNH}_2
\end{align*}
\]

\[
\begin{align*}
\text{Scheme 7: Synthesis of 2-amino-5-substituted and 2-amino-4,5-disubstituted oxazolines and thiazolines}
\end{align*}
\]