3

RESULTS AND DISCUSSION

The work carried out in this section is discussed under the two main heads:

3.1 Chemical studies and

3.2 Biological studies

3.1 CHEMICAL STUDIES

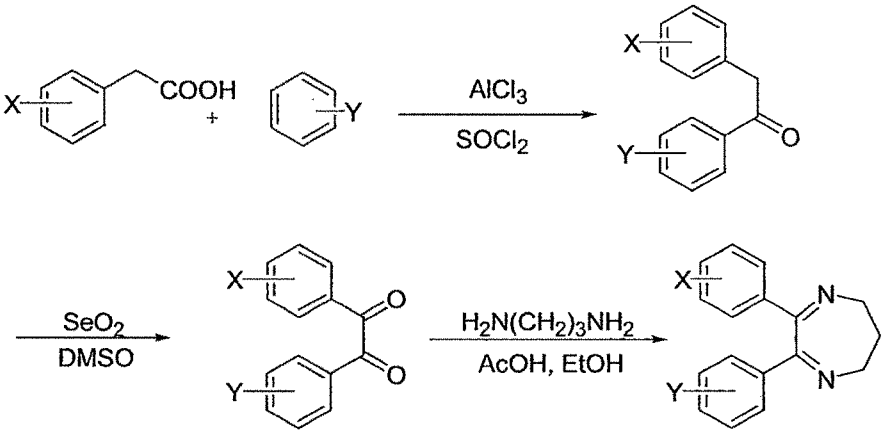
The chemical studies are broadly classified into two heads:

3.1A 2,3-Diaryl-6,7-dihydro-5*H*-1,4-diazepines

3.1B 5,7-Diaryl-2,3-dihydro-1*H*-1,4-diazepines

3.1A 2,3-Diaryl-6,7-dihydro-5*H*-1,4-diazepines (A)

It was envisaged to synthesize 2,3-diaryl-6,7-dihydro-5*H*-1,4-diazepines **(A)** as discussed earlier. For preparation of such a system the required 1,2-diarylethane-1,2-dione is synthesized so that the dione could be cyclised into the corresponding 6,7-dihydro-1,4-diazepine **(Scheme-1)**.



Scheme-1

The substituted phenylacetyl chloride was reacted with substituted benzenes through Friedel -Crafts acylation to afford the ethanones. The ethanones were

oxidized to the desired ethanedione using selenium dioxide as the oxidizing agent. The desired ethanediones derivatives thus obtained were cyclised with 1,3-propanediamine to afford the respective diazepines.

The work carried out is discussed under the following sub heads:

3.1A.1 Synthesis of substituted phenylacetic acid derivatives

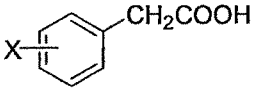
3.1A.2 Synthesis of 1,2-diaryl ethanone derivatives

3.1A.3 Synthesis of 1,2-diaryl-1,2-ethanedione derivatives

3.1A.4 Synthesis of 2,3-diaryl-1,4-diazepine derivatives

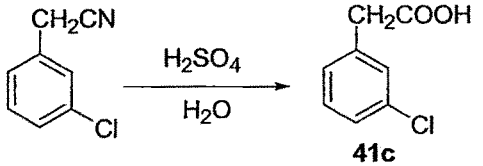
3.1A.1 Synthesis of substituted phenyl acetic acid derivatives (41a-f)

Commercially available phenylacetic acid derivatives were used without purification. The expensive phenylacetic acid derivatives were synthesized by either Kindler modified Willgerodt reaction[171, 172] or acid hydrolysis of substituted benzyl cyanides[173, 174]. Commercially available phenylacetic acid **(41a)**, 2-chlorophenylacetic acid **(41b)**, 4-chlorophenylacetic acid **(41d)** were used and 3-chlorophenylacetic acid **(41c)**, 4-methylphenylacetic acid **(41e)** and 4-nitrophenylacetic acid **(41f)** were synthesized.



41a: X = H
41b: X = 2-Cl
41c: X = 3-Cl
41d: X = 4-Cl
41e: X = 4-CH_3
41f: X = 4-NO_2

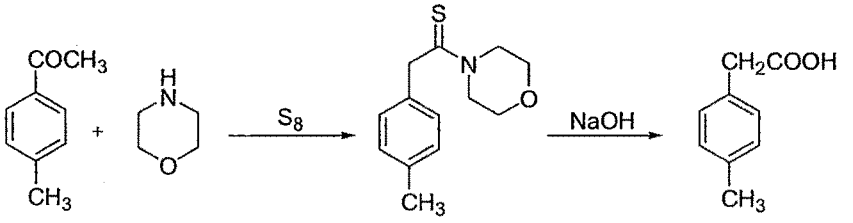
The 3-chlorophenylacetic acid **(41c)** was obtained by hydrolyzing the nitrile group of 3-chlorobenzyl cyanide in acidic medium[174].



41c

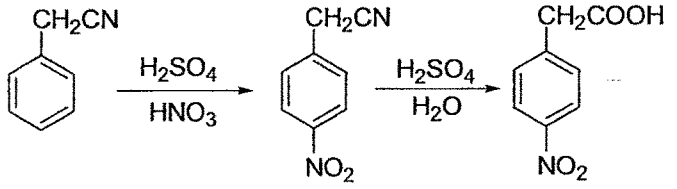
Compound **(41c)** displayed the absorption bands at 3026 (OH) and 1700 cm^{-1} (C=O) in its IR spectrum.

4-Methylphenylacetic acid[175] **(41e)** was synthesized by Kindler modified Willgerodt reaction. 4-Methylacetophenone was refluxed with sulphur in morpholine to afford the thiomorpholide. The crude thiomorpholide was hydrolysed in alkaline media to afford compound **(41e)** which showed characteristic peaks at 3000 (OH) and 1700 cm^{-1} (C=O) in the I.R spectrum.



41e

Benzyl cyanide was nitrated using nitric acid-sulphuric acid mixture to afford a sufficiently pure product. The nitrile was hydrolysed in acidic medium to afford the acid in pure form[176]. The 4-nitrophenylacetic acid[177] **(41f)** displayed bands at 2900, 1700, 1511 and 1347 cm^{-1} in its IR spectrum.



41f

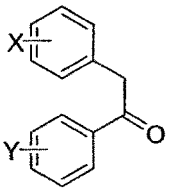
3.1A.2 Synthesis of 1,2-diaryl-1-ethanone derivatives (42a-r)

The acid chlorides were obtained by reacting the corresponding phenylacetic acids with thionyl chloride or phosphorous trichloride. The substituted phenylacetyl chlorides were treated with substituted aryl compounds in presence of $AlCl_3$ using benzene as solvent[178, 179]. After usual workup, the required 1,2-diaryl-1-ethanones **(42a-r)** were obtained in almost quantitative yield. This reaction is based on conventional Friedel's–Craft acylation reaction. The carbonyl stretching of the ethanones are given in **Table 1**.

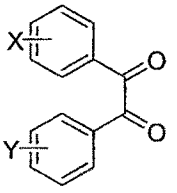
3.1A.3 Synthesis of 1,2-diarylethane-1,2-dione derivatives (43a-r)

The ethanones were converted to the respective ethandiones using selenium dioxide as oxidizing agent under microwave irradiation[180].

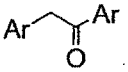
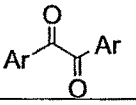
Table 1. General structure of 1,2-diarylethanone and ethanedione derivatives



42a-r



43a-r

Compound	X	Y	IR (cm^{-1})		Reaction Time (Sec)
					
42a, 43a	H	H	1676	1654	30
42b, 43b	H	4-CH_3	1680	1652	30
42c, 43c	H	4-Br	1686	1668	95
42d, 43d	H	4-F	1685	1664	80
42e, 43e	H	4-OCH_3	1674	1668	35
42f, 43f	H	4-SCH_3	1681	1666	35
42g, 43g	4-Cl	4-CH_3	1676	1666	50
42h, 43h	4-Cl	4-Cl	1690	1658	100
42i, 43i	4-Cl	4-F	1683	1663	80
42j, 43j	4-Cl	4-SCH_3	1672	1652	115
42k, 43k	4-Cl	H	1684	1667	55
42l, 43l	4-Cl	4-Br	1689	1664	155
42m, 43m	4-NO_2	H	1685	1662	20
42n, 43n	4-NO_2	4-CH_3	1680	1661	35
42o, 43o	2-Cl	4-Cl	1691	1674	40
42p, 43p	2-Cl	4-SCH_3	1670	1659	40
42q, 43q	4-CH_3	4-CH_3	1685	1660	40
42r, 43r	3-Cl	4-CH_3	1685	1677	55

Unlike the conventional methods[181, 182], microwave assisted selenium dioxide oxidation of ethanone derivatives using DMSO as solvent simplifies the workup procedure and reduction of the reaction time from 6-8 hrs to 100 seconds. The

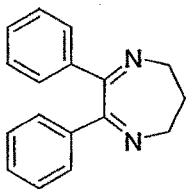
reaction time and the carbonyl stretching of the ethanediones are given in **Table 1**. Some ethanedione derivatives were obtained as either oily or sticky compound and used in the next step without further purification.

3.1A.4 Synthesis of 2,3-diaryl-1,4-diazepine derivatives (44a-r)

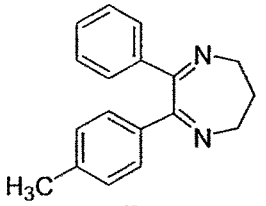
The 1,2-diarylethane-1,2-dione derivatives **(43a-r)** and 1,3-propanediamine reacted in equimolar amounts in ethanol under refluxing condition affords the cyclised product[115]. It is clearly stated in literature that synthesis of these analogs is formidable[183]. The yield of the cyclised compound is low to moderate which may be due to the formation of undesired uncyclised product with partial recovery of starting material. The IR spectrum of the uncyclised product shows an intense peak of carbonyl stretching at 1670 cm^{-1} which may be due to multiple condensation of 1,2-diaryl ethanedione with 1,3-propanediamine even when equimolar quantities of ethanedione and 1,3-propanediamine are reacted. The cyclised products are confirmed by the appearance of C=N stretching in the IR spectrum at 1590-1610 cm^{-1} and absence of carbonyl stretching. The proton NMR showed a broad hump for the methylene protons around δ 3.5; C-5 and C-7 of diazepine ring (=N-C$\underline{H_2}$-CH_2, 4H). This may be due to temperature coalescence or ring flip. In case of the compounds **44o** and **44p,** methylene protons of C-5 and C-7 of the diazepine ring appear as a multiplet. The other methylene protons of C-6 of diazepine ring (=N-CH_2-C$\underline{H_2}$, 2H) appear as a multiplet at δ 2.32-2.39. The M+1 peak was observed for all compounds and M+2 peak is observed for halo compounds in the mass spectrum.

2,3-Diphenyl-6,7-dihydro-5*H*-1,4-diazepine **(44a)** was obtained by above described condition. Compound **(44a)** showed a characteristic sharp peak at 1605 cm^{-1} for C=N stretching in IR spectrum. The PMR spectrum displayed multiplet at δ 2.31-2.40 for two protons (N-CH_2-C$\underline{H_2}$-) and a broad hump was observed at 3.47 for four protons (N-C$\underline{H_2}$-CH_2-). The ten aromatic protons were

seen as a multiplet at 7.2-7.6 (m, 10H, ArH). The M+1 peak was obtained at 249 (m/z) in its mass spectrum.



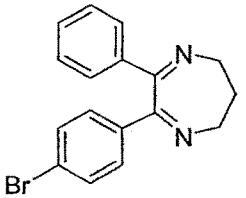
44a



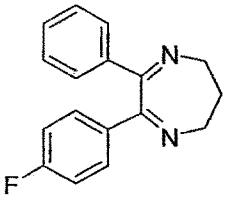
44b

Compound **(44b)** gave a characteristic peak at 1608 cm^{-1} for C=N stretching in IR spectrum. In the PMR spectrum signals were obtained at δ 2.32 (s, 3H, CH_3), 2.34-2.41 (m, 2H, N-CH_2-$C\underline{H}_2$-), 3.5 (br, 4H, N-$C\underline{H}_2$-CH_2-) and 7.1-7.6 (m, 9H, ArH). The M+1 peak was obtained at 263 (m/z) in its mass spectrum.

2-Phenyl-3-(4-bromophenyl)-6,7-dihydro-5*H*-1,4-diazepine **(44c)** showed infrared bands at 1598 cm^{-1}, a characteristic C=N stretching of the diazepine ring system. In PMR spectrum, multiplet at δ 2.33-2.41 for two protons (N-CH_2-$C\underline{H}_2$-) and a broad hump was observed at 3.5 for four protons (N-$C\underline{H}_2$-CH_2-). The nine aromatic protons showed a multiplet at 7.1-7.6 (m, 9H, ArH). The M+2 peak was obtained at 329 (m/z) in its mass spectrum.



44c

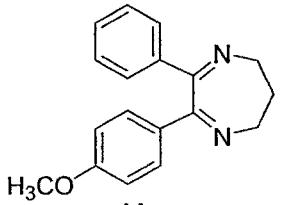


44d

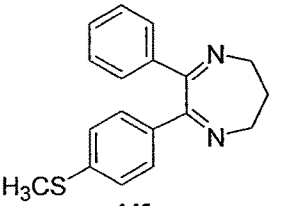
2-Phenyl-3-(4-fluorophenyl)-6,7-dihydro-5*H*-1,4-diazepine **(44d)** gave the vibrational bands at 1598 cm^{-1}, a characteristic C=N stretching of the diazepine ring system. In the PMR spectrum, signals were obtained at δ 2.31-2.4 (m, 2H, N-CH_2-$C\underline{H}_2$-), 3.5 (br, 4H, N-$C\underline{H}_2$-CH_2-) and 6.9-7.6 (m, 9H, ArH).

Compound **(44e)** showed a sharp peak for C=N at 1600 cm^{-1} and peaks at 1247 and 1029 for (Ar-O-Me) asymmetric and symmetric stretching respectively, in the IR spectrum. A sharp singlet at δ 3.77 for three protons (-OCH_3), multiplet

for C-6 methylene protons at 2.31-2.38 (2H, N-CH_2-$C\underline{H}_2$-) and a hump at δ 3.2 (br, 4H, N-$C\underline{H}_2$-CH_2-) were observed in the PMR spectrum. A multiplet was observed for aromatic protons at δ 6.8-7.3 (m, 9H, ArH). The M+1 peak was obtained at 279 (m/z) in its mass spectrum.



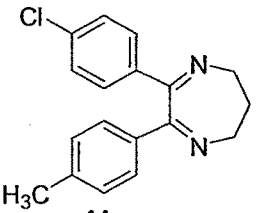
44e



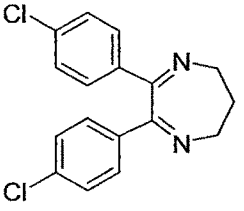
44f

2-Phenyl-3-(4-methylsulfanylphenyl)-6,7-dihydro-5*H*-1,4-diazepine **(44f)** showed a characteristic sharp peak at 1593 cm^{-1} for C=N stretching in IR spectrum. The PMR spectrum displayed a sharp singlet at δ 2.4 for three protons (-SCH_3), a multiplet was observed at δ 2.34-2.37 for two protons (N-CH_2-$C\underline{H}_2$-) and a broad hump was observed at δ 3.5 for four protons at (N-$C\underline{H}_2$-CH_2-). The nine aromatic protons appeared as a multiplet at δ 7.1-7.5 (m, 9H, ArH). The M+1 peak was obtained at 295 (m/z) in its mass spectrum.

Compound **(44g)** gave a characteristic peak at 1610 cm^{-1} for C=N stretching in the IR spectrum. In the PMR spectrum signals were obtained at δ 2.3 (s, 3H, CH_3), 2.32-2.39 (m, 2H, N-CH_2-$C\underline{H}_2$-), 3.4 (br, 4H, N-$C\underline{H}_2$-CH_2-) and δ 7.1-7.5 (m, 8H, ArH).



44g

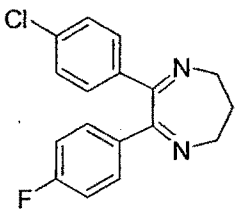


44h

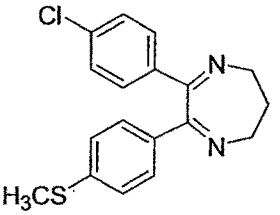
2,3-(4,4-Dichlorophenyl)-6,7-dihydro-5*H*-1,4-diazepine **(44h)** displayed a stretching band at 1614 cm^{-1} for C=N stretching in the IR spectrum. In PMR spectrum, a multiplet for C-6 methylene protons at δ 2.31-2.40 (2H, N-CH_2 -$C\underline{H}_2$-) and a hump at 3.5 (br, 4H, N-$C\underline{H}_2$-CH_2-) were observed in the PMR

spectrum. A multiplet was observed for aromatic protons at δ 7.2-7.5 (m, 8H, ArH).

2-(4-Chlorophenyl)-3-(4-fluorophenyl)-6,7-dihydro-5*H*-1,4-diazepine **(44i)** gave vibrational bands at 1614 cm^{-1} for C=N stretching in IR spectrum. The PMR spectrum showed signals at δ 2.32-2.39 (m, 2H, N-CH_2-$C\underline{H}_2$-), 3.5 (br, 4H, N-$C\underline{H}_2$-CH_2-) and 6.98-7.31 (m, 8H, ArH).



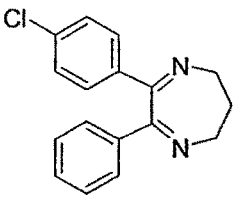
44i



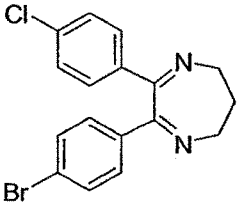
44j

A prominent sharp and strong peak was observed for compound **(44j)** at 1610 cm^{-1} (C=N imine) in IR spectrum. The PMR spectrum displayed a singlet for three protons at δ 2.45 (-SCH_3), a multiplet at δ 2.28-2.43 (2H, N-CH_2-$C\underline{H}_2$-) and a broad peak was observed at 3.51 for four methylene protons, which were adjacent to imine (N-$C\underline{H}_2$-CH_2-). The eight aromatic protons displayed as multiplet at δ 7.1-7.7.

2-(4-Chlorophenyl)-3-phenyl)-6,7-dihydro-5*H*-1,4-diazepine **(44k)** showed infrared bands at 1611 cm^{-1}, a characteristic C=N stretching of the diazepine ring system. In PMR spectrum, multiplet at δ 2.32-2.41 for C-6 methylene protons (2H, N-CH_2-$C\underline{H}_2$-) and a broad hump was observed at 3.5 for C-5 and C-7 methylene protons (N-$C\underline{H}_2$-CH_2-) of the diazepine ring. The nine aromatic protons showed a multiplet at 7.2-7.6 (m, 9H, ArH). The M+1 peak was obtained at 283 (m/z) in its mass spectrum.



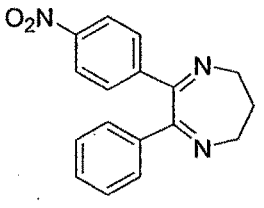
44k



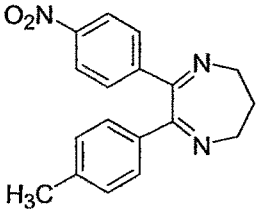
44l

Compound **(44l)** showed a prominent sharp and strong peak at 1612 cm^{-1} (C=N imine) in its IR spectrum. The PMR spectrum displayed a multiplet for C-6 of diazepine proton at δ 2.3-2.4 (2H, N-CH_2-$C\underline{H}_2$-) and a broad peak was observed at 3.5 for C-5 and C-7 of the diazepine ring (4H, N-$C\underline{H}_2$-CH_2-). The eight aromatic protons displayed as a multiplet at δ 7.2-7.5.

The IR spectrum of the compound **(44m)** showed a characteristic peak at 1608 cm^{-1} (C=N imine), asymmetric and symmetric (NO_2) stretching bands at 1520 and 1347 cm^{-1}, respectively. In PMR spectrum, the C-6 protons of diazepine were observed as a multiplet at δ 2.3-2.4 (2H, N-CH_2-$C\underline{H}_2$-), a broad peak was observed at δ 3.5 for C-5 and C-7 methylene protons of diazepine (4H, N-$C\underline{H}_2$-CH_2-). The nine aromatic protons were observed as a multiplet at δ 7.2-8.1 (9H, ArH). The M^+ peak was obtained at 293 (m/z) in its mass spectrum.



44m

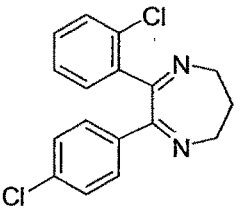


44n

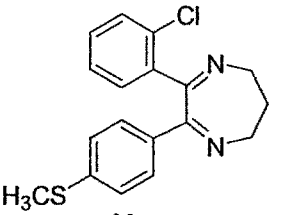
2-(4-Nitrophenyl)-3-(4-methylphenyl)-6,7-dihydro-5*H*-1,4-diazepine **(44n)** showed a characteristic peak at 1595 cm^{-1} (C=N imine), asymmetric and symmetric (NO_2) stretching bands at 1514 and 1344 cm^{-1}, respectively in IR spectrum. In PMR spectrum, a sharp singlet was observed for three protons at δ 2.33 (-CH_3), the protons of methylene at C-6 were observed as multiplet at 2.34-2.4 (2H, N-CH_2-$C\underline{H}_2$-), a broad hump was observed at 3.58 for C-5 and C-7 of diazepine ring (4H, N-$C\underline{H}_2$-CH_2-). The eight aromatic protons were observed as a multiplet at δ 7.1-8.1 (8H, ArH).

2-(2-Chlorophenyl)-3-(4-chlorophenyl)-6,7-dihydro-5*H*-1,4-diazepine **(44o)** gave a prominent sharp and strong peak at 1608 cm^{-1} (C=N imine) in the IR spectrum. The PMR spectrum displayed a multiplet at δ 2.43-2.52 (2H, N-CH_2-

CH_2-), C-7 diazepine protons appeared as a triplet at 3.58-3.62 (2H, *J* =12 Hz, N-CH_2-CH_2-) and C-5 diazepine protons displayed a triplet at 3.70-3.74 (2H, *J* =12 Hz, N-CH_2-CH_2-). A multiplet was observed for eight aromatic protons at δ 7.22-7.68.



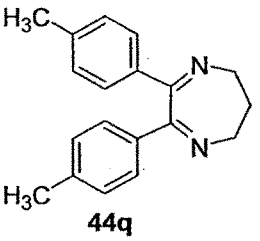
44o



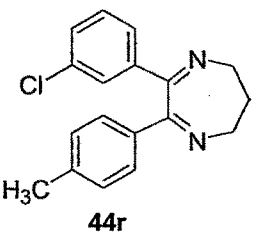
44p

The IR spectrum of the compound **(44p)** showed an intense peak at 1610 cm^{-1} for C=N (imine) stretching. The PMR spectrum displayed signals at δ 2.43 (s, 3H, SCH_3), the C-6 methylene protons of the diazepine showed multiplet at δ 2.46-2.49 (2H, N-CH_2-CH_2-), C-7 of diazepine protons showed triplet at δ 3.60-3.63 (2H, *J* =12 Hz, N-CH_2-CH_2-) and C-5 of diazepine protons displayed triplet at δ 3.70-3.73 (2H, *J* =12 Hz, N-CH_2-CH_2-). A multiplet was observed for eight aromatic protons at δ 7.1-7.3. The M+1 peak was obtained at 329 (m/z) in its mass spectrum.

In IR spectrum of compound **(44q)**, an intense and sharp peak was observed at 1608 cm^{-1} (C=N imine). In PMR spectrum, six protons of methyl observed as a singlet at δ 2.32 (6H, -CH_3), multiplet for C-6 of diazepine ring protons observed at δ 2.34-2.37 (2H, N-CH_2-CH_2-), and a broad peak observed for C-5 and C-7 of diazepine protons at δ 3.5 (br, 4H, N-CH_2-CH_2-). The eight aromatic protons were found as multiplet at δ 7.11-7.52 (8H, ArH). The M+1 peak was obtained at 277 (m/z) in its mass spectrum.



44q

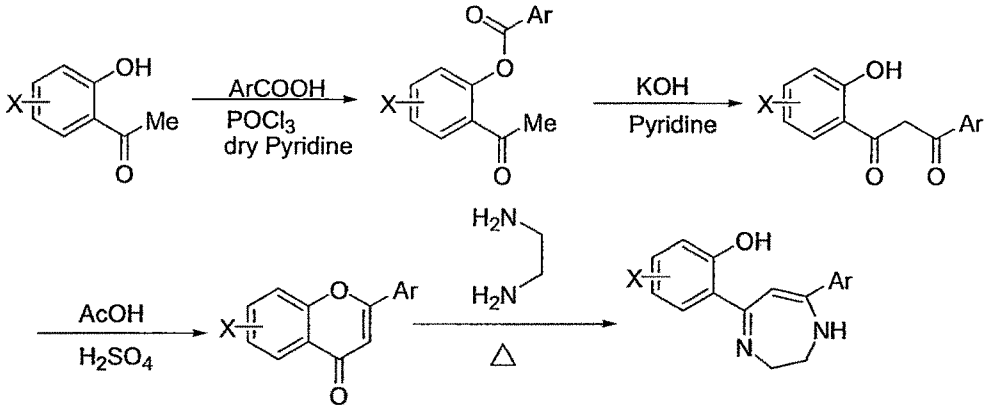


44r

2-(3-Chlorophenyl)-3-(4-methylphenyl)-6,7-dihydro-5H-1,4-diazepine **(44r)** gave a sharp peak at 1608 (C=N imine) in IR spectrum. The PMR spectrum displayed a singlet for three protons at δ 2.33 (-CH_3), C-6 of diazepine protons observed as a multiplet at δ 2.35-2.40 (2H, N-CH_2-$C\underline{H}_2$-) and a broad hump was observed for four protons at δ 3.53 (4H, N-$C\underline{H}_2$-CH_2-). The aromatic protons were found as a multiplet at δ 7.1-7.7 (m, 8H, ArH). The M+1 peak was obtained at 297 (m/z) in its mass spectrum.

3.1B 5,7-Diaryl-2,3-dihydro-1*H*-1,4-diazepines (B)

It was envisaged to synthesize 5,7-diaryl-2,3-dihydro-1*H*-1,4-diazepines **(B)** **(Scheme 2)** as discussed previously. For the preparation of such a system 1,3-diarylpropane-1,3-dione was required which could be cyclised to 2,3-dihydro-1,4-diazepine.



Scheme 2

The 2-hydroxyacetophenone derivatives were reacted with various substituted aromatic/hetero aromatic acid chlorides (generated in situ) through Schotten-Baumann reaction to afford the esters[184]. Baker-Venkatraman rearrangement of the esters furnished 1,3-propanedione derivatives[185], which were subsequently converted into 2-(substituted phenyl)chromen-4-one in acidic medium[186, 187]. The formation of diazepines was accomplished by cyclocondensation of the 2-(substituted phenyl)chromen-4-one with ethylenediamine.

The work carried out has been discussed under sub heads:

3.1B.1 Synthesis of substituted 2-hydroxyacetophenone derivatives

3.1B.2 Synthesis of 2-acetyl-1-(substituted benzoyloxy)benzene derivatives

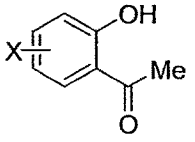
3.1B.3 Synthesis of 1,3-(substituted diaryl)-1,3-propanedione derivatives

3.1B.4 Synthesis of 2-(substituted phenyl)chromen-4-one derivatives

3.1B.5 Synthesis of 5,7-substituted diaryl-1*H*-1,4-diazepine derivatives

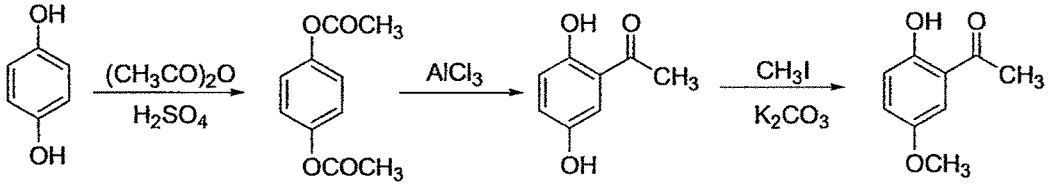
3.1B.1 Synthesis of substituted 2-hydroxyacetophenone derivatives

Commercially available 2-hydroxyacetophenone **(45a)** was used directly without purification. The 5-methoxy-2-hydroxyacetophenone **(45b)** was synthesized by reported method[188].



45a: X= H
45b: X= 5-OMe

The 5-methoxy-2-hydroxyacetophenone **(45b)** was obtained by the selective methylation of 2,5-dihydroxyacetophenone. 2,5-dihydroxyacetophenone[189] was synthesized by Fries rearrangement of hydroquinone-1,4-diacetate which was obtained by acetylation of hydroquinone in the presence of sulphuric acid[190].



Compound **(45b)** displayed the absorption bands at 3066 (OH), 1218 & 1037 (OCH_3) and 1640 cm^{-1} (C=O) in its IR spectrum.

3.1B.2 Synthesis of 2-acetyl-1-(substituted benzoyloxy)benzene (46a-s)

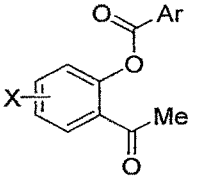
The phenoxybenzoate esters were obtained by condensation of 2-hydroxyacetophenones and various substituted aryl/heteroaryl carboxylic acid derivatives in dry pyridine and phosphorous oxychloride. The carbonyl stretching of the esters are given in the **Table 2.**

3.1B.3 Synthesis of 1,3-substituted diphenyl-1,3-propanedione derivatives (47a-s)

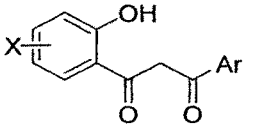
The above synthesized esters were subjected to Baker-Venkataraman rearrangement to afford yellow colored 1,3-diketones. The I.R spectrum of 1,3-diketones showed absorption bands of C=O at the range of 1615-1625cm^{-1} (**Table 3**). Moreover, the characteristic absorption bands of C=O of ketone and ester in the region of 1681 and 1740 cm^{-1} were not found in the I.R spectrum of

1,3-diketones. Most of the derivatives were isolated as yellow colored potassium salt of 1,3-diketones, confirmed by absence of OH stretching in the I.R spectrum, and used directly in the next step of the reaction without purification.

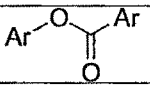
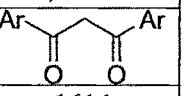
Table 2: General structure of 2-acetyl-1-(substitutedbenzoyloxy)benzene (46a-s) and 1,3-substituted diphenyl-1,3-propanedione derivatives (47a-s)



46a-s



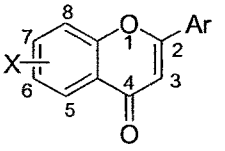
47a-s

Compound	X	Ar	IR (cm^{-1})	
				
46a, 47a	H	C_6H_5	1735	1614
46b, 47b	H	4-$CH_3C_6H_4$	1735	1616
46c, 47c	H	3-$CH_3C_6H_4$	1732	1612
46d, 47d	H	4-$CH_3OC_6H_4$	1724	1616
46e, 47e	H	3-$CH_3OC_6H_4$	1741	1621
46f, 47f	H	3-CH_3O,4-$CH_3OC_6H_3$	1732	1604
46g, 47g	H	4-ClC_6H_4	1739	1625
46h, 47h	H	3-ClC_6H_4	1739	1618
46i, 47i	H	2-ClC_6H_4	1743	1608
46j, 47j	H	2-Cl,4-ClC_6H_3	1751	1620
46k, 47k	H	4-BrC_6H_4	1739	1614
46l, 47l	H	4-FC_6H_4	1741	1625
46m, 47m	H	2-Furyl	1732	1616
46n, 47n	H	2- Thienyl	1732	1620
46o, 47o	6-CH_3O	C_6H_5	1738	1627
46p, 47p	6-CH_3O	4-$CH_3C_6H_4$	1724	1614
46q, 47q	6-CH_3O	4-ClC_6H_4	1726	1608
46r, 46r	6-CH_3O	3-ClC_6H_4	1737	1617
46s, 47s	6-CH_3O	4-$CH_3OC_6H_4$	1735	1612

3.1B.4 Synthesis of 2-(substituted phenyl)chromen-4-one derivatives (47a-s)

2-(substituted phenyl)-chromen-4-one derivatives, major intermediates, were obtained by cyclodehydration of 1,3-diketones with acetic acid containing a few drops of Con.H_2SO_4. The carbonyl stretching of flavones are given in **Table 3.**

Table 3: General structure of 2-(substituted phenyl)chromen-4-one



48a-s

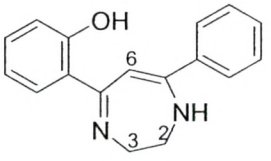
Compound	X	Ar	IR (cm^{-1})
48a	H	C_6H_5	1645
48b	H	4-$CH_3C_6H_4$	1637
48c	H	3-$CH_3C_6H_4$	1637
48d	H	4-$CH_3OC_6H_4$	1649
48e	H	3-$CH_3OC_6H_4$	1652
48f	H	3-CH_3O,4-$CH_3OC_6H_3$	1652
48g	H	4-ClC_6H_4	1662
48h	H	3-ClC_6H_4	1641
48i	H	2-ClC_6H_4	1652
48j	H	2-Cl,4-ClC_6H_3	1650
48k	H	4-BrC_6H_4	1666
48l	H	4-FC_6H_4	1639
48m	H	2-Furyl	1662
48n	H	2- Thienyl	1639
48o	6-CH_3O	C_6H_5	1641
48p	6-CH_3O	4-$CH_3C_6H_4$	1647
48q	6-CH_3O	4-ClC_6H_4	1635
48r	6-CH_3O	3-ClC_6H_4	1642
48s	6-CH_3O	4-$CH_3OC_6H_4$	1647

3.1B.4 Synthesis of 5,7-substituted diaryl-2,3-dihydro-1*H*-1,4-diazepines (49a-s)

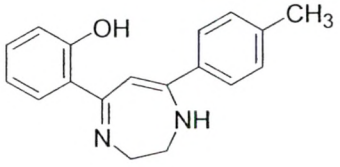
Traditionally, the synthesis of 2,3-dihyro-1,4-diazepines is based on the reaction of 1,3-diketones with ethylenediamine in acidic medium and these type of reactions are well explored by Lloyd *et al*[191-193]. This reaction condition is not

suitable for the above type of 1,3-diketones due to the competitive reaction; the formation of flavone. Also, it is known that when flavone reacts with 1,2-diamine in basic medium it undergoes cyclocondenastion and ring expansion to form diazepine[194, 195]. The 5,7-substituted diaryl-2,3-dihyro-1*H*-1,4-diazepine syntheses were carried out by reacting the flavones with aqueous ethylenediamine. The structures of 5,7-diaryldiazepines are consistent with the IR and NMR spectra. The ring expansion was confirmed by the appearance C=N stretching at 1595-1610 cm^{-1} and NH stretching at 3200 cm^{-1} in the I.R spectrum. The $-CH_2-CH_2-$ protons of the diazepine ring appeared as two broad singlets at δ 3.7 and 3.8, the N-H protons appeared as a broad singlet (δ 7.0-9.0) and the C-6 proton showed a sharp singlet at δ 5.8-6.0. The aromatic protons appeared as a multiplet. The hydroxyl proton could not be traced in the PMR spectra which range from 1-15 ppm. The mass spectra revealed the molecular ion as the base peak for most of the compounds.

5-(2-Hydroxyphenyl)-7-phenyl-2,3-dihydro-1*H*-1,4-diazepine **(49a)** was obtained by the above described reaction condition. It showed a characteristic sharp peak at 1605 cm^{-1} for C=N stretching in the IR spectrum. Peaks at 3231(NH) and 3000 for OH stretching were observed in the I.R spectra. The PMR spectrum displayed two broad singlets at δ 3.65 and 3.9 for C-2 and C-3 of diazepine ring protons (4H, $(CH_2)_2$). A sharp singlet and a broad peak were observed at δ 5.71and 8.35 for C-6 diazepine ring proton and NH proton, respectively. The nine aromatic protons appeared as a multiplet at 6.45-7.74 (m, 9H, ArH). The M^+ peak was obtained at 264 (m/z) in the mass spectrum.



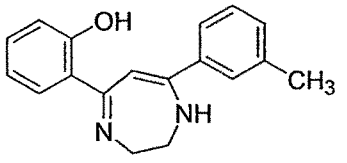
49a



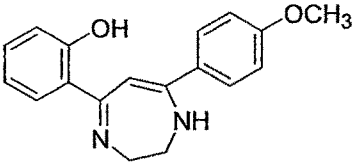
49b

Compound **(49b)** gave a characteristic peak at 1595 (C=N), 2916 (OH) and 3176 cm^{-1} for NH stretching in IR spectrum. In the PMR spectrum two sharp singlet signals were obtained for methyl and C-6 proton of diazepine at δ 2.39 and 5.8 respectively. Two separate broad singlets were observed at 3.72 & 3.82 for C-2 and C-3 diazepine ring protons (4H). The eight aromatic protons were displayed as a multiplet at δ 6.53-7.62 and the NH proton appeared at δ 9.8 as a broad singlet.

5-(2-Hydroxyphenyl)-7-(3-methylphenyl)-2,3-dihydro-1*H*-1,4-diazepine **(49c)** showed infrared bands at 1608 cm^{-1}, a characteristic C=N stretching of the diazepine ring system. The NH and OH stretching bands found at 3232 and 3000 cm^{-1} respectively. In the PMR spectrum, singlet at δ 2.45 for three protons (CH_3), two broad humps at δ 3.62 and 3.9 for four protons (C-2 & C-3 diazepine ring), a sharp singlet at δ 5.7 for C-6 proton of diazepine ring and the NH proton at δ 8.27 as a broad singlet were observed. The eight aromatic protons showed a multiplet at 6.48-7.65 (8H, ArH). The M+1 peak was obtained at 279 (m/z) in its mass spectrum.



49c

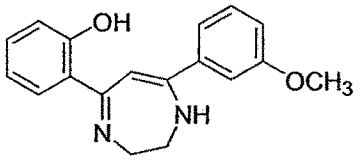


49d

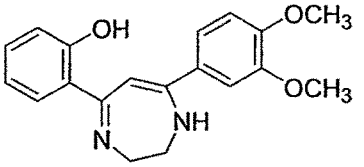
Compound **(49d)** showed a characteristic sharp peak at 1600 cm^{-1} for C=N stretching in the IR spectrum. Peaks at 3200 (NH), 3000 (OH) and asymmetric and symmetric stretching for OCH_3 were found at 1255 and 1031 cm^{-1} respectively. The PMR spectrum displayed two broad singlets at δ 3.63 and 3.88 for C-2 and C-3 of diazepine ring protons (4H, $(CH_2)_2$). A sharp singlet for three protons (OCH_3) merged with one of the above broad peaks at δ 3.88. A sharp singlet and broad peak was observed at δ 5.7 and 8.25 for C-6 diazepine ring

proton and NH proton, respectively. The eight aromatic protons were appeared as a multiplet at δ 6.45-7.65 (m, 8H, ArH).

5-(2-Hydroxyphenyl)-7-(3-methoxyphenyl)-2,3-dihydro-1*H*-1,4-diazepine **(49e)** gave a characteristic peak at 1600 (C=N), 2985 (OH) and 3200 cm^{-1} for NH stretching in IR spectrum. Also, the asymmetric and symmetric peaks for OCH_3 stretching were observed at 1230 and 1048 cm^{-1} respectively. In PMR spectrum two separate broad humps were observed at δ 3.63 and 3.9 for C-2 and C-3 diazepine ring protons (4H). Three integral protons of OCH_3 appeared along with one of the above broad peaks in the region of δ 3.9. A sharp and broad singlet was obtained for C-6 proton of diazepine ring and NH proton at δ 5.72 and 8.25, respectively. The eight aromatic protons were displayed as a multiplet at δ 6.45-7.66. The M+1 peak was obtained at 295 (m/z) in its mass spectrum.



49e

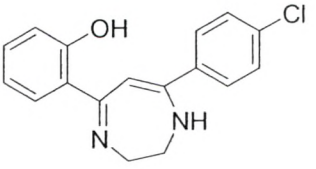


49f

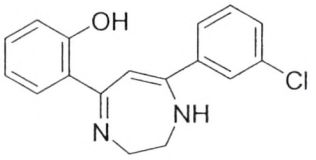
5-(2-Hydroxyphenyl)-7-(3,4-dimethoxyphenyl)-2,3-dihydro-1*H*-1,4-diazepine **(49f)** showed infrared bands at 1602 cm^{-1}, a characteristic C=N stretching of the diazepine ring system and asymmetric and symmetric stretching of OCH_3 at 1251 and 1024 cm^{-1} respectively. The NH and OH stretching bands were found at 3217 and 2929 cm^{-1} respectively. In the PMR spectrum, singlet at δ 3.88 for six protons $(OCH_3)_2$, two broad humps at δ 3.63 and 3.9 for four protons (C-2 and C-3 diazepine ring), a sharp singlet at δ 5.73 for C-6 proton of diazepine ring and the NH proton at δ 8.25 as a broad singlet were observed. The seven aromatic protons showed a multiplet at δ 6.5-7.6 (7H, ArH).

Compound **(49g)** showed a characteristic sharp peak at 1598 cm^{-1} for C=N stretching in the IR spectrum. Peaks at 3203 for NH and 3000 cm^{-1} for OH stretching were observed in the I.R spectra. The PMR spectrum displayed two

broad singlets at δ 3.75 and 3.85 for C-2 and C-3 of diazepine ring protons (4H, $(CH_2)_2$). A sharp singlet and a broad peak were observed at δ 5.74 and 7.74 for C-6 diazepine ring proton and NH proton respectively. The eight aromatic protons appeared as a multiplet at δ 6.61-7.59 (m, 8H, ArH).



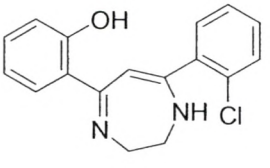
49g



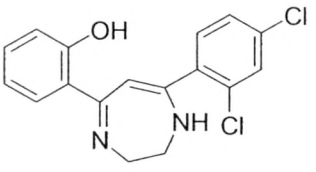
49h

5-(2-Hydroxyphenyl)-7-(3-chlorophenyl)-2,3-dihydro-1*H*-1,4-diazepine **(49h)** gave a characteristic peak at 1610 (C=N), 2916 (OH) and 3200 cm^{-1} for NH stretching in the IR spectrum. In the PMR spectrum a sharp singlet signal was obtained for C-6 proton of diazepine at δ 5.7. Two separate broad singlets were observed at δ 3.62 and 3.92 for C-2 and C-3 diazepine ring protons (4H). The eight aromatic protons were displayed as a multiplet at δ 6.5-7.7 and the NH proton appeared at δ 8.28 as a broad singlet. The M^+ peak was obtained at 298 (m/z) in its mass spectrum.

Compound **(49i)** showed infrared bands at 1598 cm^{-1}, a characteristic C=N stretching of the diazepine ring system. The NH and OH stretching bands were found at 3232 and 2916 cm^{-1} respectively. In PMR spectrum, two broad humps at δ 3.63 and 4.0 for four protons (C-2 and C-3 diazepine ring), a sharp singlet at δ 5.37 for C-6 proton of diazepine ring and the NH proton at δ 8.3 as a broad singlet were observed. The eight aromatic protons appeared as a multiplet at δ 6.49-7.64 (8H, ArH). The M^+ peak was obtained at 298 (m/z) in its mass spectrum.



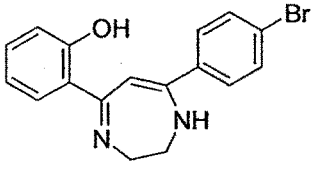
49i



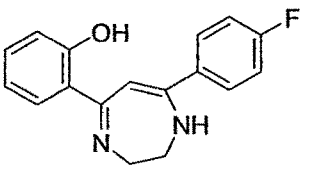
49j

5-(2-Hydroxyphenyl)-7-(2,4-dichlorophenyl)-2,3-dihydro-1*H*-1,4-diazepine **(49j)** gave a characteristic peak at 1598 (C=N), 3000 (OH) and 3087 cm^{-1} for NH stretching in its IR spectrum. In the PMR spectrum two separate broad humps were observed at δ 3.71 and 3.97 for C-2 and C-3 diazepine ring protons (4H) respectively. Sharp and broad singlets were obtained for C-6 proton of diazepine ring and NH proton at δ 5.41 and 7.54 respectively. The seven aromatic protons were displayed as a multiplet at δ 6.46-7.45. The M^+ peak was obtained at 333 (m/z) in its mass spectrum. Also, M+ 4 peak is observed due to the presence of two chlorine atoms.

5-(2-Hydroxyphenyl)-7-(4-bromophenyl)-2,3-dihydro-1*H*-1,4-diazepine **(49k)** gave a characteristic peak at 1635 (C=N), 3000 (OH) and 3064 cm^{-1} for NH stretching in the IR spectrum. In the PMR spectrum sharp singlet signal was obtained for C-6 proton of diazepine at δ 5.65. Two separate broad singlets observed at δ 3.69 and 3.89 for C-2 and C-3 diazepine ring protons (4H). The eight aromatic protons were displayed as a multiplet at δ 6.46-7.54 and the NH proton appeared at δ 8.11 as a broad hump. The M+2 peak was obtained at 345 (m/z) in its mass spectrum.



49k

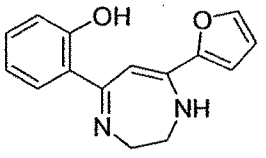


49l

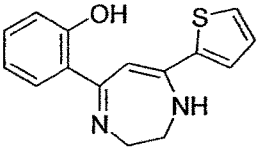
Compound **(49l)** showed infrared bands at 1604 cm^{-1}, a characteristic C=N stretching of the diazepine ring system. The NH and OH stretching bands were found at 3203 and 3000 cm^{-1} respectively. In the PMR spectrum, two broad humps at δ 3.65 & 3.90 for four protons (C-2 and C-3 diazepine ring), a sharp singlet at δ 5.68 for C-6 proton of diazepine ring and the NH proton at δ 8.38 as a broad singlet were observed. The eight aromatic protons appeared as a

multiplet at δ 6.5-7.8 (8H, ArH). The M^+ peak was obtained at 282 (m/z) in its mass spectrum.

Compound **(49m)** showed infrared bands at 1608 cm^{-1}, a characteristic C=N stretching of the diazepine ring system. The NH and OH stretching bands found at 3286 and 3000 cm^{-1} respectively. In the PMR spectrum, two broad humps at δ 3.65 and 3.9 for four protons (C-2 and C-3 diazepine ring), a sharp singlet at δ 6.1 for C-6 proton of diazepine ring and the NH proton at δ 8.28 as a broad singlet were observed. The seven aromatic protons appeared as a multiplet at δ 6.53-7.96 (7H, ArH). The M^+ peak was obtained at 254 (m/z) in its mass spectrum.



49m

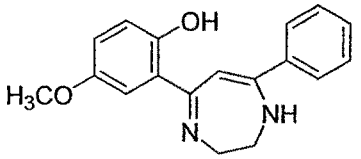


49n

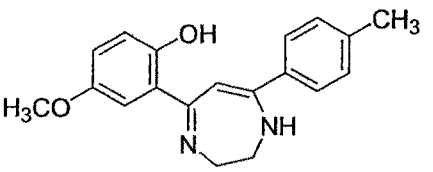
5-(2-Hydroxyphenyl)-7-(2-thienyl)-2,3-dihydro-1*H*-1,4-diazepine **(49n)** displayed a characteristic peak at 1595 (C=N), 3000 (OH) and 3203 cm^{-1} for NH stretching in IR spectrum. In the PMR spectrum two separate broad humps observed at δ 3.6 & 3.9 for C-2 and C-3 diazepine ring protons (4H). A sharp and broad singlet was obtained for C-6 proton of diazepine ring and NH proton at δ 5.92 and 8.3 respectively. The seven aromatic protons were displayed as a multiplet at δ 6.5-7.8.

5-(5-methoxy-2-hydroxyphenyl)-7-phenyl-2,3-dihydro-1*H*-1,4-diazepine **(49o)** gave a characteristic peak at 1633 (C=N), 3100 (OH) and 3423 cm^{-1} for NH stretching in the IR spectrum. Also, the asymmetric and symmetric peaks at 1255 and 1064 for OCH_3 stretching respectively. In the PMR spectrum a sharp singlet signal was obtained for C-6 proton of diazepine at δ 5.68. Two separate broad singlets observed at δ 3.65 and 3.80 for C-2 and C-3 diazepine ring protons (4H). The methoxy protons are obtained as a singlet at δ 3.71. The eight aromatic protons were displayed as a multiplet at δ 6.70-7.55 and the NH

proton appeared at δ 7.57 as a broad singlet. The M+1 peak was obtained at 295 (m/z) in its mass spectrum.



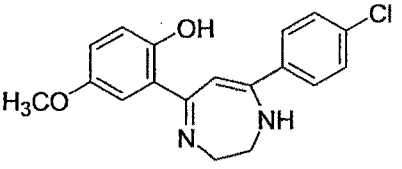
49o



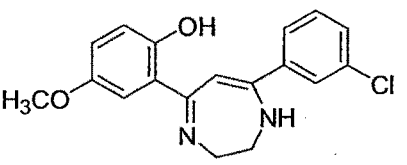
49p

Compound **(49p)** showed infrared bands at 1608 cm^{-1}, a characteristic C=N stretching of the diazepine ring system. The NH and OH stretching bands were found at 3288 and 3000 cm^{-1} respectively. Also, the asymmetric and symmetric peaks at 1218 and 1040 for OCH_3 stretching respectively. In PMR spectrum, two broad humps at δ 3.69 and 3.78 for four protons (C-2 and C-3 diazepine ring), a sharp singlet at δ 5.68 for C-6 proton of diazepine ring and the NH proton at δ 7.49 as a broad singlet were observed. Two separate singlets were found for methyl and methoxy protons at δ 2.38 and 3.71 respectively. Seven aromatic protons showed a multiplet at δ 6.75-7.47 (7H, ArH). The M+1 peak was obtained at 309 (m/z) in its mass spectrum.

5-(5-Methoxy-2-hydroxyphenyl)-7-(4-chlorophenyl)-2,3-dihydro-1*H*-1,4-diazepine **(49q)** gave a characteristic peak at 1271 and 1095 (OCH_3), 1595 (C=N), 3000 (OH) and 3299 cm^{-1} for NH stretching in IR spectrum. In the PMR spectrum a sharp singlet signal was obtained for C-6 proton of diazepine at δ 5.65. Two separate broad singlets observed at δ 3.69 and 3.92 for C-2 and C-3 diazepine ring protons (4H). A sharp singlet appeared at δ 3.73 for methoxy protons. The seven aromatic protons were displayed as a multiplet at δ 6.77-7.58 and the NH proton appeared at δ 7.87 as a broad singlet.



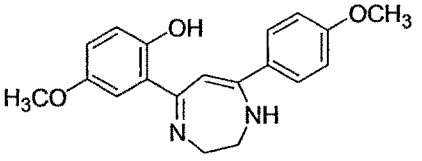
49q



49r

Compound **(49r)** showed infrared bands at 1612 cm^{-1}, a characteristic C=N stretching of the diazepine ring system. The NH and OH stretching bands were found at 3257 and 3000 cm^{-1} respectively. Also, the asymmetric and symmetric peaks were seen at 1218 and 1040 for OCH_3 stretching respectively.

5-(5-Methoxy-2-hydroxyphenyl)-7-(4-methoxyphenyl)-2,3-dihydro-1*H*-1,4-diazepine **(49s)** showed infrared bands at 1604 cm^{-1}, a characteristic C=N stretching of the diazepine ring system. Asymmetric and symmetric bands were observed at 1257 and 1033 cm^{-1} for OCH_3 stretching respectively. The NH and OH stretching bands found at 3222 and 3000 cm^{-1} respectively.



49s

In PMR spectrum, two broad humps at δ 3.66 and 3.90 for four protons (C-2 & C-3 diazepine ring), a sharp singlet at δ 5.69 for C-6 proton of diazepine ring and the NH proton at δ 7.88 as a broad singlet were observed. A sharp singlet at δ 3.72 appeared for six integral protons due to $(OCH_3)_2$. The seven aromatic protons showed a multiplet at δ 6.75-7.47 (7H, ArH). The M+ 1 peak was obtained at 325 (m/z) in its mass spectrum.

3.2 Biological Studies

The biological work carried out has been discussed under following sub heads:

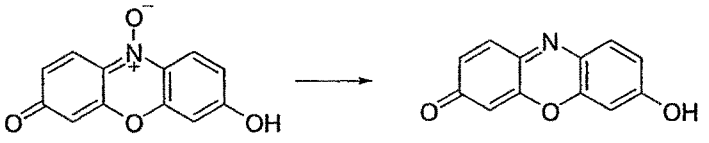
A. Antiproliferative activity

B. Antiplatelet activity

A. Antiproliferative activity

Initial evaluation of 2,3-diaryl-6,7-dihydro-5*H*-1,4-diazepines and 5,7-diaryl-2,3-dihydro-1*H*-1,4-diazepines were carried out at Memorial Sloan Kettering Cancer Center (MSKCC), New York (USA). The cytotoxic effects of the above

said compounds were tested using Alamar blue assay[196]. Alamar blue (Resazurin) is nontoxic, commonly employed as an indicator of cell number and viability, since it is reduced to a (Resorufin) pink fluorescent dye in the medium by cell activity (possibly by oxygen consumption through metabolism). Alamar blue reduction was measured on CCD-based optical imaging reader.



Resazurin (blue) Resorufin (pink)

The various human leukemic cancer cell lines Jurkat, HL60, Molt3, NCEB-1 and K562 were incubated with various concentrations of the 2,3 and 5,7-diaryldiazepines for 72 h (48h plus 24h with dye). The results of IC_{50} values of the compound are summarized in **Table 4 and 5.**

Table 4: Antiproliferative activity IC_{50} values of 2,3-diaryl-6,7-dihydro-5*H*-1,4-diazepines (44a-r)

Compound	Jurkat	HL60	Molt-3	NCEB-1	K562
44a	>100	>100	>100	>100	>100
44b	48.6	45	31.25	70	64
44c	40.59	41	24.37	54	49
44d	71.87	59	82	75	68
44e	NT	>100	NT	>100	>100
44f	NT	>100	NT	>100	>100
44g	30.1	39	22.6	>100	27.22
44h	51.78	47	27.6	54	39.19
44i	33.72	46	22.47	57	43.83
44j	26.67	30	21.4	40	36.59
44k	29	40	22.64	56	44
44l	48.26	67	28.75	>100	51.54
44m	39.73	32	25.13	65	49.18
44n	>100	>100	>100	>100	>100
44o	44	38	21	45	37.79
44p	NT	>100	NT	82.88	>100
44q	NT	>100	NT	>100	>100
44r	NT	>100	NT	>100	>100

Table 5: Antiproliferative activity IC_{50} values of 5,7-diaryl-2,3-dihydro-1*H*-1,4-diazepines (49a-s)

Compound	Jurkat	HL60	Molt-3	NCEB-1	K562
49a	10.59	60	11.4	>100	9.12
49b	50.79	>100	40.81	86	24.3
49c	8.47	22	11.6	100	4.55
49d	>100	>100	31	>100	20.85
49e	5.39	23	14.33	26	8.28
49f	N. T	>100	N. T	>100	>100
49g	81.58	>100	64	>100	65
49h	>100	>100	>100	>100	>100
49i	7.66	26	12.09	23	10.12
49j	N. T	>100	N. T	>100	>100
49k	N. T	>100	N. T	>100	>100
49l	22.75	80	38.48	70	29.46
49m	>100	>100	>100	>100	88.78
49n	N. T	>100	N. T	>100	>100
49o	N. T	88.24	N. T	>100	85.26
49p	N. T	>100	N. T	>100	>100
49q	N. T	>100	N. T	>100	>100
49r	N. T	N. T	N. T	N. T	N. T
49s	N. T	>100	N. T	>100	>100

N.T = Not tested

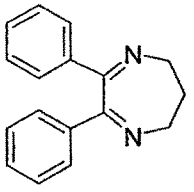
Looking to the results of antileukemic activity of 2,3-diaryl-6,7-dihydro-5*H*- 1,4-diazepines **(44a-r)** and 5,7-diaryl-2,3-dihydro-1*H*- 1,4-diazepines **(49a-s)** it is reflected that the former category of compounds do not show any significant activity. In comparison, among the 5,7-diaryl-2,3-dihydro-1*H*- 1,4-diazepines **(49a-s)** compound **49a** showed significant cytotoxic activity with IC_{50} values of 9.12 and 10.59 μM against K562 and Jurkat cell lines respectively. Introduction of methyl group at meta position of the diaryldiazepine nucleus as in compound **49c,** demonstrated high cytotoxic activity against the K562 and Jurkat cell lines with an IC_{50} values of 4.55 and 8.47 μM respectively. Compound **49e,** in which the methyl group was replaced with a greater electron donating group, a methoxy group, was also found to be cytotoxic but significantly less so

than the compound **49c**. Results indicate that among the halo substituted diaryldiazepines, compound **49i**, bearing a chloro substituent at ortho position showed significant activity with IC_{50} value of 7.66 μM against Jurkat cell line. Substitution of methoxy at 5th position of the phenyl ring attached to the 4th position of the dazepine does not show any significant activity **(49o-s)**. Also, there was no interesting activity when one of the aryl rings was replaced by furan **(49m)** and thiophene ring **(49n)**.

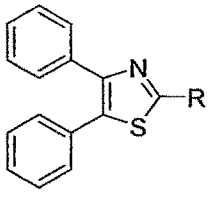
B. Antiplatelet activity

The reason for evaluation of antiplatelet activity for 2,3-diaryl-6,7-dihydro-5*H*-1,4-diazepine **(44a-r)** in this study was due to the following reason.

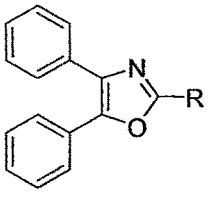
Looking to the chemical features of 2,3-diaryl-1,4-diazepines, the antiplatelet activity can be speculated, as the compounds possess both the features for such activity reported in case of certain diazepines (described in the introduction chapter) and some diaryl heterocycles[197-200] **(A-C)**.



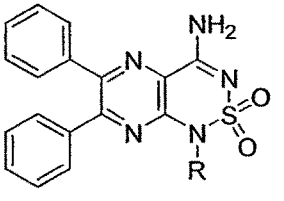
44a-r



A



B



C

The 2,3-diaryl-6,7-dihydro-5H-1,4-diazepines **(44a-r)** were evaluated to inhibit platelet aggregation of female Sprague-Dawley rat platelet-rich plasma induced by arachidonic acid[201]. 500 μM of test compounds **(44a-r)** were used in the

preliminary screening. Aspirin was used as standard drug and showed maximum inhibition (97%) at 10mM. In this screening **44c, 44d, 44h, 44i, 44k, 44m** and **44o** were found to be moderately active **(Table 6)**.

Table 6 Antiplatelet activity (500 µM)

Compound	% inhibition
44a	18.5
44b	35.7
44c	42.5
44d	60.7
44e	35.7
44f	34
44g	34
44h	44.3
44i	77.8
44j	36.4
44k	44.6
44l	43
44m	52.6
44n	22
44o	58.2
44p	N.T
44q	22.1
44r	0

N.T = Not tested

Hence, these compounds were subjected to further screening at low concentrations (100 μM). The results of the screening are displayed in **Table 7**.

Table 7 Antiplatelet activity (100 μM)

Compound	% inhibition
44c	18.5
44d	18.8
44h	24
44i	36.4
44k	24
44m	24.2
44o	28.3
Aspirin	14

From these studies, **44i** was the most active one with 36.4% inhibition. Compound **44o** showed 28.3% inhibition with two fold greater activity as compared to aspirin (14%). The compound **44h, 44k** and **44m** showed moderately good activity.