The thesis entitled “DESIGN, SYNTHESIS AND BIOLOGICAL EVALUATION OF NEW AZOLE HETEROCYCLES” has been divided into four chapters.

CHAPTER I: This chapter describes the introduction, natural abundance and biological importance of azoles i.e. benzoazoles, benzoxazolones and 1,2,4-triazoles and cyclic amines i.e. piperizines and piperidines.

CHAPTER II: This chapter is divided into two sections; Section A and Section B.

SECTION A: This section describes the synthesis and biological evaluation of 2-cyclic amine substituted benzoazoles.

SECTION B: This section describes the synthesis and biological evaluation of 3-substituted-2(3H)- benzoxazolones.

CHAPTER III: This chapter describes the synthesis and biological evaluation of 3-mercapto / -thione- substituted-1,2,4-triazoles.

CHAPTER IV: This chapter describes the stereoselective synthesis of 2,4-disubstituted piperidines.
CHAPTER-I

Introduction

Human health is impacted by a large variety of chemical substances, including those essential to human life, such as vitamins and nutrients and medicines. Natural substances are intrinsically exhibit superior properties with regard to efficacy and safety in matters related to human health. As it is difficult to meet the worldwide demand of the requirement of the natural products due to their low abundance in nature, it is essential to produce synthetic substances in large quantities. This can extend to so-called nature-identical materials that are natural substances produced synthetically in an identical or with slight modification of the molecular form, in order to increase the biological activity of the molecule.

Heterocycles are among the most frequently encountered scaffolds in drugs and pharmaceutically relevant substances. The remarkable ability of heterocyclic nuclei to serve both as biomimetics and reactive pharmacophores has largely contributed to their unique value as traditional key elements of numerous drugs. The development of heterocycles as scaffolds, containing a high degree of diversity has become a leading focus in modern drug discovery. In this research program the derivatives of various diverse classes of heterocycles e.g., benzoxazole, benzoazolone, triazole, piperazine and piperidine were developed. Certain possible modifications on the heterocyclic ring by the addition of diverse substituents may lead to new products with better biological profiles. As a result of the biological activity exhibited by the heterocyclic molecules, the development of new chemical entities (NCEs) is the focus of intense activity in pharmaceutical industry. Various types of newly synthesized molecules may be very good at blocking the action or killing the pathogens without harming the human cells so as to prevent or cure the disease. The nitrogen, oxygen and sulphur heterocycles are an attractive source of compounds for the identification of new biological probes. The main aim is to design and synthesize molecules involving the use of structural motif commonly found in majority of well-established drug molecules. The research work is mainly concentrated on the above points.
Benzoxazoles
Substituted benzoxazole derivatives and their analogues such as benzimidazoles and benzothiazoles have been the aim of researchers for many years, because they constitute an important class of heterocyclic compounds. Benzoxazole form core of a variety of cytotoxic natural products. Benzoxazole derivatives are biologically significant compounds and known to exhibit various biological activities such as anticancer, antimicrobial, anti HIV and dopamine D4 agonists.

Benzoxazolones
2(3H)-Benzoxazolone and its bioisosters are considered ‘privileged scaffolds’ in the design of pharmacological probes. Various derivatives of 2(3H)-benzoxazolone have been marketed as drug like Benzolone (myorelaxant), Paraflex or Chlorzoxazone (sedative analgesic) and Vinizene (topical antiseptic). 2(3H)-Benzoxazolone derivatives have been reported with various biological activities like anti-inflammatory, analgesic, antitubercular, antimalarial, anticancer, anticonvulsant, anti-HIV, antinociceptive, antioxidant, 5-HT antagonist, PPARγ agonist and antimicrobial.

1,2,4-Triazoles
In the last few decades, the chemistry of 1,2,4-triazoles and their fused heterocyclic derivatives has received considerable attention owing to their synthetic and effective biological importance. A large number of 1,2,4-triazole-containing ring system have been incorporated into a wide variety of therapeutically interesting drug candidates. The mercapto / thione-substituted-1,2,4-triazoles have been well studied and so far a variety of biological activities have been reported for a large number of their derivatives, such as antitubercular, antimycobacterial, anticancer, antibacterial, antifungal, diuretic and hypoglycemic properties.

Piperazines
The piperazine scaffold has been classified as a privileged structure and is frequently found in biologically active compounds across a number of different therapeutic areas. This motif is found in drug candidates displaying anti-allergenic, antibacterial, anti-anxiety, anti-emetic and antimigraine activity.
Piperidines

The piperidine ring is a ubiquitous structural feature of numerous naturally occurring alkaloids. They are also being often embedded within scaffolds recognized as privileged structures by medicinal chemists. Substituted piperidines are appear in many drugs and drug candidates. Compounds with the piperidine sub-structure exhibit anti-hypertensive, antibacterial, anticonvulsant, anti-inflammatory and anti-proliferative activities.

CHAPTER-II

SECTION A: Synthesis and biological evaluation of 2-cyclic amine substituted benzoxazoles

Chemistry

In this section, we are reporting the application of zinc dust as a reusable reagent in synthesis of 2-cyclic amine benzoxazoles under microwave irradiation conditions in the absence of a solvent. The starting material 2-chlorobenzoxazole (4), is commercially available chemical and also otherwise was prepared starting from 2-nitrophenol (1). The 2-nitrophenol (1) was subjected to hydrogenation under Pd-C in ethanol to obtain 2-aminophenol (2). The 2-aminophenol (2) was treated with carbon disulfide under basic conditions to obtain 2-mercapto-1,3-benzoxazole (3). 2-Mercapto-1,3-benzoxazole (3) was reacted with phosphorus pentachloride (PCl₅) to obtain 2-chlorobenzoxazole (4) (Scheme 1). The formation of 4 was confirmed by ¹H NMR, IR and mass spectral data.

\[
\begin{align*}
1 & \xrightarrow{(i) \text{H}_2/\text{Pd-C, Ethanol, 24 h}} 2 & \xrightarrow{(ii) \text{CS}_2, \text{KOH, Ethanol, Reflux, 8 h}} 3 & \xrightarrow{(iii) \text{PCl}_5, \text{dry Toluene, 3 h}} 4
\end{align*}
\]

Scheme 1

The 2-chlorobenzoxazole was treated with various cyclic amines under conventional and microwave irradiation conditions. In the conventional procedure 2-chlorobenzoxazole was treated with various cyclic amines in acetonitrile at 0°C-rt to
obtain the 2-cyclic amine benzoxazoles (5a-l) (Scheme 2) for an appropriate time (Method A). The formation of the products was confirmed by $^1$H NMR, $^{13}$C NMR, IR and mass spectroscopy. The presence of zinc dust did not improve the reaction time and yield of the product under conventional condition. The product was obtained with less purity in low yield when the reaction was carried out under microwave irradiation in the absence of zinc dust. The microwave irradiation experiments were carried out using focused microwave instrument Discover (CEM) model at 2450 MHz frequency and 450 watts power. Improvement in yields was not obtained even by increasing the irradiation time and voltage.

![Scheme 2](image_url)

Reagents: (i) Method A: Acetonitrile, 0°C-rt

X = N-methyl, N-ethyl, N-benzyl, N-phenyl, N-pyridyl, N-pyrimidyl, N-(3-chlorophenyl), N-(2-hydroxyethyl), CH$_2$, O.

entry 9: 2-(3-methylpiperazin-1-yl)-1,3-benzoxazole.

entry 10: 2-(1,4-Diazepan-1-yl)-1,3-benzoxazole.

On other hand, when the reaction was carried out under microwave conditions in the presence of activated zinc, the product was obtained in excellent yield with high purity and the reaction time has decreased from minutes to seconds. This is a remarkable achievement over the existing conventional method. It was also investigated for the possibility of zinc dust in catalytical or less than stoichiometric quantities. The high yields of the products with high purity were obtained with one equivalent of zinc dust. A mixture of 2-chlorobenzoxazole and cyclic amine was subjected to microwave irradiation in the presence of zinc dust (Scheme 3) for an appropriate time (Method B). The reaction is remarkably fast and led to very good yields of the products with high purity. The yields and the reaction time under conventional and microwave conditions were compared.
Reagents: (i) Method B: Zinc dust, Microwave, 450 W

\[ X = N\text{-}\text{methyl}, \, N\text{-}\text{ethyl}, \, N\text{-}\text{benzyl}, \, N\text{-}\text{phenyl}, \, N\text{-}\text{pyridyl}, \, N\text{-}\text{pyrimidyl}, \, N\text{-}(3\text{-}\text{chlorophenyl}), \]

entry 9: 2-(3-methylpiperazin-1-yl)-1,3-benzoazole.
entry 10: 2-(1,4-Diazepan-1-yl)-1,3-benzoazole.

Scheme 3

Reusability of zinc dust under microwave conditions was also studied. The zinc dust was reactivated and used for subsequent runs. It has shown nearly same activity after each use. This makes the process more economical.

The significant feature of this method is the isolation of the pure product by simple work up in a short reaction time with high purity.

**Biology**

The synthesized 2-cyclic amine benzoxazoles were evaluated for anticancer, antibacterial, antifungal and anti-inflammatory activities.

**Anticancer activity**

Among the compounds (n=9) evaluated for their cytotoxic potential, eight entries showed dose dependent inhibition of cancer cell proliferation (IC\(_{50}\) values are of less than 100 \(\mu\)M). The results indicate that these compounds are comparatively more effective on human breast cancer (MCF-7) and lung cancer cells (A549) than cervical cancer (HeLa) and colon cancer (SW-480) cells. From the cytotoxicity results, it is evident that cytotoxicity of the benzoxazole is increasing when substituted cyclic amine is incorporated at its 2\(^{\text{nd}}\) position.

**Antibacterial activity**

All the twelve synthesized compounds (5a-l) were evaluated for antibacterial activity. The minimum inhibitory concentrations (MIC) of synthesized compounds were tested against three representative Gram-positive organisms and Gram-negative organisms.
Among the synthesized compounds (5a-l), only 5h, 5j and 5l with N-(2-hydroxyethyl) group, NH and O respectively at the 4th position of cyclic amine showed moderate antibacterial activity while other compounds were inactive.

**Antifungal activity**

All the twelve synthesized compounds (5a-l) were evaluated for antifungal activity. In vitro antifungal activity of the synthesized compounds was studied against the four fungal strains. Among the synthesized compounds 5a-l, only 5b and 5c with N-ethyl group and N-benzyl group respectively at the 4th position of cyclic amine and 5k with piperidine as cyclic amine showed moderate antifungal activity while other compounds were inactive.

**Anti-inflammatory activity**

The synthesized compounds 5a-l were subjected to carrageenan induced oedema in hind paw of rats as an assay for anti-inflammatory drugs. Among tested compounds, moderate anti-inflammatory activity was exhibited by 5a and 5g with N-methyl group and N-chloro phenyl group respectively at the 4th position of cyclic amine. Other compounds have shown very less or no activity.

The antibacterial, antifungal and anti-inflammatory activities results evidence that larger groups at 4th position of cyclic amine have no significant contribution to these activities of the compounds.

**CHAPTER-II**

**SECTION B: Synthesis and biological evaluation of 3-substituted-2(3H)-benzoxazolones**

- Chemistry
- Zinc dust mediated alkylation of cyclic amines

As a part of our ongoing research work on the synthesis of bio molecules, our group has developed environmentally benign, economical and reusable catalyst for alkylation and sulfonylation of amines using zinc dust as the reaction medium. In the present
work, we adapted the same methodology for alkylation reactions and the details are illustrated as follows.

Various 5-chloro-3-substituted-1,3-benzoxazol-2(3H)-ones (4a-f) and 5-chloro-3-[3-(cyclic amine)propyl]-1,3-benzoxazol-2(3H)-ones (6a-f) were synthesized starting from commercially available 2-amino-4-chlorophenol (1).

5-Chloro-2(3H)-benzoxazolone (2) was synthesized by the reaction of 2-amino-4-chlorophenol (1) and urea in concentrated HCl at 140-170°C (Scheme 1). 5-Chloro-2(3H)-benzoxazolone (2), K₂CO₃ and various alkyl bromides (3a-f) were reacted in acetonitrile at ambient temperature to obtain the N-alkylated products i.e. 5-chloro-3-substituted-1,3-benzoxazol-2(3H)-ones (4a-f) as shown in the Scheme 1. The N-alkylation was confirmed by the two observations: (1) The 5-chlorobenzoxazolone (2) carbonyl stretching (ν C=O, 1771 cm⁻¹) was retained in the IR spectrum of 4a (ν C=O, 1774 cm⁻¹). (2) The ¹³C chemical shift value for the carbonyl carbon (δ = 152.6 ppm) of the 5-chloro-2(3H)-benzoxazolone (2) was retained in the ¹³C spectrum of 4a (δ = 154.2 ppm). The alkylation of 2 with 3a-f in the presence of zinc dust yielded the products with less purity in low yield. Hence the reaction was carried out under conventional method using a base.

Reagents and conditions: (i) Conc. HCl, 140-170°C, 6 h; (ii) K₂CO₃, CH₃CN, Room temperature, 8 h; (iii) Method A: K₂CO₃, KI, DMF, 100°C, 4 h; (iv) Method B: Zn, THF, Reflux, 3 h

Scheme 1
Compounds 6a-f were synthesized by means of the following two methods as shown in the Scheme 1: (i) Method A: Compound 4a was reacted with various cyclic amines (5a-f) using K$_2$CO$_3$ in dry DMF at 100°C to obtain different 5-chloro-3-[3-(cyclic amine)propyl]-1,3-benzoazol-2(3H)-ones (6a-f). (ii) Method B: A simple and convenient new synthesis of 6a-f was carried out using zinc dust under neutral conditions. Compound 4a was reacted with various cyclic amines (5a-f) using zinc dust in dry THF to obtain different 6a-f. The products were confirmed by $^1$H NMR, $^{13}$C NMR, IR and Mass Spectroscopy. Thus the synthesis of 6a-f was illustrated by the use of environmentally benign protocol that involves the utilization of the recyclable metal, zinc dust as the reaction medium that enables economical, efficient and high yield products. The corresponding yields of the two methods were compared and it was found that method which utilizes zinc was superior to K$_2$CO$_3$. It was also investigated for the possibility of zinc dust in catalytical or less than stoichiometric quantities. However, high yields of the products with high purity were obtained with one equivalent of zinc dust. Reusability of zinc dust was also studied. The zinc dust was reactivated and used in subsequent runs. It has shown nearly same activity after each use.

**Alkylations under heterogeneous conditions: CsF–Celite catalyzed alkylation of 2(3H)-benzoxazolone**

The alkylation of 2(3H)-benzoxazolone under basic conditions which makes use of bases like K$_2$CO$_3$, NaOEt is known in literature. However, there is still a demand for base catalysts for alkylation of 2(3H)-benzoxazolone via an environmentally friendly process as resulting products are medicinally important heterocycles. As it is mentioned above, we have prepared various N-alkylated 2(3H)-benzoxazolone derivatives 4a-f and 6a-f. There was till a scope for improvement in terms of yields and reaction time. The use of Cesium fluoride-Celite (CsF-Celite) in various transformations is well reported in literature. Organic reactions on solid supported reagents coupled with microwaves are currently of increasing interest due to their greater selectivity, enhanced reaction rates, cleaner reaction products and operational simplicity.
Herein a practical and convenient method for the alkylation of 5-chloro-2(3H)-benzoxazolone using CsF-Celite as catalyst under microwave conditions is reported. 5-Chloro-2(3H)-benzoxazolone (2) was reacted with various alkyl bromides (3a-f) and 3-cyclic amine substituted propyl chlorides (7a-f) in presence of CsF–Celite under microwave to obtain 5-chloro-3-substituted-1,3-benzoxazol-2(3H)-ones (4a-f) and 5-chloro-3-[3-(cyclic amine)propyl]-1,3-benzoxazol-2(3H)-ones (6a-f) respectively (Scheme 2). The microwave irradiation experiments were carried out using focused microwave instrument Discover (CEM) model operating at 2450 MHz frequency. So as a general procedure, one equivalent of each 5-chloro-2(3H)-benzoxazolone (2), either of halides 3a-f or 7a-f and CsF–Celite were mixed well and subjected to microwave irradiation at 140 watts for 60 seconds (Method A). The products were confirmed by comparison of spectral data with that of the products prepared earlier in this section.

\[
\begin{align*}
\text{(i)} & \quad \text{Method A} \quad \text{CsF-Celite, Microwave, 60 sec} \\
\text{(ii)} & \quad \text{Method B} \\
\end{align*}
\]

| R = 3-chloropropyl, allyl, propargyl, benzyl, 3-butenyl, 3-methyl-2-butenyl |
| X = N-benzyl, N-phenyl, N-Pyridyl, N-Pyrimidyl, N-3-chlorophenyl, O |

Reagents and Conditions: (i) Method A: CsF-Celite, Microwave, 60 sec; (ii) Method B: CsF-Celite, THF, rt or reflux, 10 h; (iii) Zn, THF, rt, 1-2 h

Scheme 2
The results were compared with that of CsF–Celite under conventional conditions. The alkylation of 2 using CsF-Celite under conventional conditions was carried out in tetrahydrofuran (THF) at ambient temperature or reflux (Method B). The preparation of the CsF–Celite was carried out in the same manner as described earlier. 3-Cyclic amine substituted propyl chlorides (7a-f) were prepared by reaction of various cyclic amines (5a-f), 1-bromo-3-chloropropene (3a) and zinc dust in THF at ambient temperature (Scheme 2).

For CsF–Celite under microwave conditions the yields were higher and reaction time was decreased from hours to seconds with compared to conventional conditions. This was a remarkable achievement. The yields of 4a-f were lower when reaction was carried out with a single 60 second exposure to microwave irradiation. This was mostly due to volatility of alkyl bromide 3a-f. So to overcome this problem while performing reaction with 3a-f, a 30 second interval was given after a 30 second exposure to microwave irradiation. The best results were obtained with the 1 equivalent of CsF–Celite. Reusability of CsF–Celite was also studied. It has shown nearly same activity after each use. This makes the method more economical.

**Biology**

The synthesized benzoxazole-2(3H)-ones derivatives (4a-f, 6a-f) were evaluated for anticancer, antibacterial and antifungal activities.

**Anticancer activity**

Among the compounds (n=7) evaluated for their cytotoxic potential, five entries showed dose dependent inhibition of cancer cell proliferation (IC\textsubscript{50} values are of less than 100 µM). It is evident from the results that these compounds are comparatively more effective on human lung cancer cells (A549) and breast cancer (MCF-7) than cervical cancer (HeLa) and colon cancer (SW-480) cells. From the cytotoxicity results, it was evident that cytotoxicity of the 2(3H)-benzoxazolone is increasing when substituted cyclic amine is incorporated at its 3\textsuperscript{rd} position with a three carbon spacer.
Antibacterial activity
All the twelve synthesized compounds 4a-f and 6a-f were evaluated for antibacterial activity. The minimum inhibitory concentrations (MIC) of synthesized compounds were tested against three representative Gram-positive organisms and Gram-negative organisms. Compounds 4c with \(N\)-propargyl group and 6f with \(N\)-(3-morpholinyl)propyl group at 3\(^{rd}\) position of benzoxazolone exhibited moderate activity against both Gram-positive and Gram-negative bacteria.

Antifungal activity
All the twelve synthesized compounds 4a-f and 6a-f were evaluated for antifungal activity. Amongst the antifungal activity of the 4 series compounds, 4a with \(N\)-3-chloropropyl group and 4c with \(N\)-propargyl group at 3\(^{rd}\) position of benzoxazolone showed good antifungal activity while 4f with \(N\)-3-methyl-2-butenyl group at 3\(^{rd}\) position of benzoxazolone showed moderate activity. In the 6 series compounds, only 6f with \(N\)-(3-morpholinyl)propyl group at 3\(^{rd}\) position of benzoxazolone showed moderate activity. The antibacterial and antifungal activities results indicate that larger groups at 3\(^{rd}\) position of benzoxazolone and 4\(^{th}\) position of cyclic amine have no significant contribution to these activities of the compounds.

CHAPTER-III
Synthesis and biological evaluation of 3-mercapto/-thione-substituted-1,2,4-triazoles

Chemistry
The chemistry of 1,2,4-triazoles and their fused heterocyclic derivatives has received considerable attention in the last few decades, owing to their synthetic and effective biological importance. Several cyclic amine (e.g. substituted piperazine) derivatives were synthesized in the last decade, as useful chemotherapeutic agents for various diseases. In the present work, synthesis of 3-[3-[4-(substituted)-1-cyclicamine]propyl]thio-5-substituted[1,2,4]triazoles (8) and 2-[3-[4-(substituted)-1-cyclicamine]propyl]-5-(substituted)-2,4-dihydro-3H[1,2,4]triazole-3-thiones (11) is
described. The anticancer, antibacterial and antifungal activities of these derivatives were studied.

- **Retro synthetic analysis**

It was planned to prepare 3-[3-[4-(substituted)-1-cyclicamine]propyl]thio-5-substituted[1,2,4]triazole derivatives (8) as it contains three important parts from a structural point of view as shown in **Figure 1**. (i) a pharmacophoric portion constituted by a substituted 1,2,4-triazole, (ii) a terminal fragment constituted by a substituted cyclic amine like aryl piperazine which is again a pharmacophore and (iii) a three carbon linker between these two substructures i.e. pharmacophores. So idea was that this structural future could have influence on their biological activity.

![Retro synthetic analysis](image)

The 3-[3-[4-(substituted)-1-cyclicamine]propyl]thio-5-substituted[1,2,4]triazole derivatives (8) could be synthesized from the corresponding aryl carboxylic acids (1) (Scheme 1).

![Scheme 1: Retro synthetic analysis](image)

- **Synthesis of the 3-[3-[4-(substituted)-1-cyclicamine]propyl]thio-5-substituted[1,2,4]triazole derivatives (8)**
5-Substituted[1,2,4]triazole-3-thiones (5) were synthesized by following reported sequence of the reactions from corresponding aryl carboxylic acids (1) (Scheme 2). The aryl carboxylic acids (1) were converted to corresponding methyl esters (2) with catalytic amount of sulfuric acid in methanol. The methyl esters (2) were converted to corresponding acid hydrazides (3) using hydrazine hydrate in methanol. The acid hydrazides (3) were reacted with potassium thiocyanate and concentrated hydrochloric acid to produce corresponding thiosemicarbazides (4). Thiosemicarbazides (4) were reacted with aqueous sodium hydroxide to produce 5-substituted[1,2,4]triazole-3-thiones (5). They can also exist in tautomeric thiol form. Here it was found that they mainly exist in thione form which is also supported by some earlier reports.

The 5-substituted[1,2,4]triazole-3-thiones (5) were reacted with 1-bromo-3-chloropropane in presence of K₂CO₃ in acetonitrile at ambient temperature to obtain the corresponding 3-(3-chloropropyl)-sulfanyl-5-substituted[1,2,4]triazoles (6) (Scheme 3). Now to obtain the 3-[3-[4-(substituted)-1-cyclicamine]propyl]thio-5-substituted[1,2,4]triazole derivatives (8), 3-(3-chloropropyl)-sulfanyl-5-substituted[1,2,4]triazoles (6) were reacted with cyclic amines (7) in presence of K₂CO₃ and catalytic amount of KI in acetonitrile (Scheme 3). After work up when product was analyzed it was not the desired 8 because characteristic cyclic amine CH₂ protons peaks
were absent in $^1$H NMR spectra. In some cases desired product 8 was formed but yield was less than 10% while other product was major.

![Diagram](image.png)

So we have studied the particular example of 3-(3-chloropropyl)-sulfanyl-5-(4-tert-butylphenyl)-1,2,4-triazoles (6a). 6a was reacted with morpholine (7a) in presence of K$_2$CO$_3$ and catalytic amount of KI in acetonitrile (Scheme 4). A CH$_2$ peak as triplet with chemical shift 4.36 was observed in $^1$H NMR spectrum of the product which is characteristic triazole $N$-CH$_2$ peak. Characteristic NH stretching of triazole was absent in IR spectra and mass spectrum of product was showing elimination of HCl. All the three observations were pointing structure of the product as 9a. This was further supported by some reports which states formation of linear ring rather than angular.

![Diagram](image.png)

So we did further study with changing alkyl chain length and substituents on phenyl ring to observe that whether it favors formation of 8 or 9 (Scheme 5). The product 9 was obtained with carbon chain length 2, 3, 4 and 5. The different substituents on phenyl ring had also always favored the product 9. In no cases the desired 8 was obtained.
SYNOPSIS

Initially, 1-(3-chloropropyl)-4-substituted cyclic amines (10) were prepared by the reaction of cyclic amines (7) with 1-bromo-3-chloropropane in presence of activated zinc in THF at ambient temperature (Scheme 6).

By careful literature work the methods for the selective N and S-alkylation of 4-amino-5-aryl-1,2,4-triazole-3-thiols were found. Now it was planned to prepare 2-[3-[4-(substituted)-1-cyclicamine]propyl]-5-(substituted)-2,4-dihydro-3H[1,2,4]triazole-3-thiones (11) in addition to 8 as this may leads some interesting results in biological activity point of view. Thus to obtain the target compounds 8 and 11 in shorter reaction time, good yields and selectivity, we did slight modifications in the reported methods.
When the 5-substituted[1,2,4]triazole-3-thiones (5) and 1-(3-chloropropyl)-4-substituted cyclic amines (10) were reacted in the presence of triethyl amine in ethanol with a catalytic amount of tetra butyl ammonium iodide (TBAI). Exclusively 3-[3-[4-(substituted)-1-cyclic amine]propyl]thio-5-substituted[1,2,4]triazoles (8) were formed i.e. S-alkylation (Scheme 7). In the derivatives 8, a triplet for the methylene group of the propyl chain that connects the cyclic amine moiety and the triazole part of the molecule is observed with a chemical shift in the range of δ 3.25-3.15, which is typical for S–CH₂ connectivity. Further the products (8a-m) were confirmed by mass and IR spectroscopy.

When the 5-substituted-1,2,4-triazole-3-thiones (5) and 1-(3-chloropropyl)-4-substituted cyclic amines (10) were reacted in the presence of K₂CO₃ in acetonitrile with a catalytic amount of TBAI. The 2-[3-[4-(substituted)-1-cyclic amine]propyl]-5-(substituted)-2,4-dihydro-3H[1,2,4]triazole-3-thiones (11) were formed i.e. N-alkylation with the traces of 8 (Scheme 8). In the derivatives 11, a triplet for the methylene group of the propyl chain that connects the piperizine moiety and the triazole part of the molecule is observed with a chemical shift in the range of δ 4.45-4.35, which is typical for NCSN–CH₂ connectivity. Further the products (11a-h) were confirmed by mass and IR spectroscopy.
All the 21 compounds 8a-m and 11a-h were tested for anticancer, antibacterial and antifungal activities.

**Anticancer activity**

The sensitivity of the human leukemic cell lines, HL-60 (myeloid leukemia) and U937 (leukemic monocyte lymphoma) to the synthetic triazole derivatives 8a-m and 11a-h was evaluated. The compounds with chloro group at 3\textsuperscript{rd} or 4\textsuperscript{th} position of the phenyl ring and piperazine containing N-3-chlorophenyl, N-2-pyrimidyl and N-2-pyridyl groups had shown good cytotoxic activity. Overall it was observed from the results that cytotoxicity depends on the particular substituents on the phenyl ring and piperazine ring rather than S and N alkylation of triazoles.

**Antibacterial activity**

All the 21 compounds 8a-m and 11a-h were evaluated for antibacterial activity. The compounds had not shown any remarkable antibacterial activity.

**Antifungal activity**

All the 21 compounds 8a-m and 11a-h were evaluated for antifungal activity. Most of the compounds had shown moderate activity especially against *C.rugosa*. The compounds 8b, 11d and 11e had shown comparatively more antifungal activity than other compounds. None of the compounds had shown higher activity than the standard Amphotericin-B.
CHAPTER-IV

Stereoselective synthesis of 2,4-disubstituted piperidines

**BiCl₃ promoted aza-Prins type cyclization: a rapid and efficient synthesis of 2,4-disubstituted piperidines**

Epoxides are extensively used as starting materials and intermediates in organic synthesis because of their ease of formation and versatile reactivity towards nucleophiles. Bismuth salts have attracted attention due to their low toxicity, low cost and good stability. They have been reported as environmentally benign Lewis acid catalysts. Due to the importance of bismuth compounds in organic synthesis, here the utility of bismuth(III) chloride (BiCl₃) as a catalyst for the opening of epoxides with N-protected homoallyl amines was studied. In this note, the stereoselective synthesis of trans-4-chloro-2-substituted piperidines by the reaction of epoxides and N-protected homoallyl amines using BiCl₃ as a Lewis acid catalyst under mild reaction conditions is described (**Scheme 1**).

![Scheme 1](image)

Various mono-N-protected homoallyl amines were treated with structurally diverse epoxides in the presence of BiCl₃ in dichloromethane at 0°C - room temperature for a period of 30-60 min. The products 3 were obtained in good to high yields. In all cases the trans-2,4-diastereomer was the major product, the trans stereochemistry of the products being confirmed by comparison of the ¹H NMR data of 3a with those reported. With N-benzyl and N-allyl homoallyl amines the reaction was not successful. The reaction was carried out in various solvents and the best results were obtained in dichloromethane. Other Lewis acids were also used to observe their effect on reaction time, yields and diastereoselectivity. In all cases diastereoselectivity remained the same.
(9:1) but shorter reaction times and higher yields were obtained with BiCl$_3$. Epoxides were always attacked on the less hindered carbon.

The probable mechanism for this reaction is shown in Scheme 2. The reaction is expected to proceed through iminium ion formation stabilized by adjacent sulfonyl or carbonyl functionalities.

In conclusion, a novel and facile synthesis of piperidine derivatives using BiCl$_3$ as a catalyst for epoxide opening using N-protected homoallyl amines is reported. Several N-protected homoallyl amines and epoxides were subjected to an aza-Prins cyclization. A rapid and efficient BiCl$_3$ promoted stereoselective synthesis of trans-2,4-disubstituted piperidine derivatives was achieved. The catalyst is insensitive to air moisture and the products were formed in high yields with high stereoselectivity.

- Zn(OTf)$_2$ -catalyzed synthesis of 4-halo-2-substituted piperidines in the presence of TBDMSX

In continuation of our work on the stereoselective synthesis of substituted piperidines, herein an efficient and novel synthesis of various 4-halo-2-substituted piperidines by the reaction of aldehydes and N-protected homoallyl amines with use of
tert-butyldimethylsilyl halides (TBDMSX) and catalytic amount of Zn(OTf)₂ by means of aza-Prins type cyclization under mild conditions is described (Scheme 3).

\[
\text{NHR} + \text{R'-CHO} \xrightarrow{\text{25 Mol% Zn(OTf)₂}} \text{N' R'}
\]

\[
\begin{array}{c}
\text{NHTs} + \text{CHO} \xrightarrow{\text{25 Mol% Zn(OTf)₂}} \text{Cl} \\
\end{array}
\]

\[
\begin{array}{c}
\text{R= Tosyl, Mesyl; R'= Aryl, Heterocycle, Alkyl; X= Cl, Br, I} \\
\end{array}
\]

Scheme 3

Initially, N-tosyl homoallyl amine 1a (1 equivalent) and cyclohexane carboxaldehyde 4a (1 equivalent) were treated with TBDMSCl (1 equivalent) in the presence of Zn(OTf)₂ (25 mol%) at 0°C-rt in dichloromethane. The reaction proceeded with 94% yield in 6 h with trans-4-chloro-2-cyclohexyl-1-tosylpiperidine (5a) as a major product. The other isomer was negligible and was not isolated (Scheme 4).

To optimize the Zn(OTf)₂ mol% we repeated same experiment with 5, 10, 15, 20, 25 and 30 mol% of Zn(OTf)₂. Best results (94% yield) obtained with 25 mol% of Zn(OTf)₂. The reaction was unsuccessful in the absence of Zn(OTf)₂. Now, we treated N-tosyl homoallyl amine (1 equivalent) and cyclohexane carboxaldehyde (1 equivalent) with TBDMSBr (1 equivalent) and TBDMSI (1 equivalent) respectively in the two separate experiments in the presence of Zn(OTf)₂ (25 mol%). Very good results were obtained with the trans-diastereoselectivity. The TBDMSBr and TBDMSI were generated in situ by 1 equivalent TBDMSCl + 1 equivalent KBr and 1 equivalent TBDMSCl + 1 equivalent KI respectively. To check the generality of the reaction N-tosyl homoallyl amine was treated with various aldehydes like benzaldehyde, anisaldehyde and thiophene-2-carboxaldehyde. The reaction proceeded with the trans-
2,4-diastereomer as the major product with high yields. The reaction was also successful with N-mesyl homoallyl amine. In the presence of TBDMSI reaction was faster with high yield. With chlorotrimethylsilane (TMSCl) reaction proceeded fast (2-3 h) but with low diastereoselectivity. Among the various triflates tried best results were obtained with Zn(OTf)₂. Dichloromethane was found the most suitable solvent. Here the Zn(OTf)₂ believed to activate the aldehyde and facilitate the amine attack while TBDMSX is acting as a halide donor (Scheme 5).

Various halo-substituted piperidines were clearly distinguished by the comparison of the chemical shift of the carbon at the 4th position in ¹³C NMR spectrum of the compounds. The trans-diastereoselectivity of the products was confirmed by ¹H NMR and was supported by NOE study. Further it was confirmed by the comparison of ¹H NMR and ¹³C NMR data of 5a, 5b and 5f with those of reported.

In conclusion, a simple and efficient procedure for the synthesis of trans-4-halo-2-substituted piperidines by using TBDMSX and catalytic amount of Zn(OTf)₂ was reported. The products were formed in high yields with high stereoselectivity. The highlight of this method is chloro, bromo and iodo piperidines can be obtained using same method. The catalytic use of Zn(OTf)₂ makes the method economical.