CHAPTER - III

Results And Discussion

A. Chemical Work

B. Biological Studies

RESULTS AND DISCUSSION

A. CHEMICAL WORK

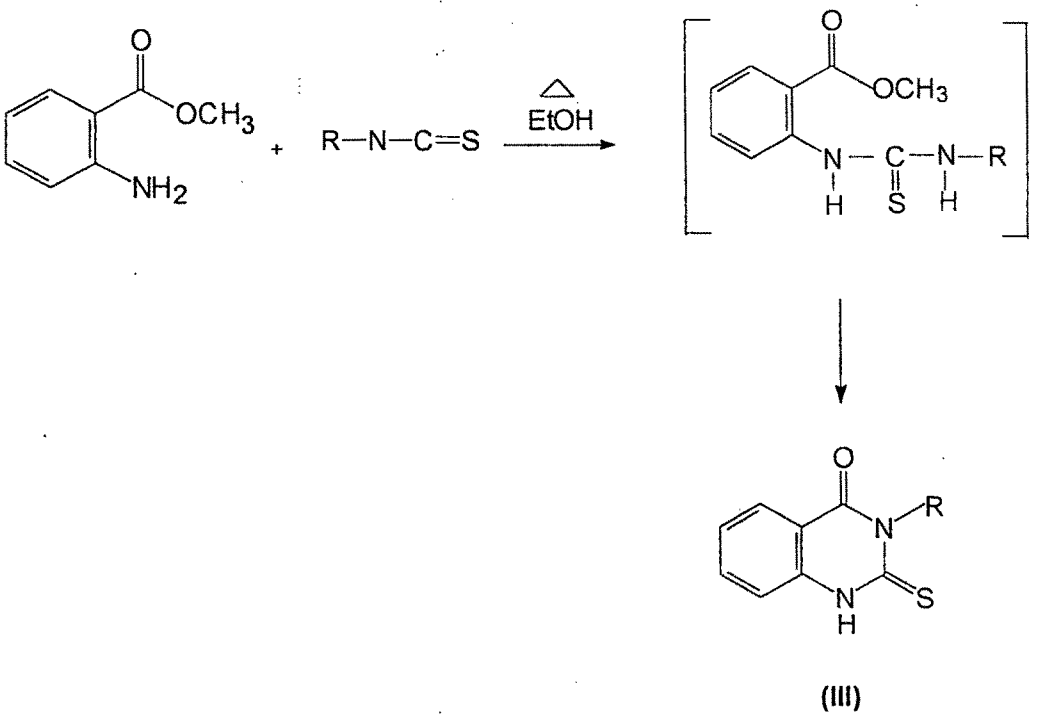
The results of the present work is discussed under the following heads

- **Synthesis of 2-thioxo-3-substituted quinazolin-4-(3*H*)-ones (III)**
- **Synthesis of 2-methylthio-3-substituted quinazolin-4(3*H*)-ones (IV)**
- **Synthesis of 2-hydrazino-3-substituted quinazolin-4(3*H*)-ones (V)**
- **Synthesis of 1-substituted-4-aryl-*s*-triazolo[4,3-*a*]quinazolin–5(4*H*)-ones (1-30)**

Synthesis of 2-thioxo-3-substituted quinazolin-4-(3*H*)-ones (III)

METHOD-I

The 2-thioxo-3-substituted quinazolin-4-(3*H*)-one **(III)** was obtained by refluxing methylanthranilate with arylisothiocyanates in ethanol (Scheme-I).



(III)

SCHEME-I

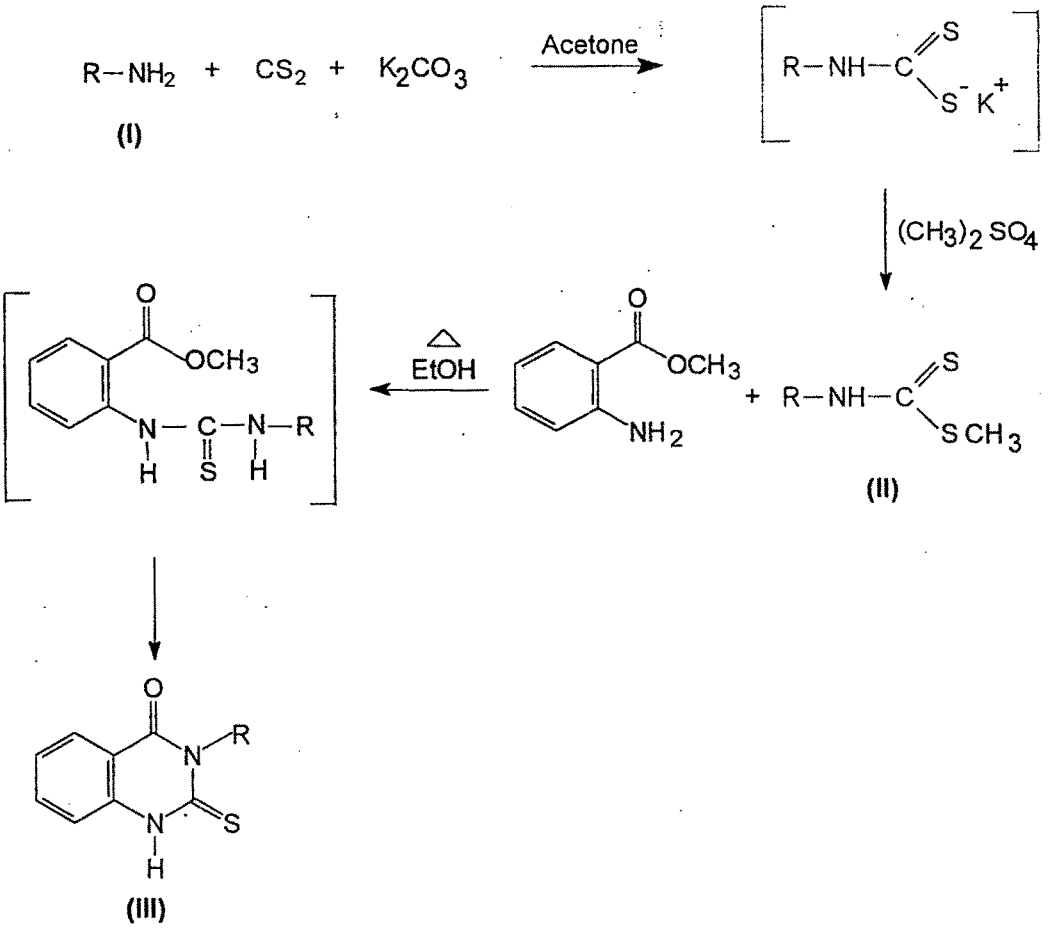
The synthesis of 2-thioxo-3-substituted quinazoline **(III)** by the described route have the following disadvantages:

i) Preparation of arylisothiocyanates required for the reaction is a tedious and and time consuming process.

ii) The yield was also low (60-70%)

Hence an alternate route was attempted to synthesize the 2-thioxo-3-substituted quinazoline **(III)** as depicted in Scheme-II

METHOD-II



SCHEME II

This process of synthesizing **III** by this scheme suffers from the following drawbacks

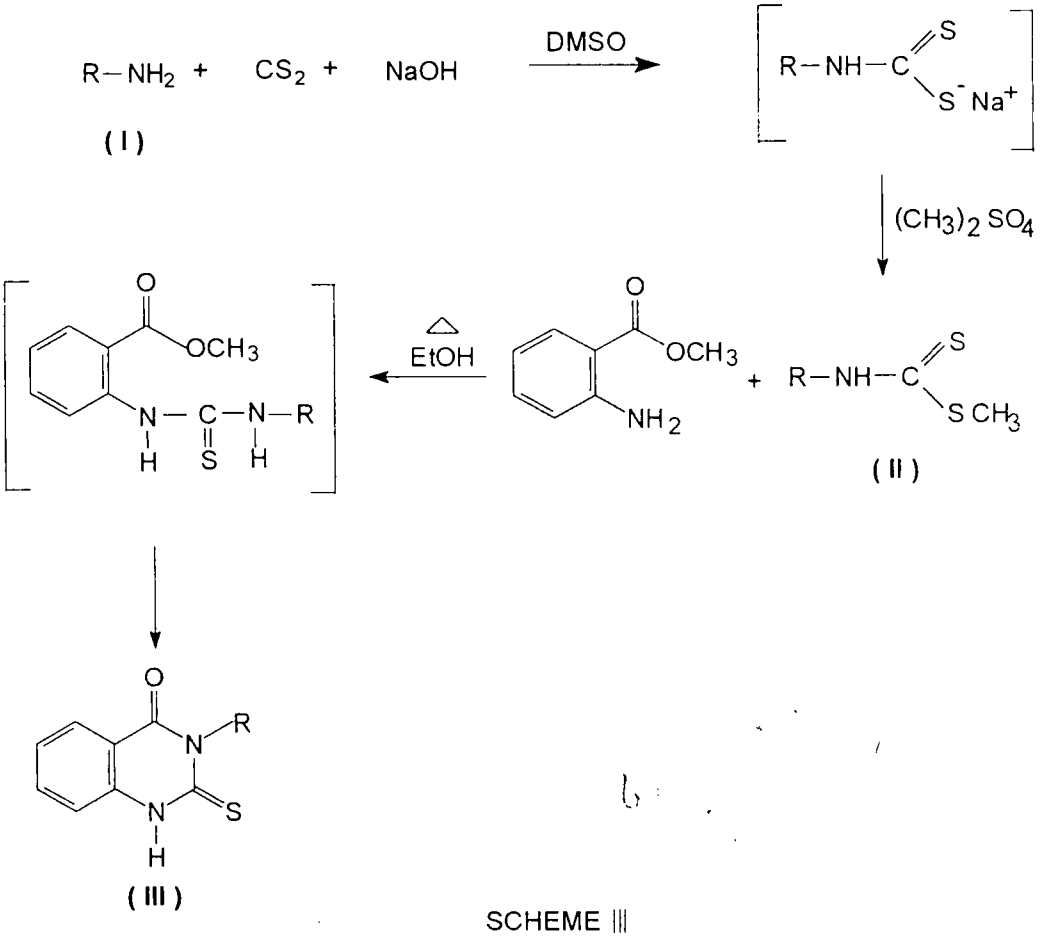
i) It is a multistep process

ii) It requires prolonged reaction time (37 h)

iii) The yield is also very low (20-30%)

METHOD-III

Hence, improvisation was carried out on this method (Scheme-III). Aq. NaOH (20 molar solution) was used as a base instead of anhydrous K_2CO_3 and dimethyl suphoxide (DMSO) was substituted for acetone as the reaction solvent. The use of DMSO as the reaction solvent enhanced the rate of reaction and the use of alkali in higher concentration helped in preventing the hydrolysis of the intermediate probably, due to less solvation.

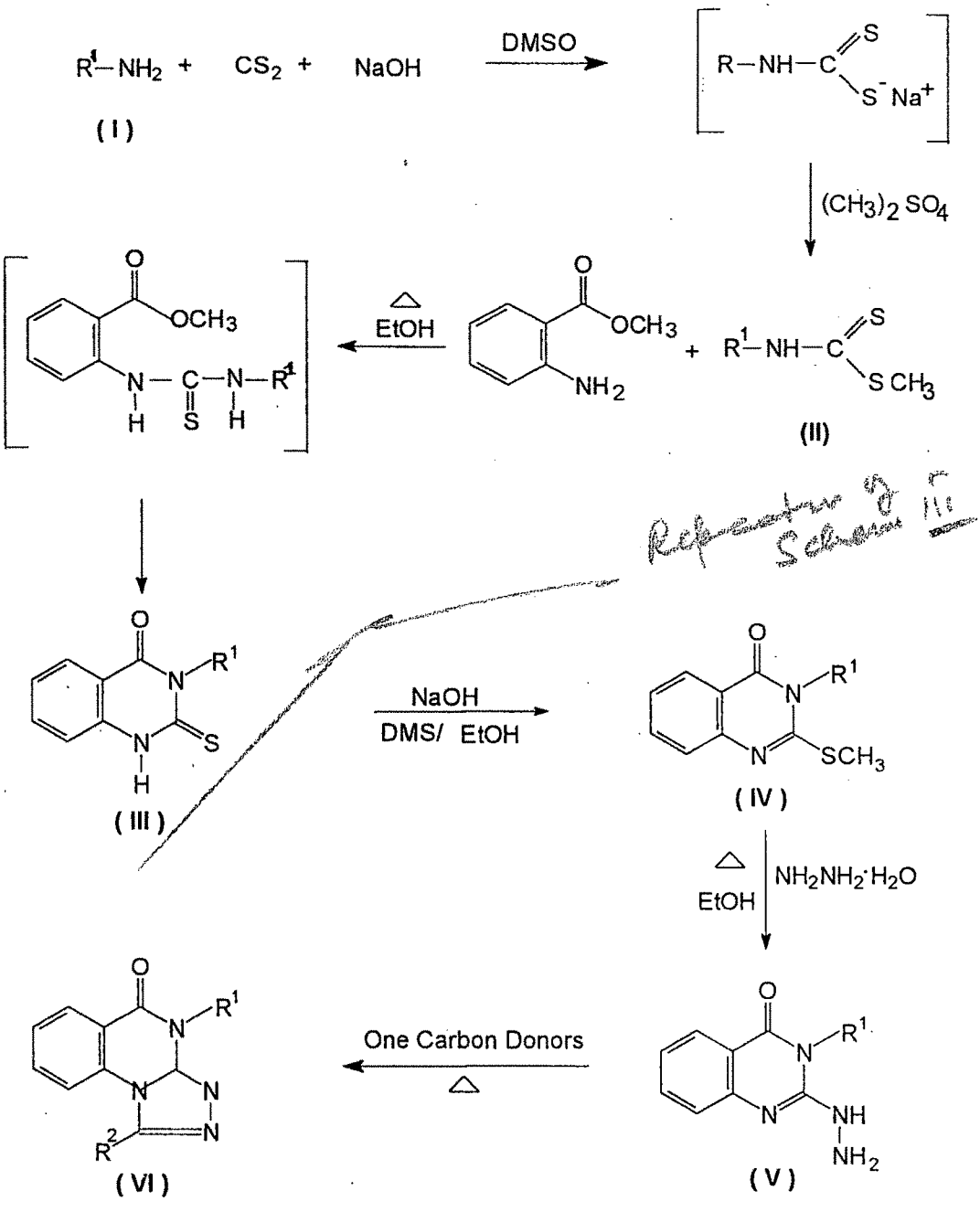


SCHEME III

The aromatic primary amine **(I)** was reacted with carbon disulphide to give dithiocarbamate salt which was methylated with dimethylsulphate to afford the dithiocarbomic acid methyl ester **(II)**. Compound **II** on reflux with methylanthranilate in ethanol yielded the desired 2-thioxo-3-substituted quinazoline **(III)** via the thiourea intermediate in good yield (around 80%)

The product obtained was cyclic and not an open chain thiourea. It was confirmed by its low Rf value, high melting point and its solubility in sodium hydroxide solution. The IR spectrum of these compounds show intense peaks around 3220 cm^{-1} for amino (NH), 1660 cm^{-1} for carbonyl (C=O) and 1200 cm^{-1} for thioxo (C=S) stretching. In the NMR spectrum thioxo derivatives showed signals at δ 7-9 (m, -ArH) and 10.5 (s, 1*H*, -NH).

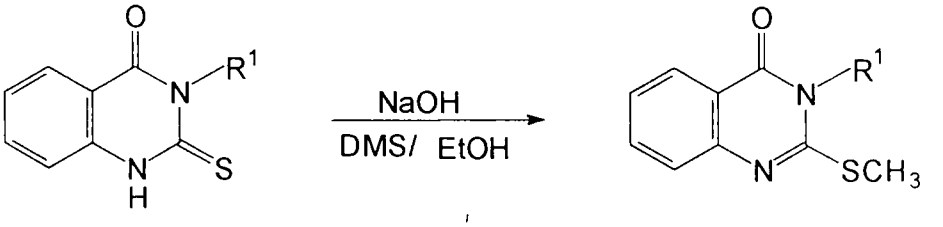
The synthetic route depicted in Scheme-IV outlines the general path adopted in the present work.


R–NH2 + CS2 + NaOH
DMSO
(I)
(CH3)2 SO4
EtOH
OCH3
NH2
(II)
NaOH
DMS/ EtOH
(III)
(IV)
EtOH
NH2NH2·H2O
One Carbon Donors
(V)
(VI)

(SCHEME-IV)

Synthesis of 2-methylthio-3-substituted quinazolin-4(3*H*)-ones (IV)

The 2-methylthio derivative **(IV)** was obtained by dissolving **III** in 2% alc. NaOH solution and methylating with dimethyl sulphate with stirring at room temperature.

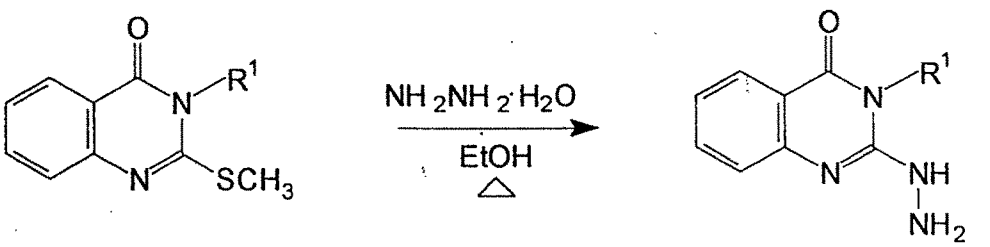


(III) (IV)

The IR spectrum of the 2-methylthio derivative **(IV)** showed disappearance of amino (-NH) and thioxo (C=S) stretching signals of the starting material. It showed a peak for carbonyl (C=O) stretching around 1680 cm^{-1}. The NMR spectrum of compound **IV** showed signals around δ2.5 (s, 3*H*, $-SCH_3$) and 7-8.6 (m, -ArH).

Synthesis of 2-hydrazino-3-substituted quinazolin-4(3*H*)-ones (V)

Nucleophilic displacement of methylthio group with hydrazine hydrate was carried out using ethanol as solvent. The long duration of reaction (22 to 31 h) required may be due to the presence of bulky aromatic ring at position 3, which might have reduced the reactivity of quinazoline ring system at 2 position.



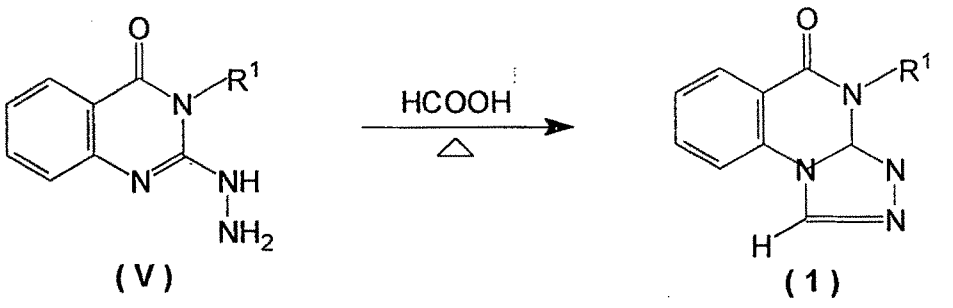
(IV) (V)

The formation of **V** was confirmed by the presence of amino ($-NHNH_2$) signals around 3334–3280 cm^{-1} in the IR spectrum. It also showed a peak for carbonyl (C=O) around 1680 cm^{-1}. The NMR spectrum of the compound **V** showed signals at or around δ 5.0 (s, 2*H*, $-NHN\underline{H_2}$), 7.0–8.1 (m, -ArH) and 8.7 (s, 1*H*, $-N\underline{H}NH_2$).

Synthesis of 1-substituted-4-aryl-*s*-triazolo[4,3-*a*]quinazolin-5(4*H*)-ones (1-30).

The cyclization of 2-hydrazino-3-substituted quinazolin-4(3*H*)-ones to 1-substituted-4-aryl-*s*-triazolo[4,3-*a*]quinazolin-5(4*H*)-ones was studied with various one carbon donors like formic acid and acetic acid etc.

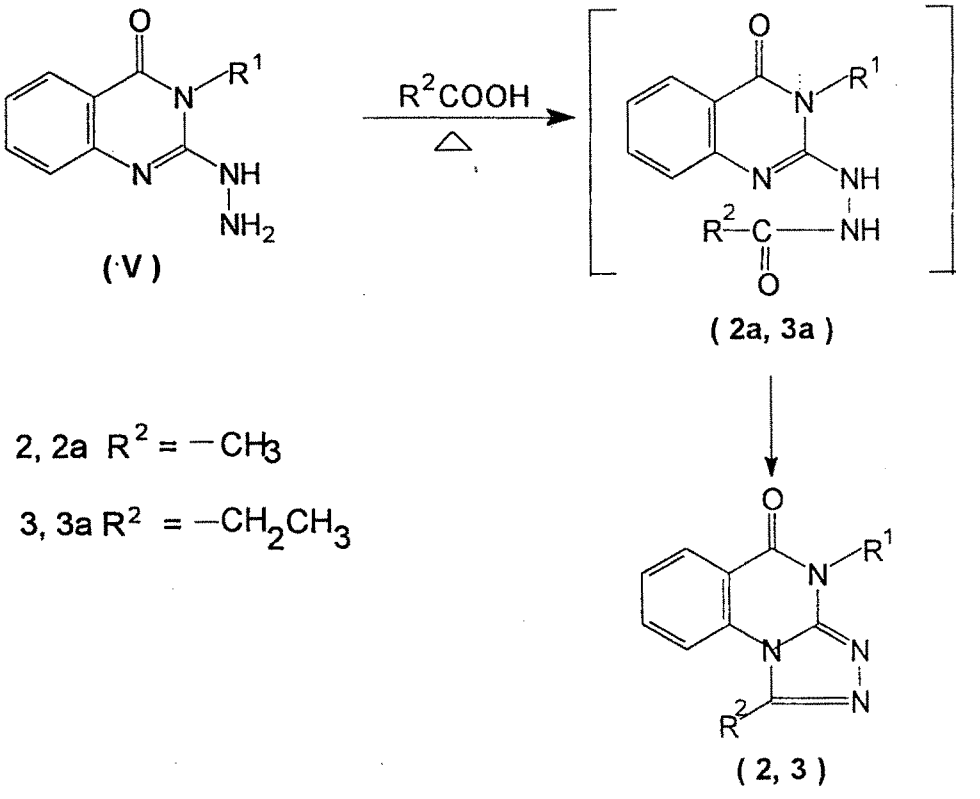
The reaction of **V** with formic acid at reflux temperature yielded the expected triazoloquinazolin-5(4*H*)-one **(1)**.



(V) (1)

The formation of cyclic product is indictated by the disappearance of peaks due to NH and NH_2 at 3400–3200 cm^{-1} in IR spectrum of all the compounds **(1-30)**. It was also characterized by the NMR spectrum of **1** which showed the absence of NH and NH_2 signals around δ 5.0 (s, 2*H*, -NHN<u>H</u>$_2$) and 8.7 (s, 1*H*, -N<u>H</u>NH_2). A multiplet at 7–8.0 integrating for aromatic protons was observed. Data from the elemental analyses have been found to be in conformity with the assigned structure. Further the molecular ion recorded in the mass spectrum is also in agreement with the molecular weight of the compound.

Analogously the reaction of hydrazine derivative **V** with acetic acid and propionic acid yielded desired 1-methyl-4-substituted-*s*-triazolo [4,3-*a*]quinazolin-5(4*H*)-one **(2)** and 1-ethyl-4-substituted-*s*-triazolo [4,3-*a*]quinazolin-5(4*H*)-one **(3)** respectively. The intermediates acetyl amine **(2a)** and propionyl amine **(3a)** could not be isolated.

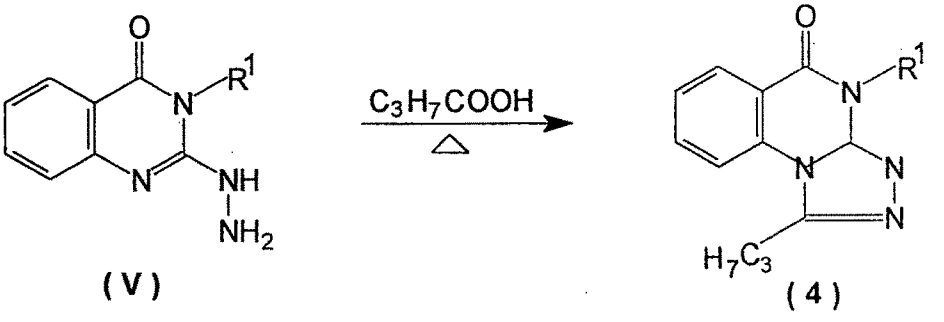


Scheme V

The compounds **2** and **3** was characterized by IR and NMR spectra. In the IR and NMR spectrum of the compound **2** and **3** disappearance of NH and NH_2 signals of starting material confirms the cyclization. The NMR spectrum of compound **2** showed characteristic signal for methyl ($-CH_3$) δ 2.5 (s, 3*H*,) and a multiplet at 7-8 integrating for aromatic protons. The NMR spectrum of compound **3** showed characteristic signal for ethyl ($-CH_2-CH_3$) around δ 1.3-1.4 (t, 3*H*, $-CH_2\underline{CH_3}$) and δ 2.7-2.8 (q, 2*H*, $-\underline{CH_2}CH_3$) and a multiplet around 7.1-8.5 integrating for aromatic protons. Its data from the elemental analyses have been found to be in conformity with the assigned structure. Further

the molecular ion recorded in the mass spectrum is also in agreement with the molecular weight of the compound.

Similarly the reaction of **V** with butyric acid at reflux temperature in presence of a few drops of concentrated sulphuric acid also led directly to the isolation of the product.

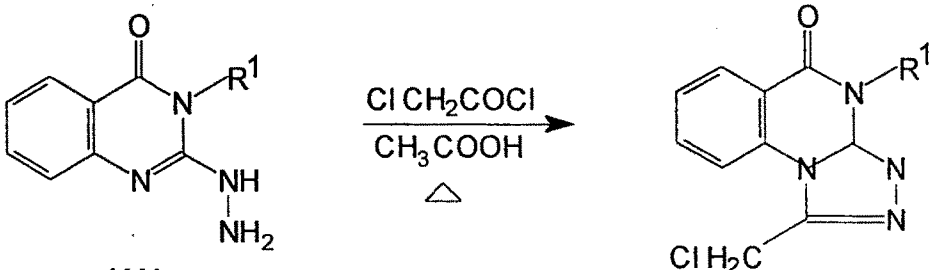


(V) (4)

The IR and NMR spectrum of the title compounds showed the disappearance of peaks due to NH and NH_2 confirms the cyclization. The IR spectrum of **4** showed a peak around 1680-1690 cm^{-1} for carbonyl (C=O). In the NMR spectrum of **4** signals were seen for propyl ($-CH_2CH_2CH_3$) around δ 0.9-1.0 (t, 2*H*, $-\underline{CH_2}CH_2CH_3$), δ 1.7-1.8 (hext, 2*H*, $-CH_2\underline{CH_2}CH_3$), 2.7-2.8 (t, 3*H*, $-CH_2CH_2\underline{CH_3}$) and a multiplet around 7.2-8.5 for aromatic protons. Its data from the elemental analyses have been found to be in conformity with the assigned structure. Further the molecular ion recorded in the mass spectrum is also in agreement with the molecular weight of the compound.

As the moleculer weight of the carboxylic acids increased the reactivity decreased and hence the reaction time required for cyclization increased.

Hydrazino compound **V** could be readily cyclized to the desired 1-chloromethyl-4-substituted-*s*-triazolo[4,3-*a*]quinazolin-5(4*H*)-ones **(5)** by its reaction with chloroacetyl chloride in acetic acid medium.



(V) **(5)**

The compound **5** was characterized by IR and NMR spectra. The IR and NMR spectra of **5** showed the absence of signals for amino (-$NHNH_2$) which confirms the cyclization. The IR spectrum of **5** showed a peak around 1690-1700 cm^{-1} for carbonyl (C=O). In the NMR spectrum signals were seen around δ 2.0 (s, 2*H*) for -CH_2 of -CH_2Cl and a multiplet around 7.3-8.0 integrating for aromatic protons. Its data from the elemental analyses have been found to be in conformity with the assigned structure. Further the molecular ion recorded in the mass spectrum is also in agreement with the molecular weight of the compound.

BIOLOGICAL STUDIES

B.1 Alpha adrenergic blocking activity

B.1.1 *In vivo study*

B.1.1.1 Pressor Responses To Various Agonists After Intravenous Administration of Test Compound (24)

The experiment with the test compound was carried out at different doses (2, 6, 10 and 20 mg/kg). From the study it was found that at a dose of 2 mg/kg of the test compound (**24**), the pressor effect of adrenaline (1µg/Kg, iv), noradrenaline (1µg/kg, iv), angiotensin-II (100 ng/kg, iv) and depressor response of histamine (1µg/kg, iv) were not modified (Fig 1A, 2A, 3A, 4A). There was a significant ($p<0.01$) blockade response of adrenaline and noradrenaline by test compound (**24**) at the dose of 6 mg/kg (Fig 1B, 2B). The pressor response of angiotensin-II (100 ng/kg) and depressor response of histamine (1µg/kg, iv) were not modified at 6 mg/kg dose of **24** (Fig 3B,4B). After the administration of **24** (10 and 20 mg/kg) to anesthetized rats, there was a complete reduction of pressor response of adrenaline (1µg/kg) and noradrenaline (1µg/kg) (Fig 1C, 1D, 2C and 2D) and adrenaline reversal was also produced (Fig 6). The blood pressure response to higher doses of adrenaline (5 µg/kg) and noradrenaline (5 µ/kg) were significantly ($P<0.01$) blocked by higher doses of **24** (10, 20 mg/kg). Response to angiotensin-II and histamine were not blocked by any of the dose studied (Fig 3C, 3D, 4C and 4D). The effect of DMSO (Dimethyl sulfoxide) and **24** on normal systolic blood pressure was studied

in anesthetized rats. There was a reduction in the blood pressure after administration of DMSO and also

there was significant reduction in the blood pressure after administration of **24** (10 and 20 mg/kg) as compared to corresponding control response (Fig 5).

B.1.1.1.1 Effect of Adrenaline Before And After Intravenous Administration of 24 on Rat Blood Pressure

Figure 1: Mean change in blood pressure by acute intravenous injection of adrenaline (μg/kg) in anesthetized rats before and after intravenous **24** (2, 6, 10 and 20 mg/kg). Abscissa indicates doses of adrenaline and ordinate mean change in blood pressure (mm Hg). Vertical line represents SEM (n = 5)

Fig 1A = $p>0.5$(NS)

1B = ** $p<0.01$

1C and 1D = *** $p<0.001$ as compared with corresponding control

Figure 1A

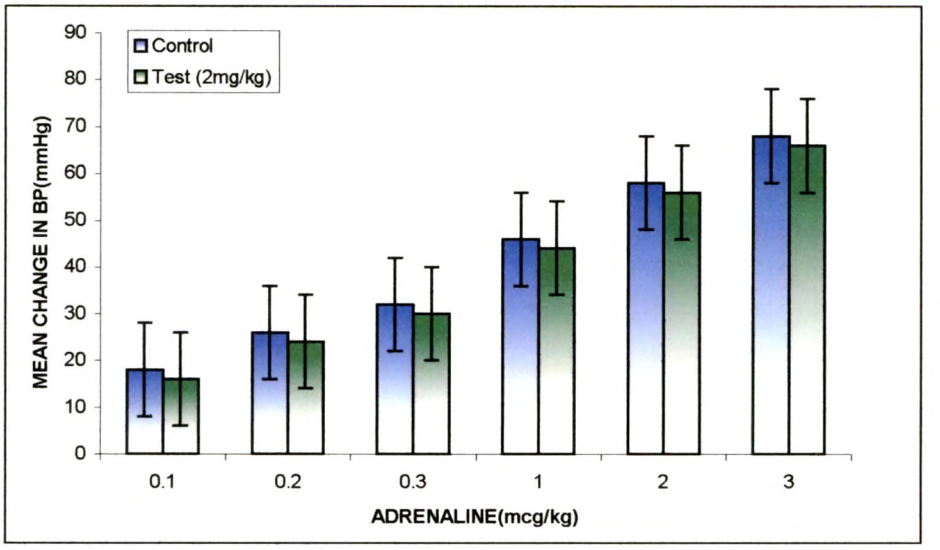


Figure 1B

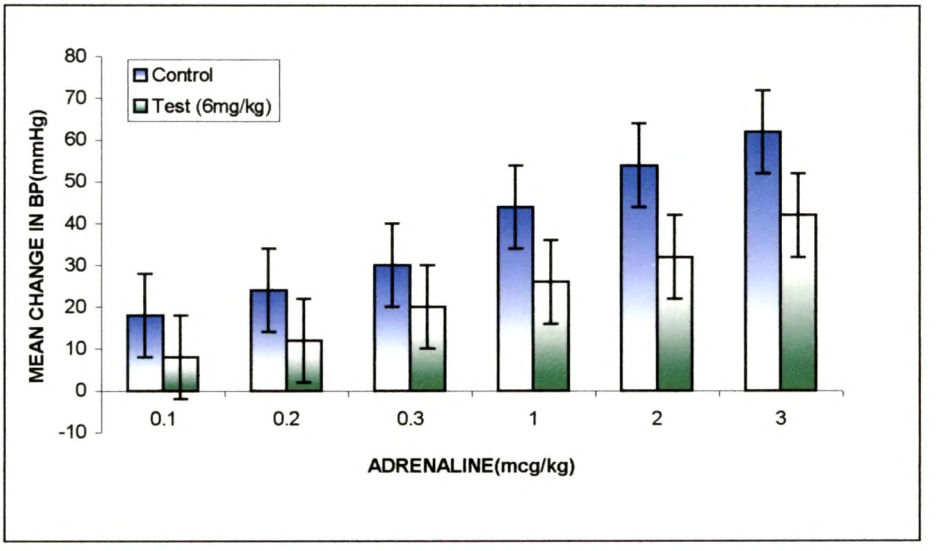


Figure 1C

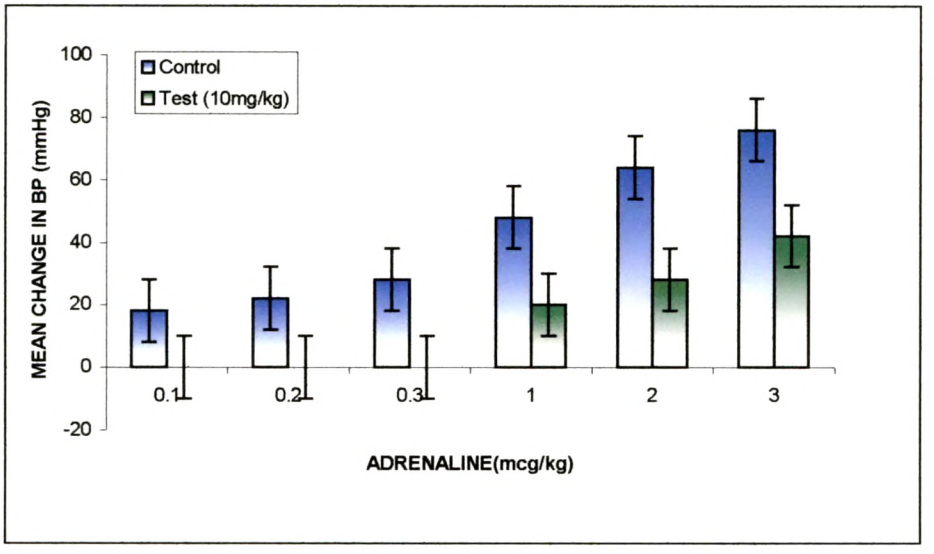
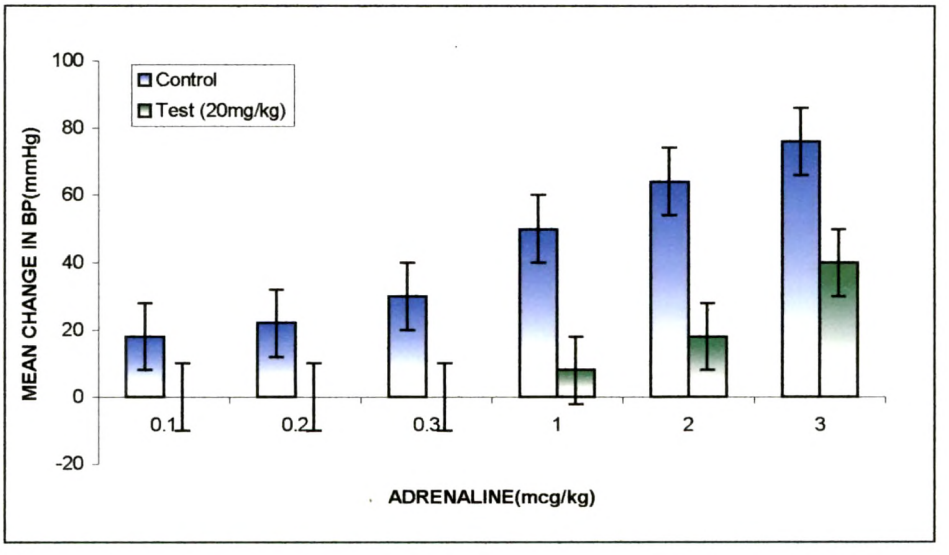


Figure 1D



B.1.1.1.2 Effect of Noradrenaline Before And After Intravenous Administration of 24 on Rat Blood Pressure

Figure 2: Mean change in blood pressure by acute intravenous injection of noradrenaline (µg/kg) in anesthetized rats before and after intravenous **24** (2, 6, 10 and 20 mg/kg). Abscissa indicates doses of NA and ordinate mean change in blood pressure (mm Hg). Vertical lines represent SEM (n = 5).

Fig 2A = $p>0.05$ (NS)

2B =** $p<0.01$

2C = *** $p<0.001$

2D =*** $p< 0.001$ as compared with corresponding control

Figure 2A

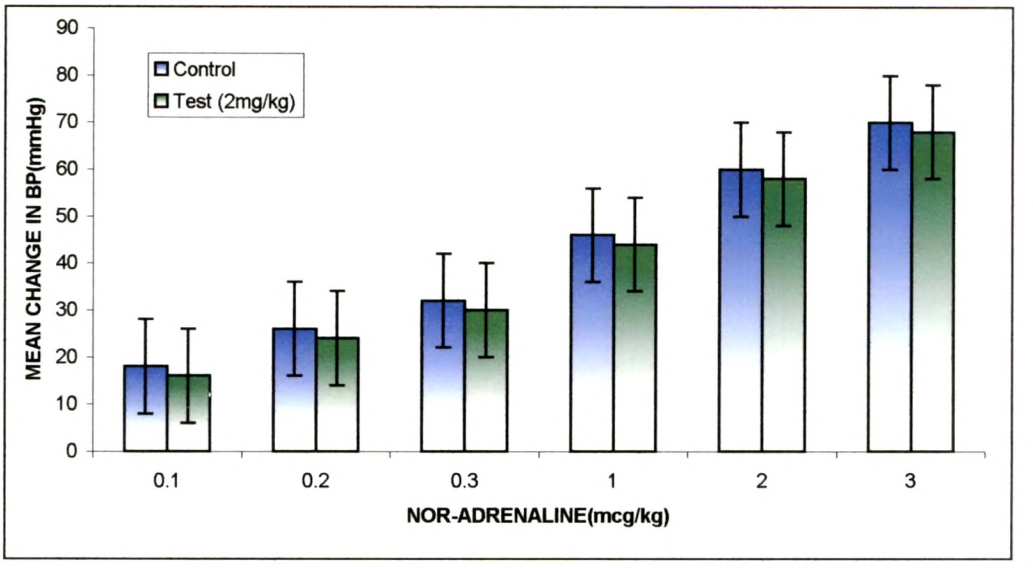


Figure 2B

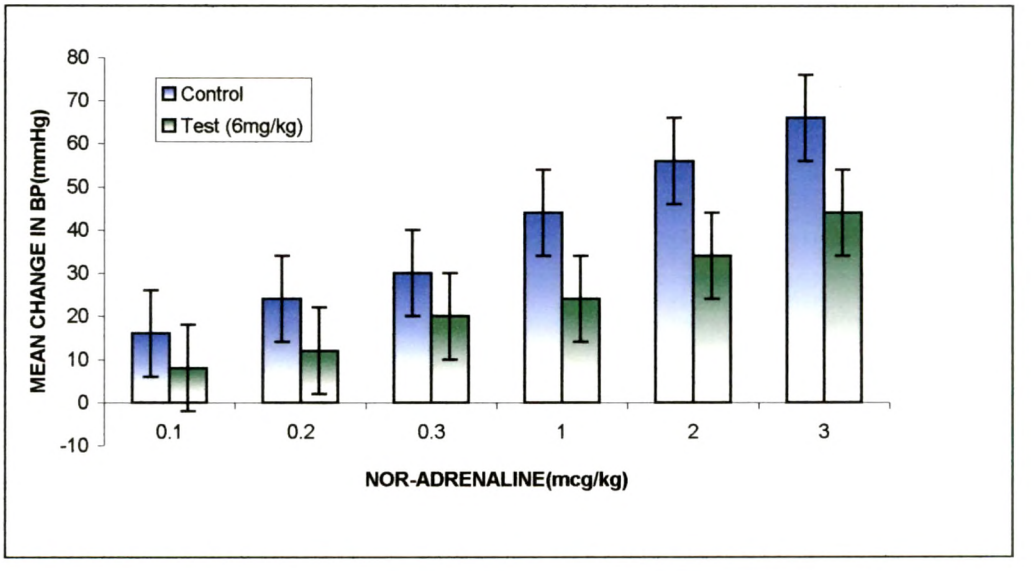


Figure 2C

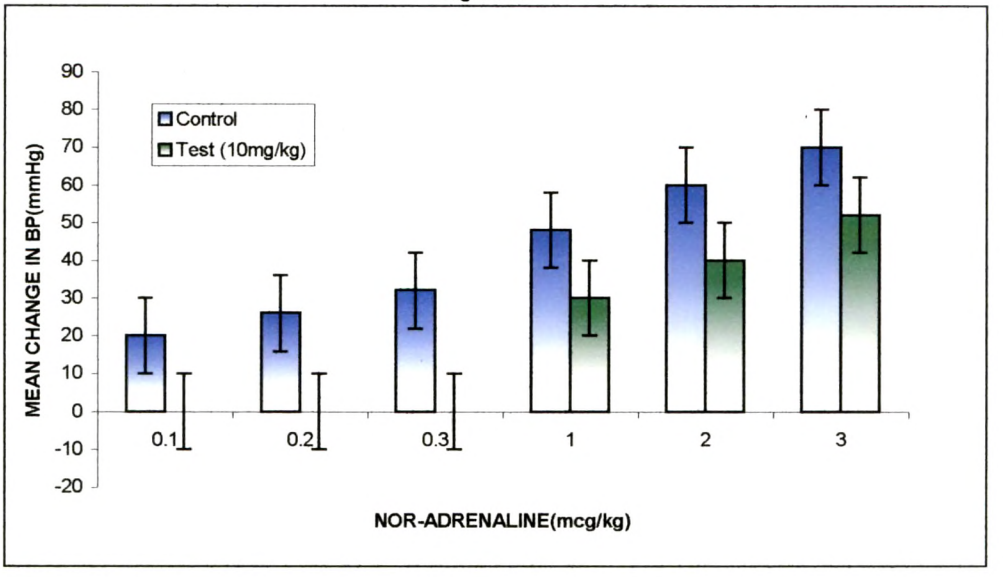
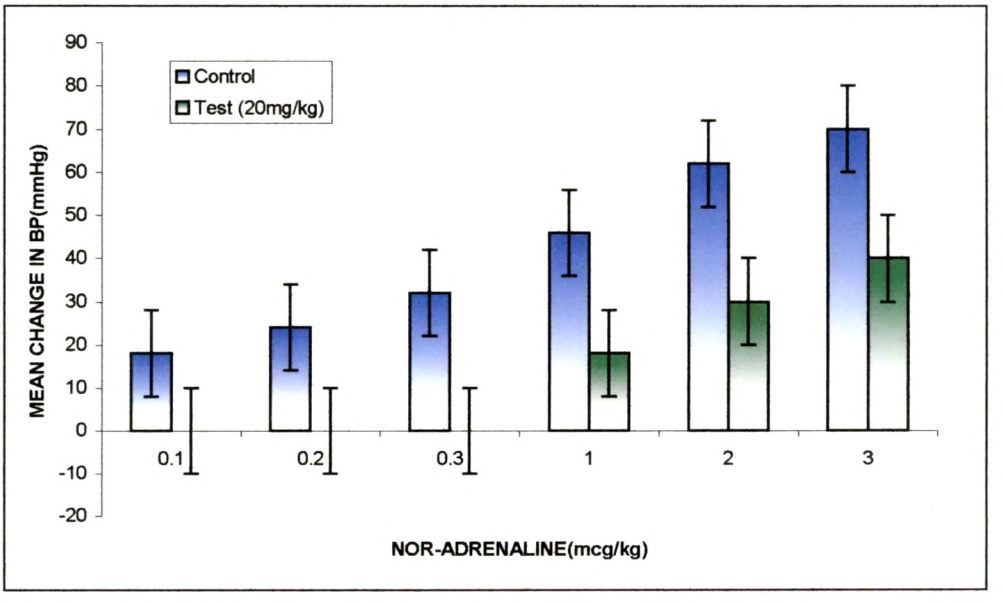


Figure 2D



B.1.1.1.3 Effect of Angiotensin-II Before And After Intravenous Administration of 24 on Rat Blood Pressure

Figure 3: Mean change in blood pressure by acute intravenous injection of angiotensin-II (10, 20, 30 ng) in anaesthetized rats before and after intravenous **24** (2, 6, 10 and 20 mg/kg). Abscissa indicates doses of angiotensin-II and ordinate indicates mean change in blood pressure (mm Hg). Vertical line indicates SEM (n = 5).

Figure 3A

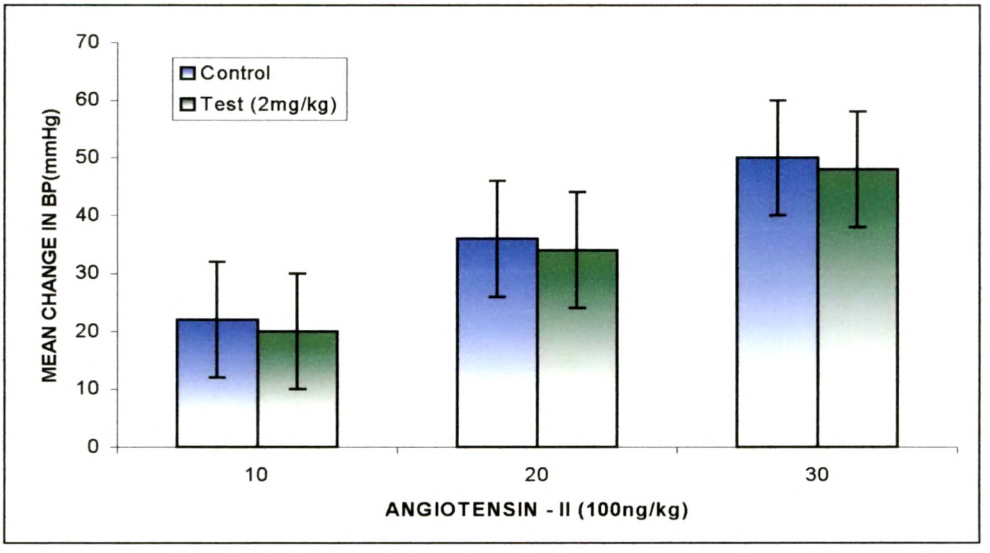


Figure 3B

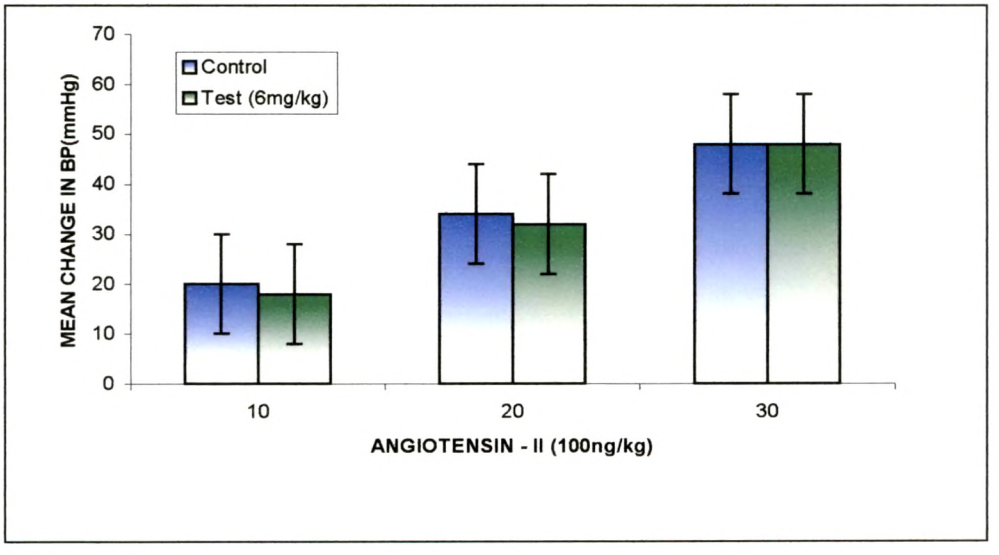


Figure 3C

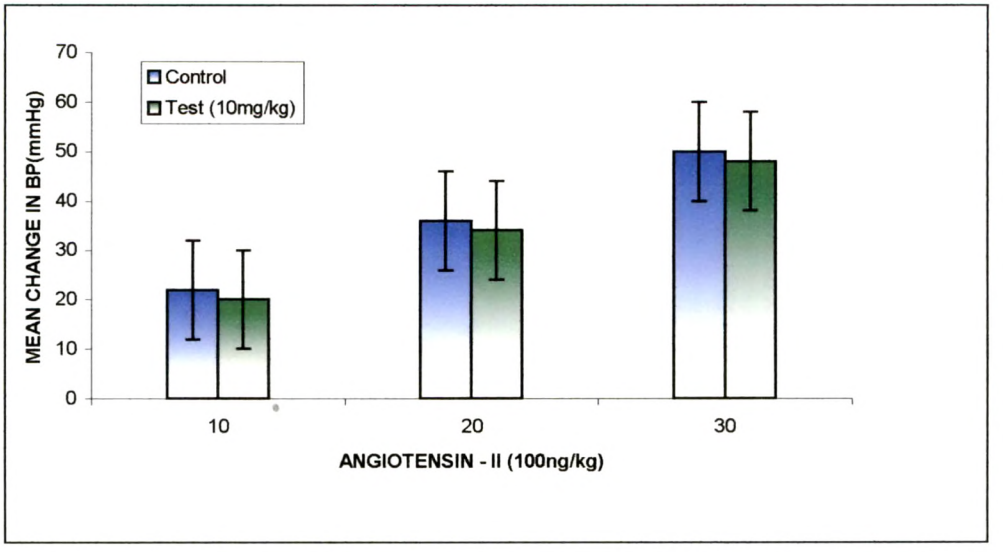
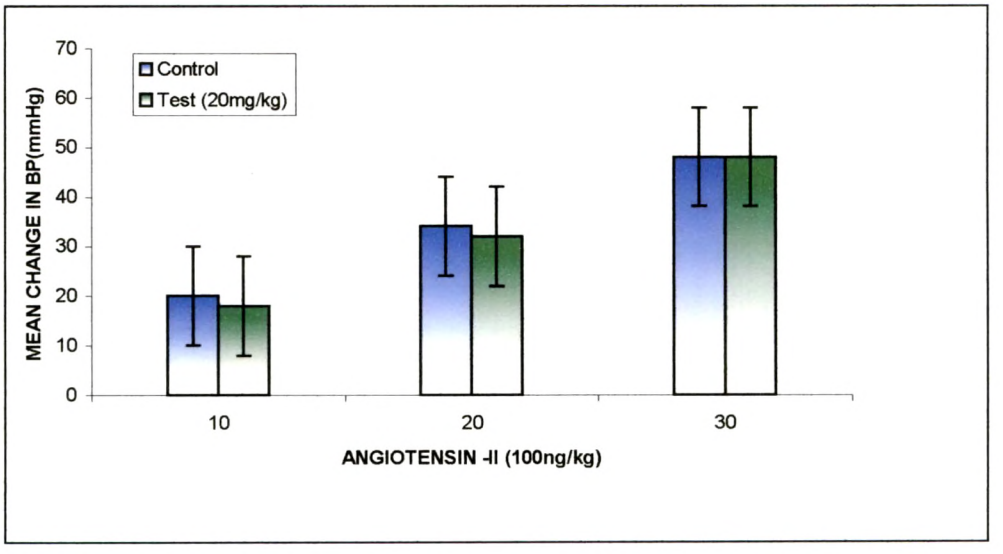


Figure 3D



B.1.1.1.4 Effect of Histamine Before And After Intravenous Administration of 24 on Rat Blood Pressure

Figure 4: Mean change in blood pressure by acute intravenous injection of histamine (0.1, 0.2, 0.3. 0.4 μg) in anesthetized rats before and after intravenous **24** (2, 6, 10 and 20 mg/kg). Abscissa indicates doses of histamine and ordinate indicates mean change in blood pressure (mm Hg). Vertical line indicates SEM (n = 5).

Figure 4A

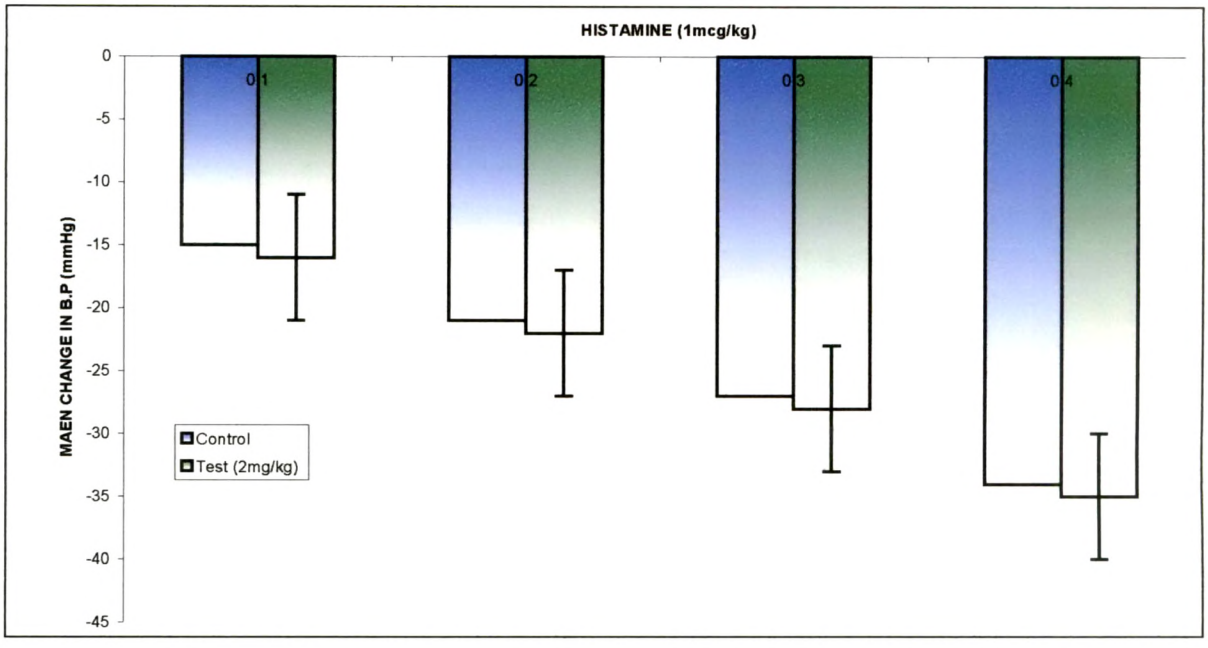


Figure 4B

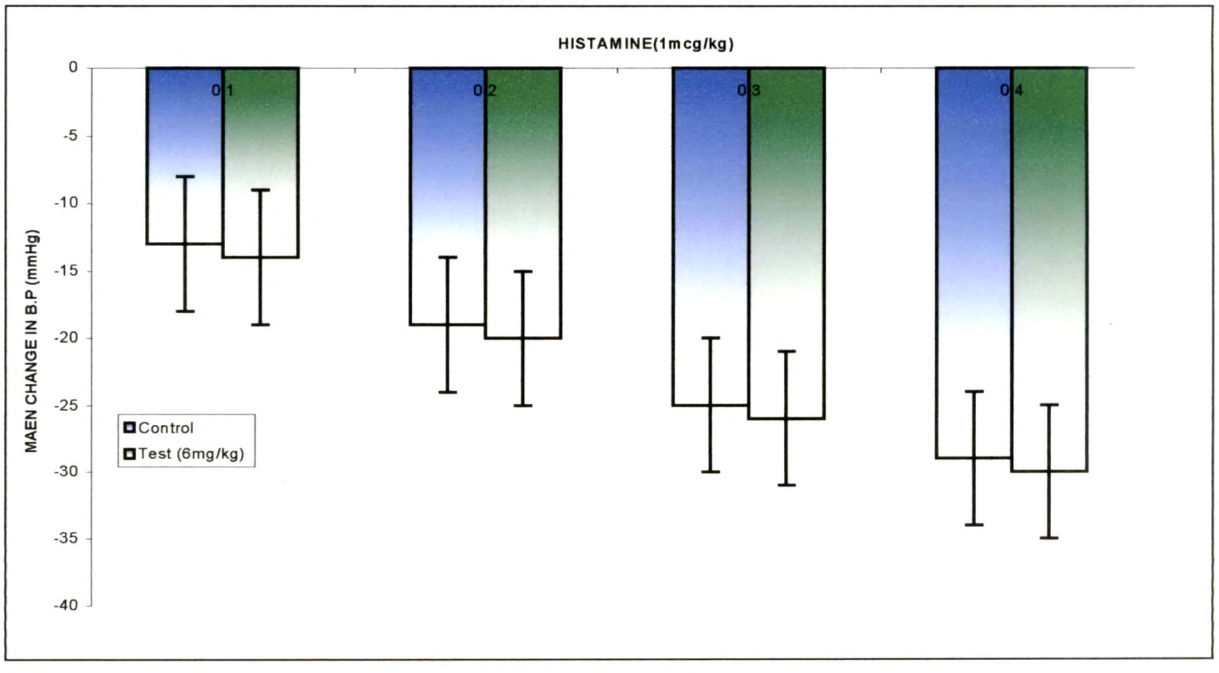


Figure 4A

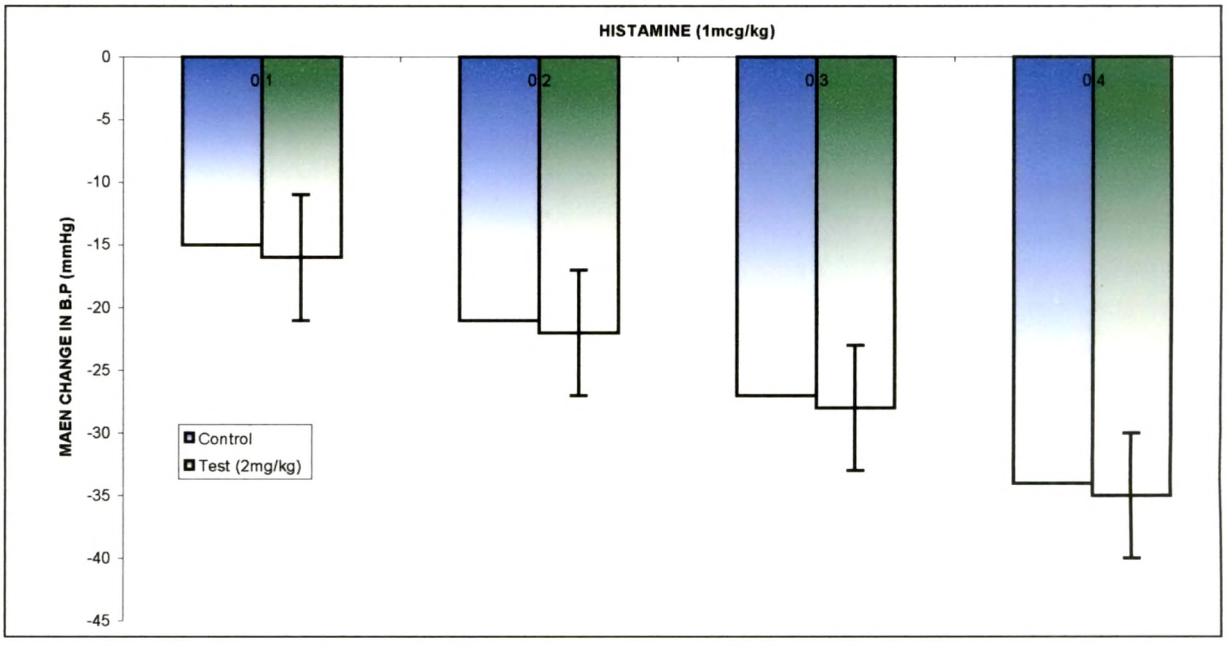
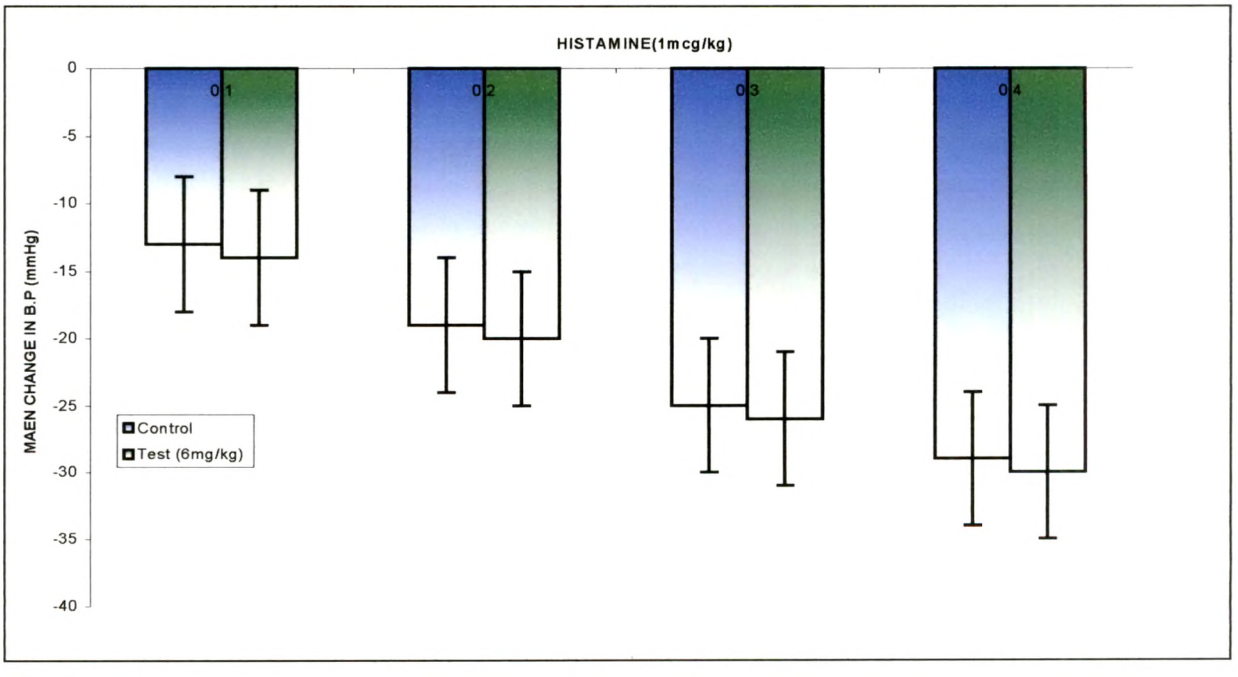


Figure 4B



B.1.1.1.5. Comparison of Mean Change In Rat Blood Pressure Before And After Intravenous Administration of 24 And DMSO.

Figure 5: Mean change is normal systolic blood pressure by acute intravenous injection of DMSO and 2, 6, 10 and 20 mg/kg of **24** in anesthetized rats. Vertical line on histograms represents SEM (n = 5).

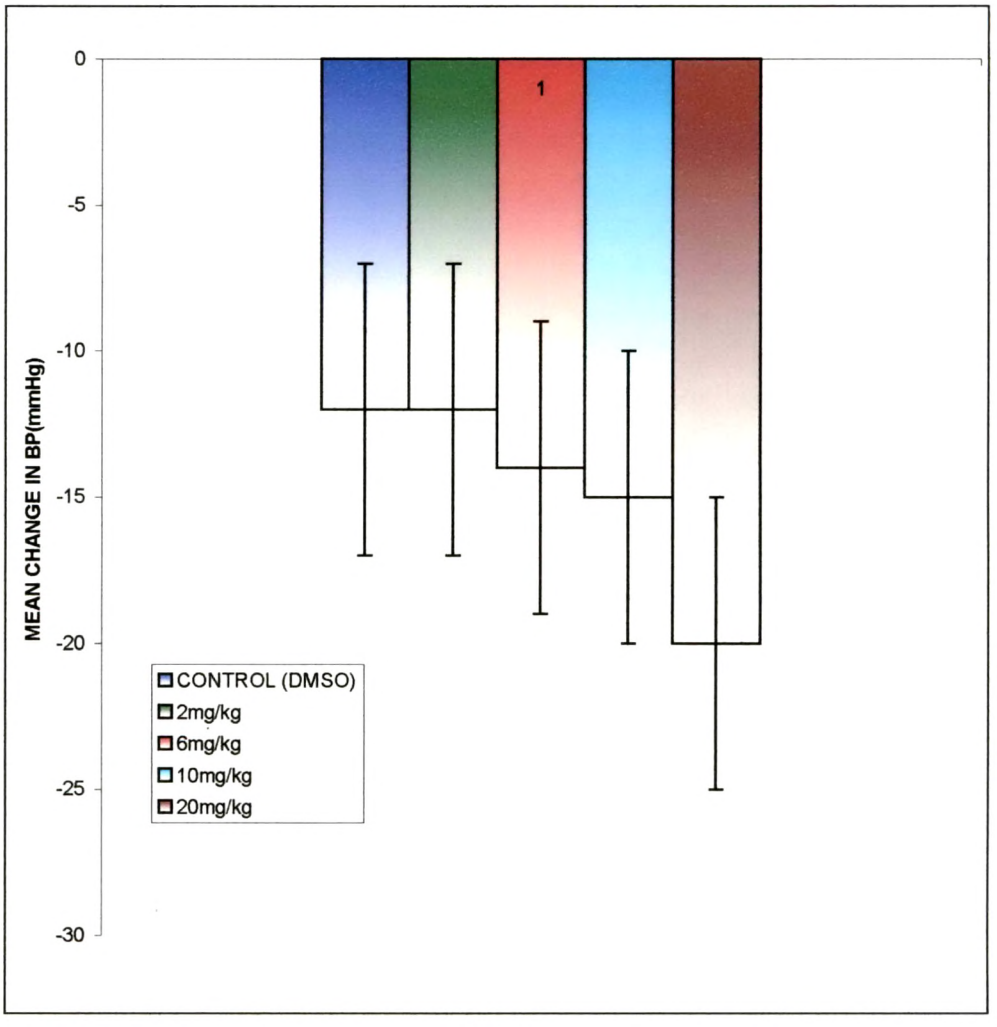
A vs B = $p > 0.05$ (NS)

A vs. C = $p > 0.05$(NS)

A vs. D = ** $p < 0.01$

A vs. E = ** $p < 0.01$ as compared with corresponding control

Figure 5


0
-5
-10
-15
-20
-25
-30
MEAN CHANGE IN BP(mmHg)
1
CONTROL (DMSO)
2mg/kg
6mg/kg
10mg/kg
20mg/kg

B.1.1.1.6 Adrenaline Reversal Responses of 24 After Intravenous Injection

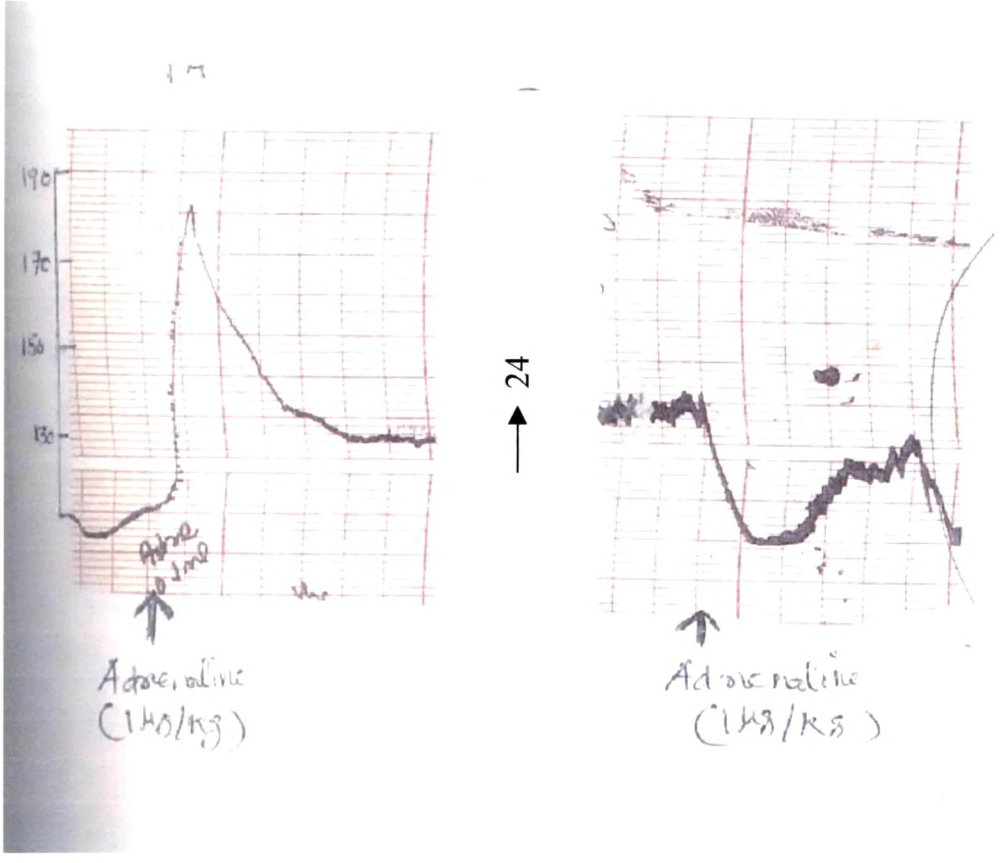
Figure 6: Tracing of adrenaline reversal responses of **24** of an anesthetized urethane (120 mg/kg, iv) rats. Abscissa indicates dose of adrenaline and ordinate the blood pressure (mm Hg)

A –Adrenaline (1 μg/kg) response (control)

B- Adrenaline (1 μg/kg) response in **24** treated animals.

A

B



B.1.1.2. Effect of Various Agonists Before And After The Intraperitoneal Administration Of Test Compounds on Rat Blood Pressure

The *in vivo* study indicates that acute intraperitoneal injection of **1** (5 mg/kg) showed nonsignificant change in pressor response to noradrenaline (NA 1 μg/kg, and 2 μg/kg, i.v.), adrenaline (Adr 0.5 μg/kg, and 1 μg/kg, i.v.), and histamine (His 1 μg/kg, and 2 μg/kg, i.v.) as compared to control (Figure 7).

Acute intraperitoneal administration of **26** (5 mg/kg) showed significant change in pressor response to noradrenaline (NA 1 μg/kg, and 2 μg/kg, i.v.), and adrenaline (Adr 0.5 μg/kg, and 1 μg/kg, i.v.), but nonsignificant change in pressor response to histamine (His 1 μg/kg, and 2 μg/kg, i.v.) as compared to control (Figure 8 A).

Acute intraperitoneal administration of **27** (5 mg/kg) showed significant change in pressor response to noradrenaline (NA 1 μg/kg, and 2 μg/kg, i.v.), and adrenaline (Adr 0.5 μg/kg, and 1 μg/kg, i.v.), it also showed significant change in pressor response to histamine (His 1 μg/kg, and 2 μg/kg, i.v.) as compared to control (Figure 9 A). Also tracing of blood pressure response showed the "adrenaline reversal" which indicates α-adrenoceptor blocking activity (Figure 9 B).

Acute intraperitoneal administration of **29** (5 mg/kg) showed insignificant change in pressor response to noradrenaline (NA 1 μg/kg, and 2

μg/kg, i.v.), adrenaline (Adr 0.5 μg/kg, and 1 μg/kg, i.v.), and histamine (His 1 μg/kg, and 2 μg/kg, i.v.) as compared to control (Figure 10).

Acute intraperitoneal administration of **6** (5 mg/kg) showed nonsignificant change in pressor response to noradrenaline (NA 1 μg/kg, and 2 μg/kg, i.v.), adrenaline (Adr 0.5 μg/kg, and 1 μg/kg, i.v.), and histamine (His 1 μg/kg, and 2 μg/kg, i.v.) as compared to control (Figure 11).

Acute intraperitoneal administration of **8** (5 mg/kg) showed significant change in pressor response to noradrenaline (NA 1 μg/kg, and 2 μg/kg, i.v.), and adrenaline (Adr 0.5 μg/kg, and 1 μg/kg, i.v.), but nonsignificant change in pressor response to histamine (His 1 μg/kg, and 2 μg/kg, i.v.) as compared to control (Figure 12 A).

Acute intraperitoneal administration of Prazosin (50 μg/kg) showed significant change in pressor response to noradrenaline (NA 1 μg/kg, and 2 μg/kg, i.v.), and adrenaline (Adr 0.5 μg/kg, and 1 μg/kg, i.v.), but nonsignificant change in pressor response to histamine (His 1 μg/kg, and 2 μg/kg, i.v.) as compared to control (Figure 13A).

B.1.1.2.1 Effect of various agonists before and after the intraperitoneal administration of 1 (5 mg/kg) on Rat Blood Pressure

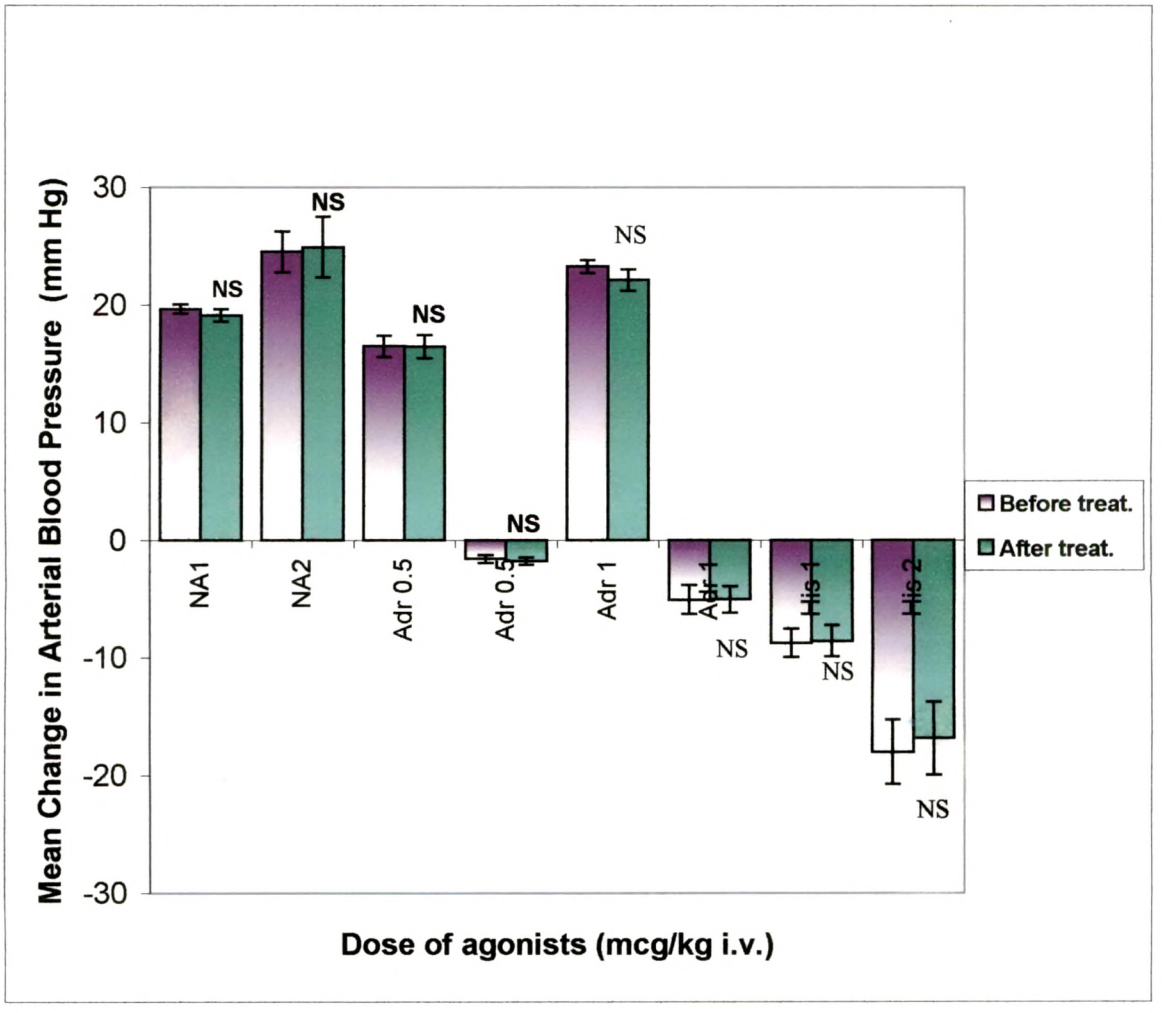
Figure 7: Mean change in arterial blood pressure produced by acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (Adr 0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats before and after intraperitoneal injection of **1** (5 mg/kg).

Abscissa indicates doses of agonists like noradrenaline, adrenaline, and histamine (μg/kg i.v.) and ordinate indicates mean change in blood pressure (mm Hg).

Vertical line on bars represents SEM (n = 6).

NS = nonsignificant

Figure 7


Mean Change in Arterial Blood Pressure (mm Hg)
30
20
10
0
-10
-20
-30
NS
NS
NS
NS
NS
NS
NS
NA1
NA2
Adr 0.5
Adr 0.5
Adr 1
Adr 1
His 1
His 2
Before treat.
After treat.
Dose of agonists (mcg/kg i.v.)

B.1.1.2.2 Effect of various agonists before and after the intraperitoneal administration of 26 (5 mg/kg) on Rat Blood Pressure

Figure 8 A: Mean change in arterial blood pressure (mm Hg) produced by acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (Adr 0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats before and after intraperitoneal injection of **26** (5 mg/kg).

Abscissa indicates doses of agonists like noradrenaline, adrenaline, and histamine (μg/kg i.v.) and ordinate indicates mean change in blood pressure (mm Hg).

Vertical line on bars represents SEM (n = 6).

NS = insignificant

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

Figure 8 A

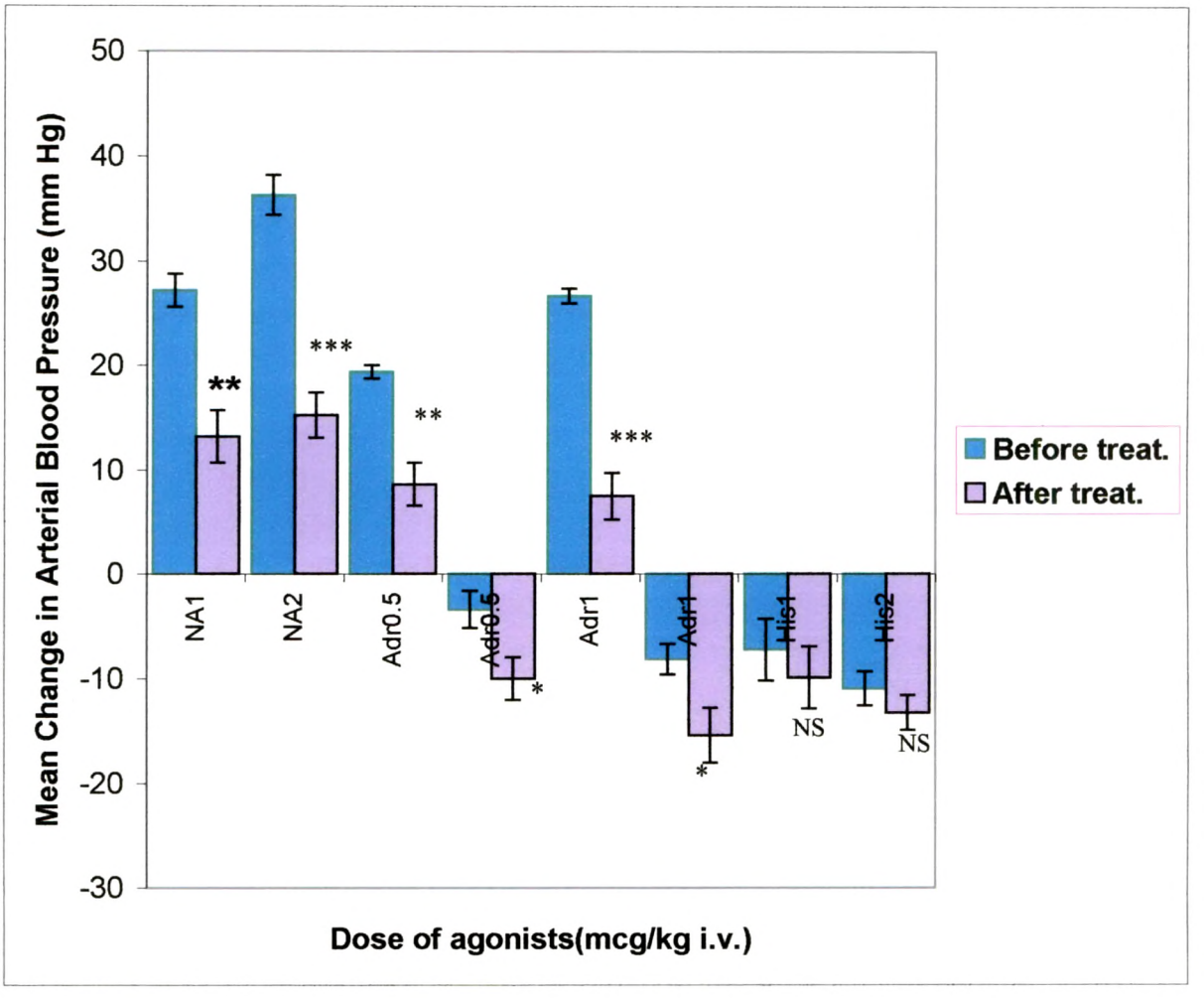
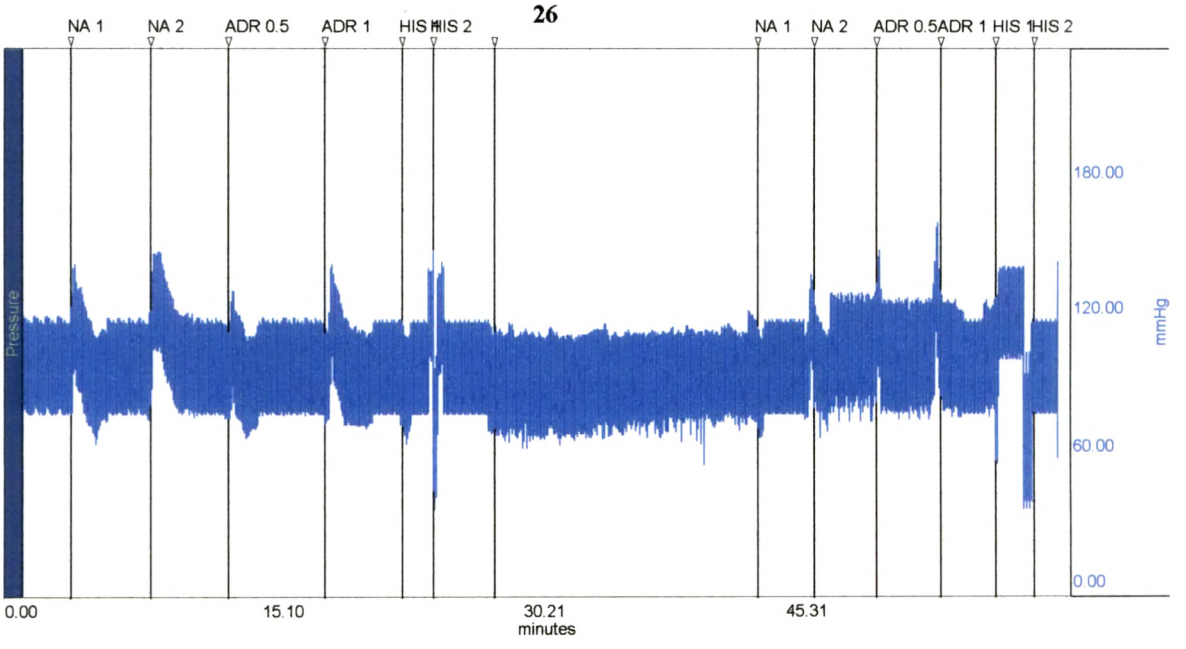

Mean Change in Arterial Blood Pressure (mm Hg)
50
40
30
20
10
0
-10
-20
-30
**
***
**
*
***
*
NS
NS
NA1
NA2
Adr0.5
Adr0.5
Adr1
Adr1
His1
His2
Dose of agonists(mcg/kg i.v.)
Before treat.
After treat.

Figure 8 B: Tracing of blood pressure response after acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats before and after intraperitoneal injection of **26** (5 mg/kg).

Abscissa indicates time in minutes and ordinate indicates change in blood pressure (mm Hg).

Figure 8 B


26
NA 1
NA 2
ADR 0.5
ADR 1
HIS 1
HIS 2
NA 1
NA 2
ADR 0.5
ADR 1
HIS 1
HIS 2
Pressure
180.00
120.00
60.00
0.00
mmHg
0.00
15.10
30.21
45.31
minutes

B.1.1.2.3 Effect of various agonists before and after the intraperitoneal administration of 27 (5 mg/kg) on Rat Blood Pressure

Figure 9 A: Mean change in arterial blood pressure (mm Hg) produced by acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (Adr 0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats before and after intraperitoneal injection of **27** (5 mg/kg).

Abscissa indicates doses of agonists like noradrenaline, adrenaline, and histamine (μg/kg i.v.) and ordinate indicates mean change in blood pressure (mm Hg).

Vertical line on bars represents SEM (n = 6).

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

Figure 9A

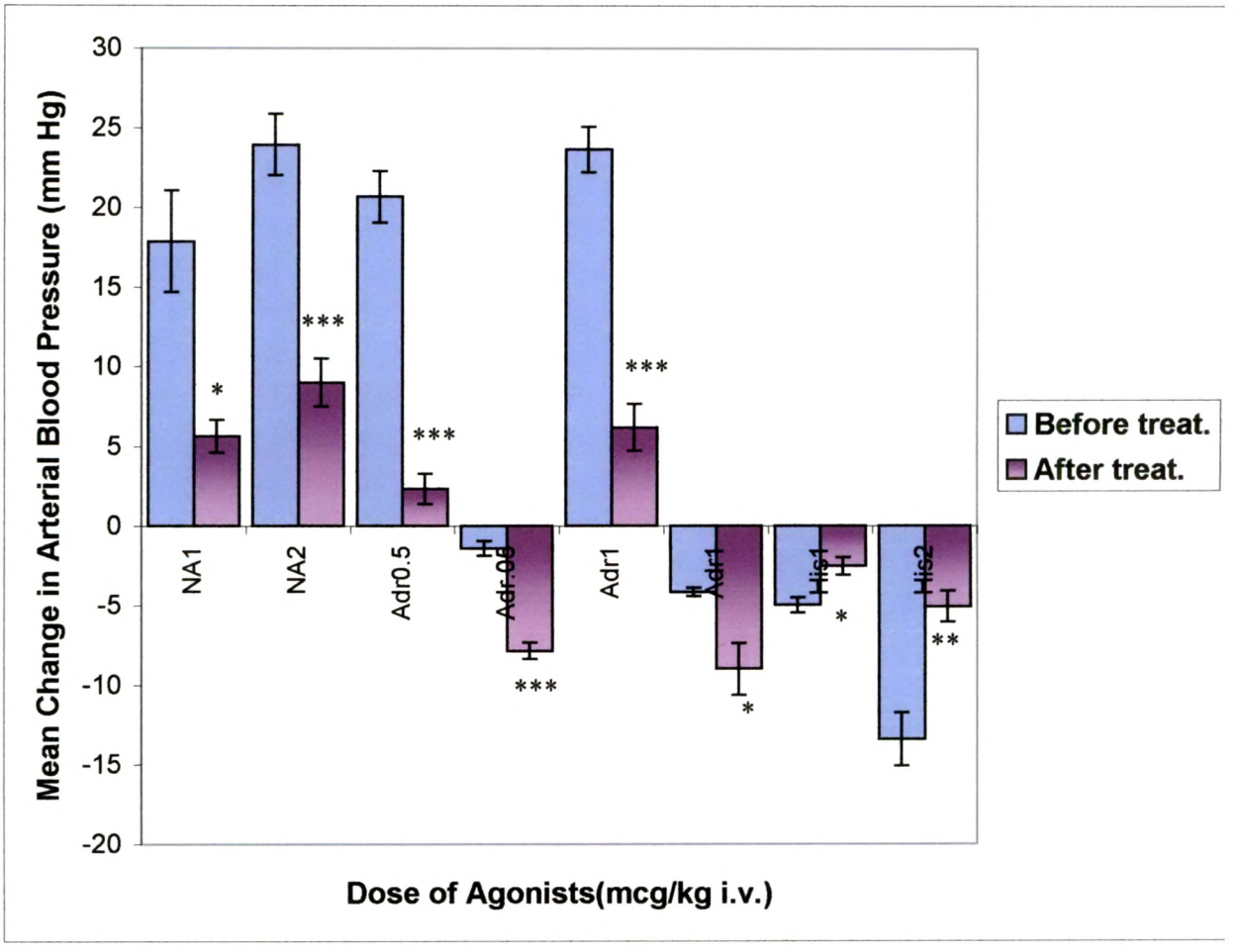
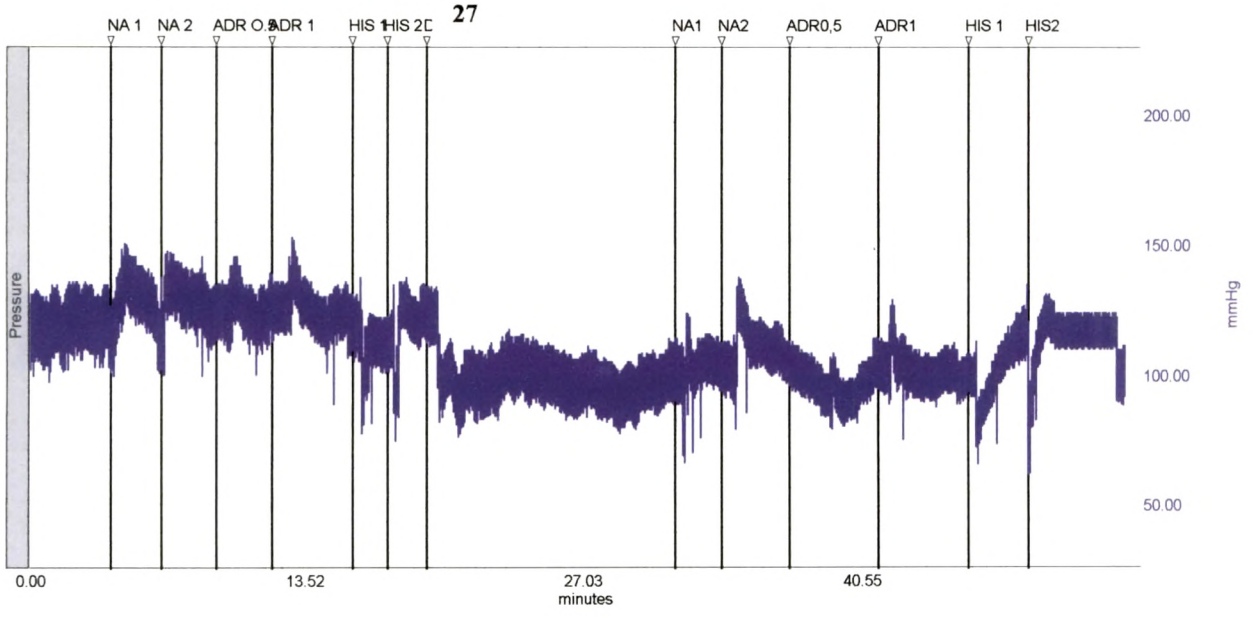

Mean Change in Arterial Blood Pressure (mm Hg)
30
25
20
15
10
5
0
-5
-10
-15
-20
NA1
NA2
Adr0.5
Adr1
Adr1
His1
His2
*
***
***
***
*
*
**
Before treat.
After treat.
Dose of Agonists(mcg/kg i.v.)

Figure 9 B: Tracing of blood pressure response after acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats before and after intraperitoneal injection of **27** (5 mg/kg).

Abscissa indicates time in minutes and ordinate indicates change in blood pressure (mm Hg).

Tracing of blood pressure response (Figure 9 B) clearly showed the *adrenaline reversal* response (Dale's vasomotor reversal) after i.p. treatment of **27** which indicates the α-adrenoceptor blocking activity of the test compound.

Figure 9B


27
NA 1
NA 2
ADR 0,5
ADR 1
HIS 1
HIS 2
NA1
NA2
ADR0,5
ADR1
HIS 1
HIS2
Pressure
200.00
150.00
100.00
50.00
mmHg
0.00
13.52
27.03
40.55
minutes

B.1.1.2.4 Effect of various agonists before and after the intraperitoneal administration of 29 (5 mg/kg) on Rat Blood Pressure

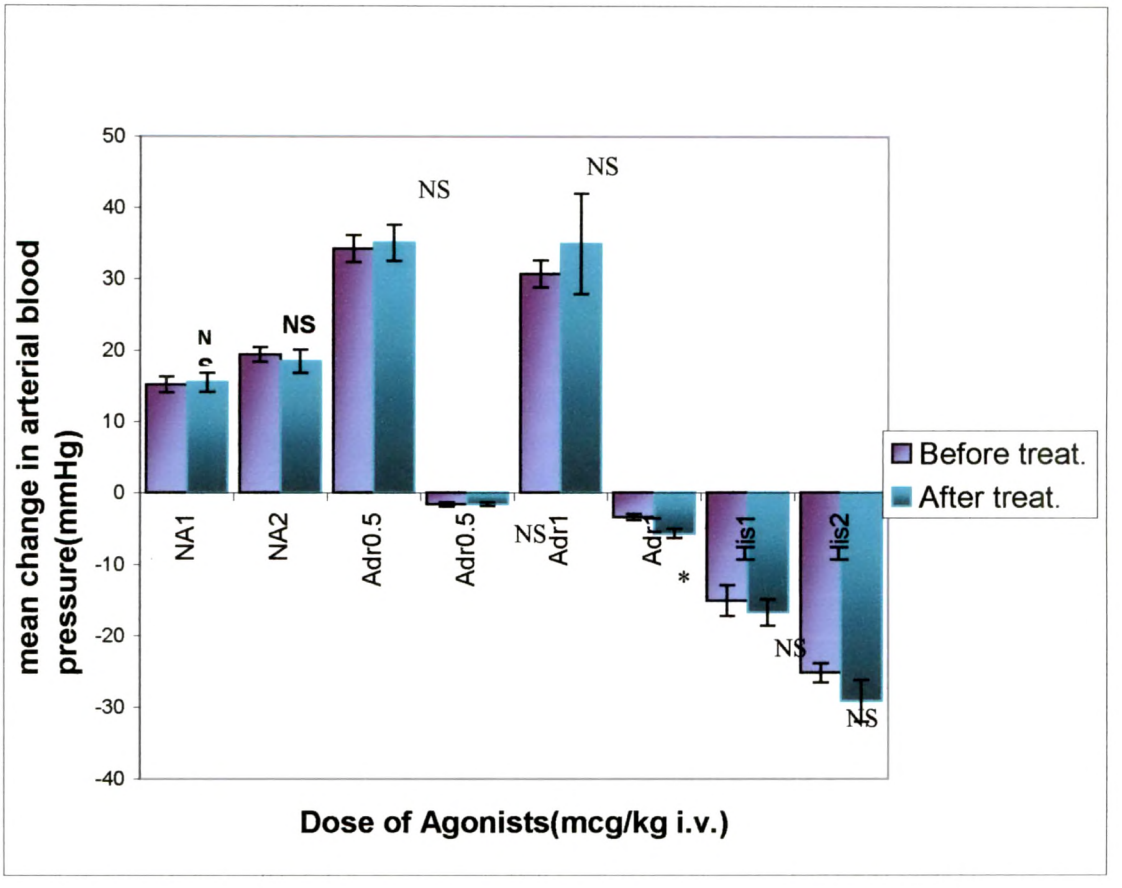
Figure 10: Mean change in arterial blood pressure (mm Hg) produced by acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (Adr 0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats before and after intraperitoneal injection of **29** (5 mg/kg).

Abscissa indicates doses of agonists like noradrenaline, adrenaline, and histamine (μg/kg i.v.) and ordinate indicates mean change in blood pressure (mm Hg).

Vertical line on bars represents SEM (n = 6).

NS = insignificant

Figure 10


mean change in arterial blood pressure(mmHg)
50
40
30
20
10
0
-10
-20
-30
-40
NS
NS
NS
NS
NS
*
NS
NS
NA1
NA2
Adr0.5
Adr0.5
NS Adr1
His1
His2
Before treat.
After treat.
Dose of Agonists(mcg/kg i.v.)

B.1.1.2.5. Effect of various agonists before and after the intraperitoneal administration of 6 (5 mg/kg) on Rat Blood Pressure

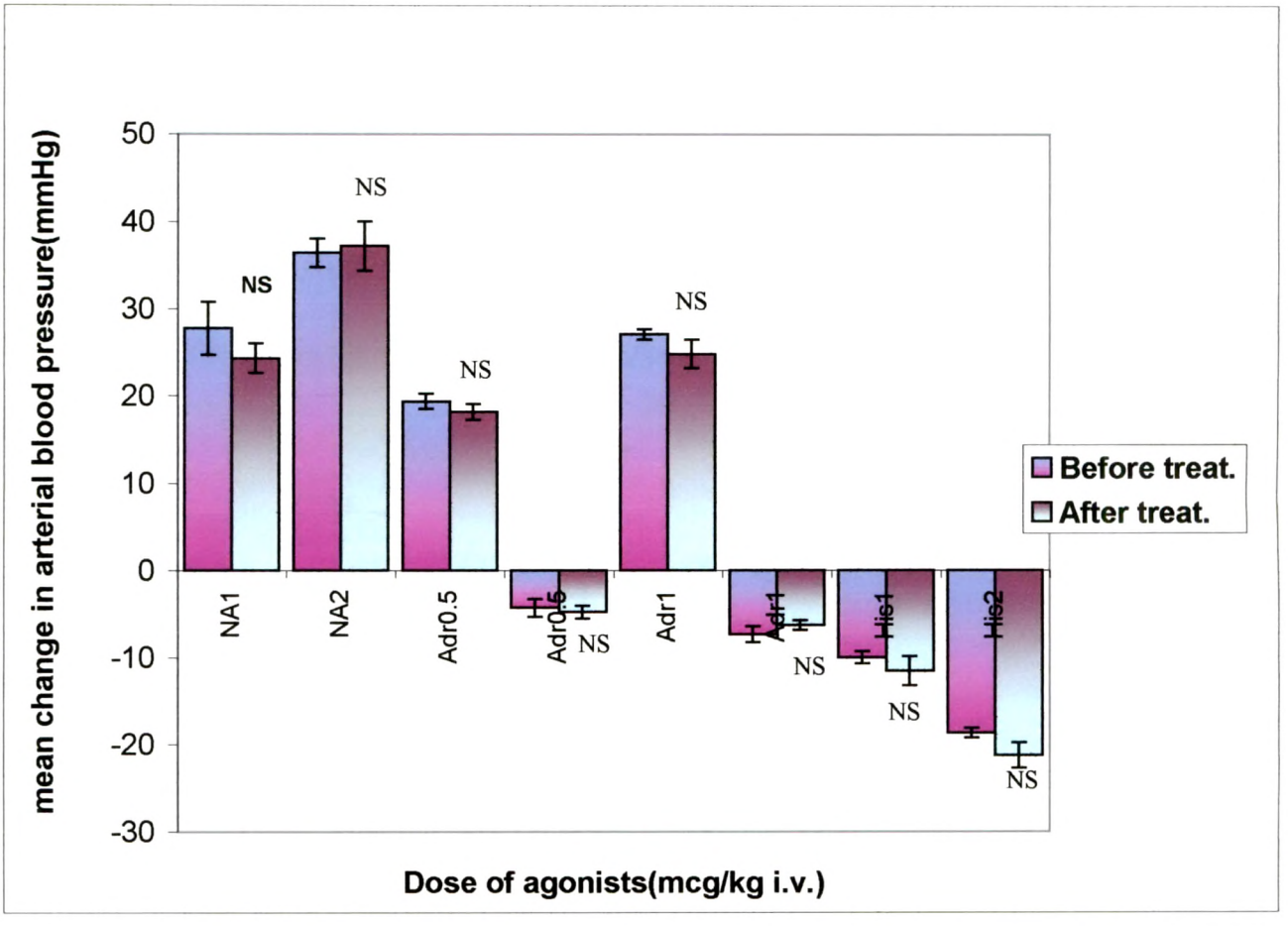
Figure 11: Mean change in arterial blood pressure (mm Hg) produced by acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (Adr 0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats before and after intraperitoneal injection of **6** (5 mg/kg).

Abscissa indicates doses of agonists like noradrenaline, adrenaline, and histamine (μg/kg i.v.) and ordinate indicates mean change in blood pressure (mm Hg).

Vertical line on bars represents SEM (n = 6).

NS = insignificant

Figure 11


mean change in arterial blood pressure(mmHg)
50
40
30
20
10
0
-10
-20
-30
NS
NS
NS
NS
NS
NS
NS
NS
NA1
NA2
Adr0.5
Adr0.5
Adr1
Adr1
His1
His2
Before treat.
After treat.
Dose of agonists(mcg/kg i.v.)

B.1.1.2.6 Effect of various agonists before and after the intraperitoneal administration of 8 (5 mg/kg) on Rat Blood Pressure

Figure 12 A: Mean change in arterial blood pressure (mm Hg) produced by acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (Adr 0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats before and after intraperitoneal injection of **8** (5 mg/kg).

Abscissa indicates doses of agonists like noradrenaline, adrenaline, and histamine (μg/kg i.v.) and ordinate indicates mean change in blood pressure (mm Hg).

Vertical line on bars represents SEM (n = 6).

NS = insignificant

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

Figure 12A

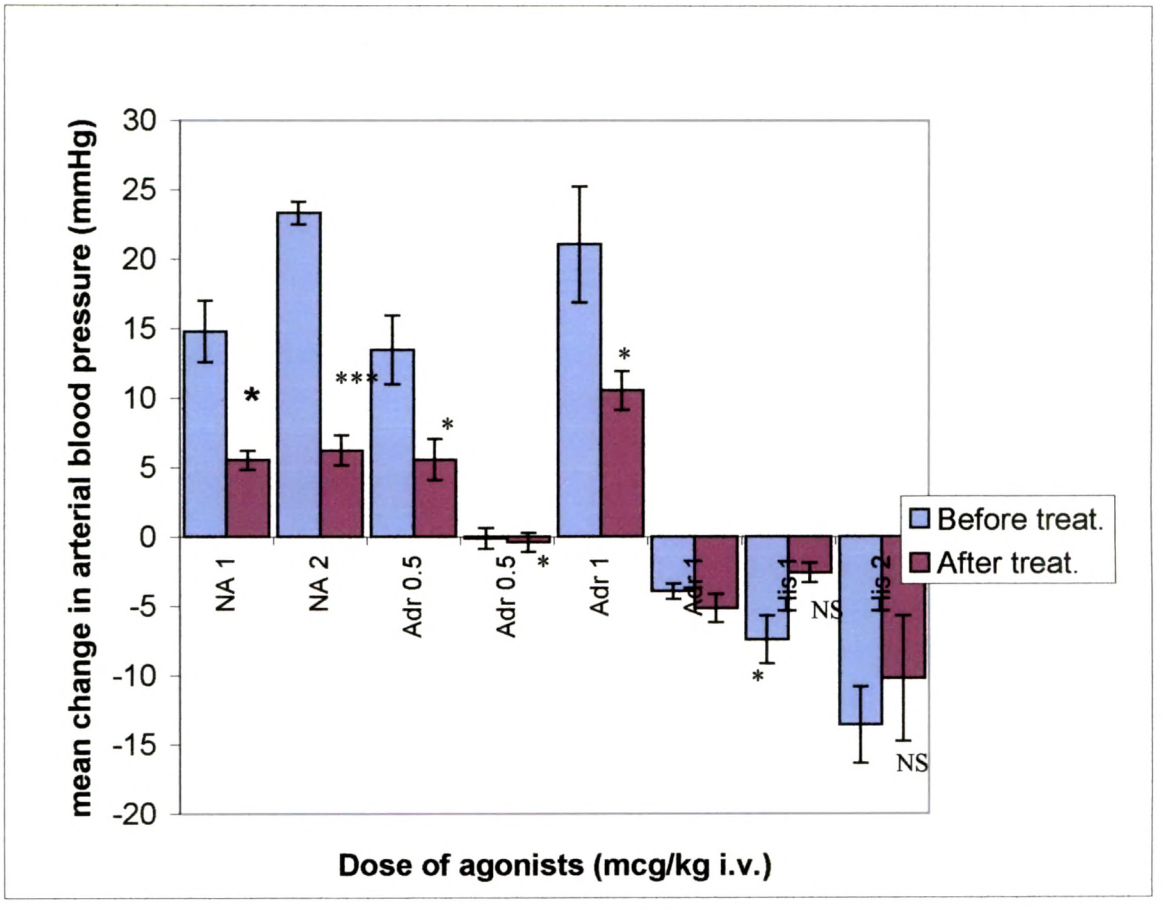
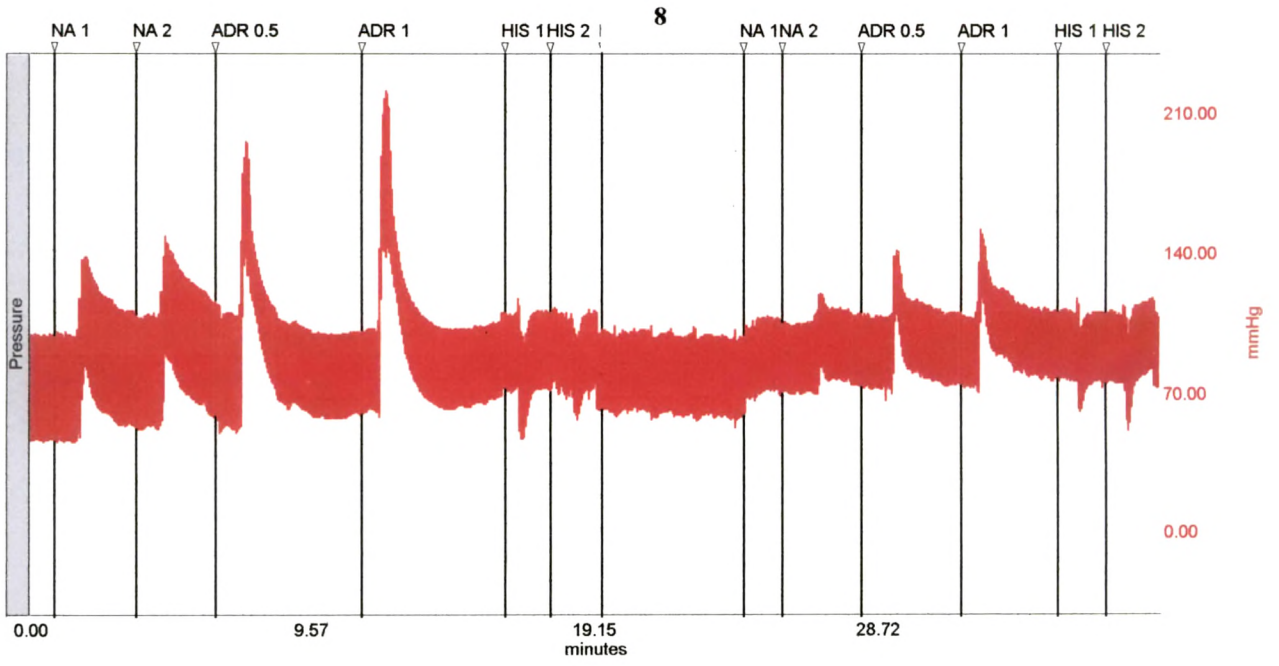

mean change in arterial blood pressure (mmHg)
30
25
20
15
10
5
0
-5
-10
-15
-20
NA 1
NA 2
Adr 0.5
Adr 0.5
Adr 1
Adr 1
His 1
His 2
*
***
*
*
*
*
NS
NS
Before treat.
After treat.
Dose of agonists (mcg/kg i.v.)

Figure 12 B: Tracing of blood pressure response after acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats before and after intraperitoneal injection of **8** (5 mg/kg).

Abscissa indicates time in minutes and ordinate indicates change in blood pressure (mm Hg).

Figure 12B


8
NA 1
NA 2
ADR 0.5
ADR 1
HIS 1
HIS 2
NA 1
NA 2
ADR 0.5
ADR 1
HIS 1
HIS 2
Pressure
210.00
140.00
70.00
0.00
mmHg
0.00
9.57
19.15
28.72
minutes

B.1.1.2.7 Effect of various agonists before and after the intraperitoneal administration of Prazosin (50 μg/kg) on Rat Blood Pressure

Figure 13 A: Mean change in arterial blood pressure (mm Hg) produced by acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (Adr 0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats before and after intraperitoneal injection of Prazosin (50 μg/kg, i.p.).

Abscissa indicates doses of agonists like noradrenaline, adrenaline, and histamine (μg/kg i.v.) and ordinate indicates mean change in blood pressure (mm Hg).

Vertical line on bars represents SEM (n = 6).

NS = insignificant

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

Figure 13A

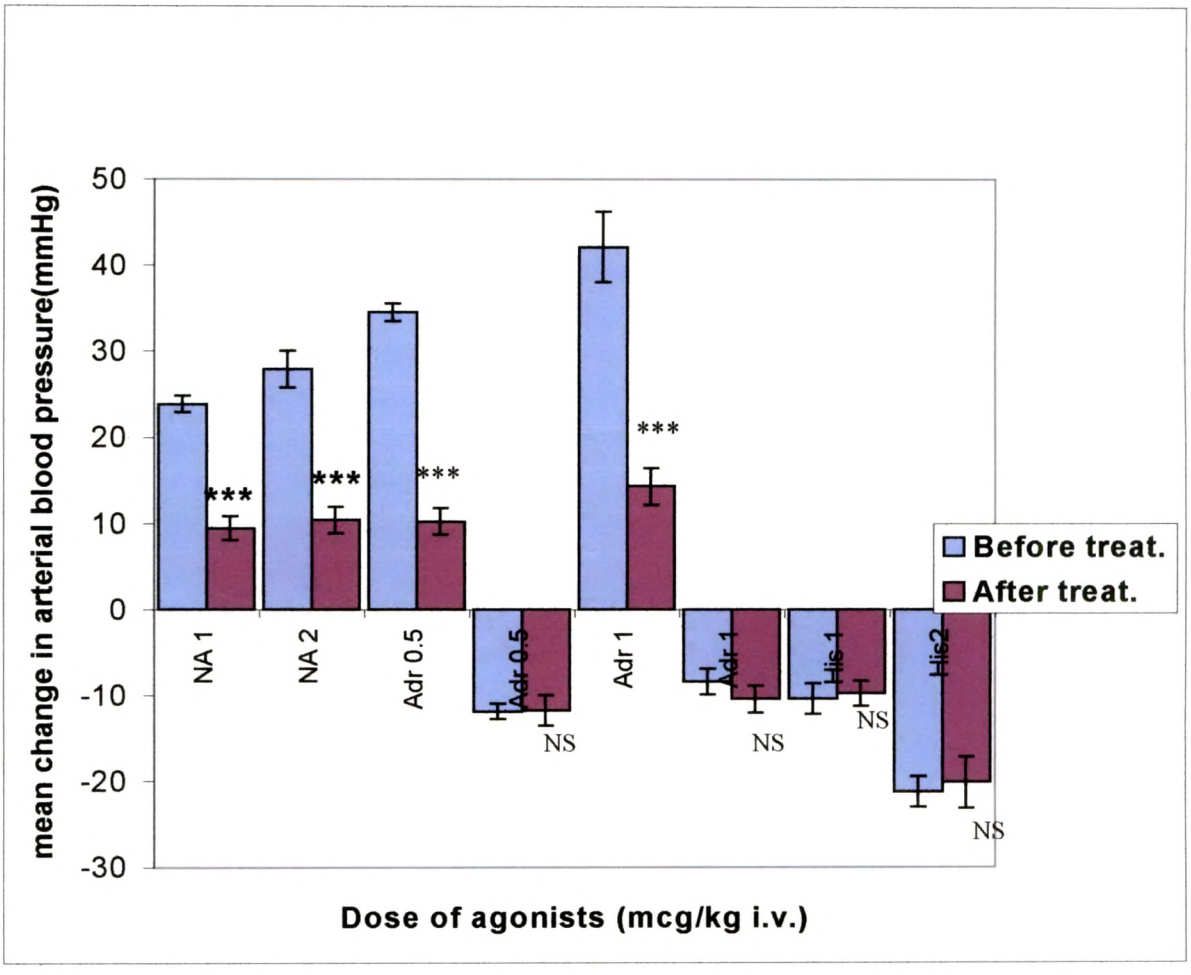
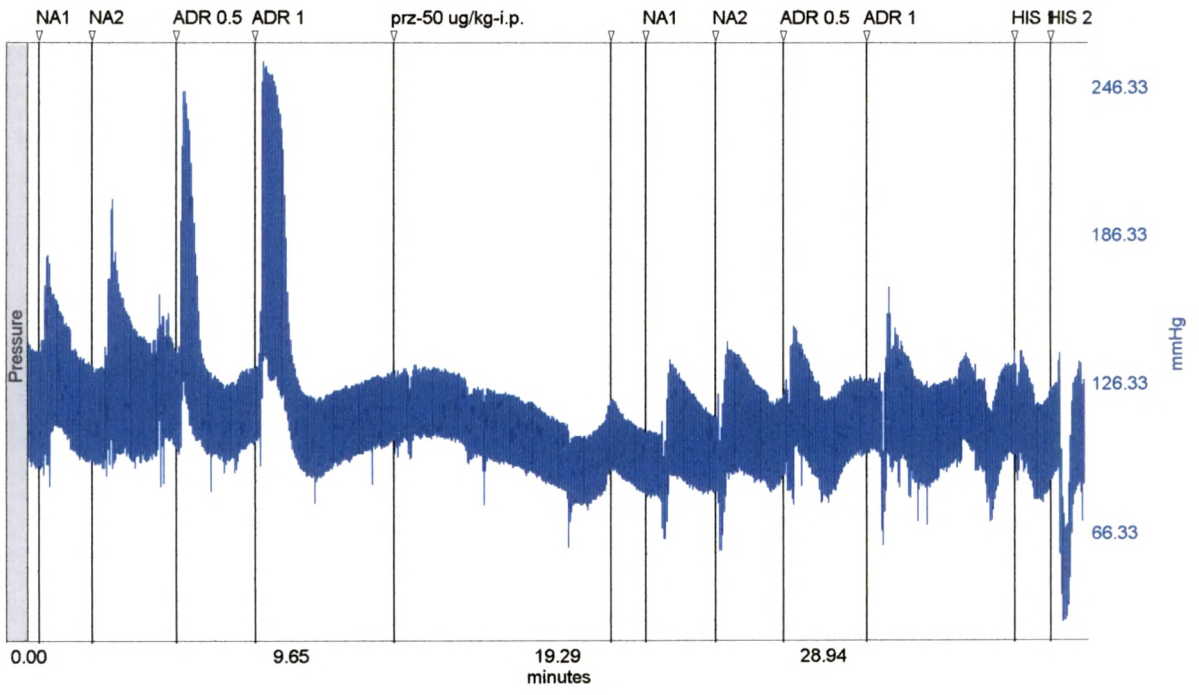

mean change in arterial blood pressure(mmHg)
50
40
30
20
10
0
-10
-20
-30
***
***
***
***
NS
NS
NS
NS
NA 1
NA 2
Adr 0.5
Adr 0.5
Adr 1
Adr 1
His 1
His2
Before treat.
After treat.
Dose of agonists (mcg/kg i.v.)

Figure 13 B: Tracing of blood pressure response after acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (Adr 0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats before and after intraperitoneal injection of Prazosin (50 μg/kg,).

Abscissa indicates time in minutes and ordinate indicates change in blood pressure (mm Hg).

Figure 13B


NA1
NA2
ADR 0.5
ADR 1
prz-50 ug/kg-i.p.
NA1
NA2
ADR 0.5
ADR 1
HIS 1
HIS 2
Pressure
246.33
186.33
126.33
66.33
mmHg
0.00
9.65
19.29
28.94
minutes

B.1.1.3 Effect of Various Agonists Before And After The Oral Administration of Test Compounds on Rat Blood Pressure

Acute oral administration of **1** (10 mg/kg) showed significant change in pressor response to noradrenaline (NA 1 μg/kg, and 2 μg/kg, i.v.), and adrenaline (Adr 0.5 μg/kg, and 1 μg/kg, i.v.), but insignificant change in pressor response to histamine (His 1 μg/kg, and 2 μg/kg, i.v.) as compared to control (Figure 14 A). Also tracing of blood pressure response showed the "adrenaline reversal" which indicates α-adrenoceptor blocking activity (Figure 14 B).

Acute oral administration of **26** (10 mg/kg) showed significant change in pressor response to noradrenaline (NA 1 μg/kg, and 2 μg/kg, i.v.), and adrenaline (Adr 0.5 μg/kg, and 1 μg/kg, i.v.), but nonsignificant change in pressor response to histamine (His 1 μg/kg, and 2 μg/kg, i.v.) as compared to control (Figure 15 A). Also tracing of blood pressure response showed the "adrenaline reversal" indicating α-adrenoceptor blocking activity (Figure 15 B).

Acute oral administration of **27** (10 mg/kg) showed significant change in pressor response to noradrenaline (NA 1 μg/kg, and 2 μg/kg, i.v.), and adrenaline (Adr 0.5 μg/kg, and 1 μg/kg, i.v.), it also showed significant change in pressor response to histamine (His 1 μg/kg, and 2 μg/kg, i.v.) as compared to control (Figure 16 A). Also tracing of blood pressure response

showed the "adrenaline reversal" which indicates α-adrenoceptor blocking activity (Figure 16 B).

Acute oral administration of **8** (10 mg/kg) showed significant change in pressor response to noradrenaline (NA 1 μg/kg, and 2 μg/kg, i.v.), and adrenaline (Adr 0.5 μg/kg, and 1 μg/kg, i.v.), but nonsignificant change in pressor response to histamine (His 1 μg/kg, and 2 μg/kg, i.v.) as compared to control (Figure 17 A). Also tracing of blood pressure response showed the "adrenaline reversal" indicating α-adrenoceptor blocking activity (Figure 17 B).

Acute oral administration of Prazosin (500 μg/kg) showed significant change in pressor response to noradrenaline (NA 1 μg/kg, and 2 μg/kg, i.v.), and adrenaline (Adr 0.5 μg/kg, and 1 μg/kg, i.v.), but nonsignificant change in pressor response to histamine (His 1 μg/kg, and 2 μg/kg, i.v.) as compared to control (Figure 24 A).

Acute oral administration of **1**, **26**, **27**, and **8** (10 mg/kg) showed significant change in pressor response to phenylephrine (2.5, 5, 10, 20, 25, 50, and 100 μg/kg, i.v.) as compared to control (Figure 25).

B.1.1.3.1 Effect of various agonists after the oral administration of 1 (10 mg/kg) on Rat Blood Pressure

Figure 14 A: Mean change in arterial blood pressure (mm Hg) produced by acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (Adr 0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats after oral injection of **1** (10 mg/kg).

Abscissa indicates doses of agonists like noradrenaline, adrenaline, and histamine (μg/kg i.v.) and ordinate indicates mean change in arterial blood pressure (mm Hg).

Vertical line on bars represents SEM (n = 6).

NS = insignificant

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

Figure 14A

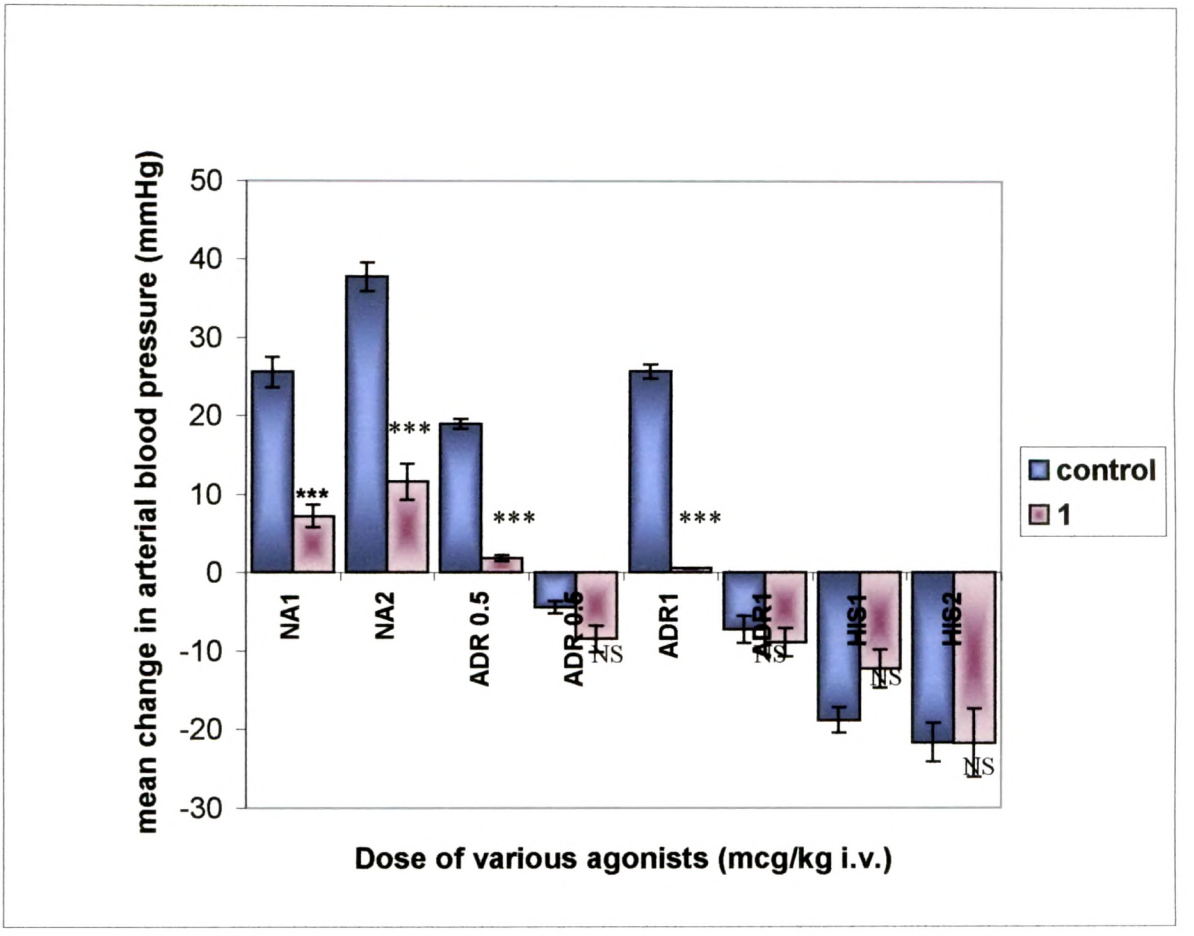
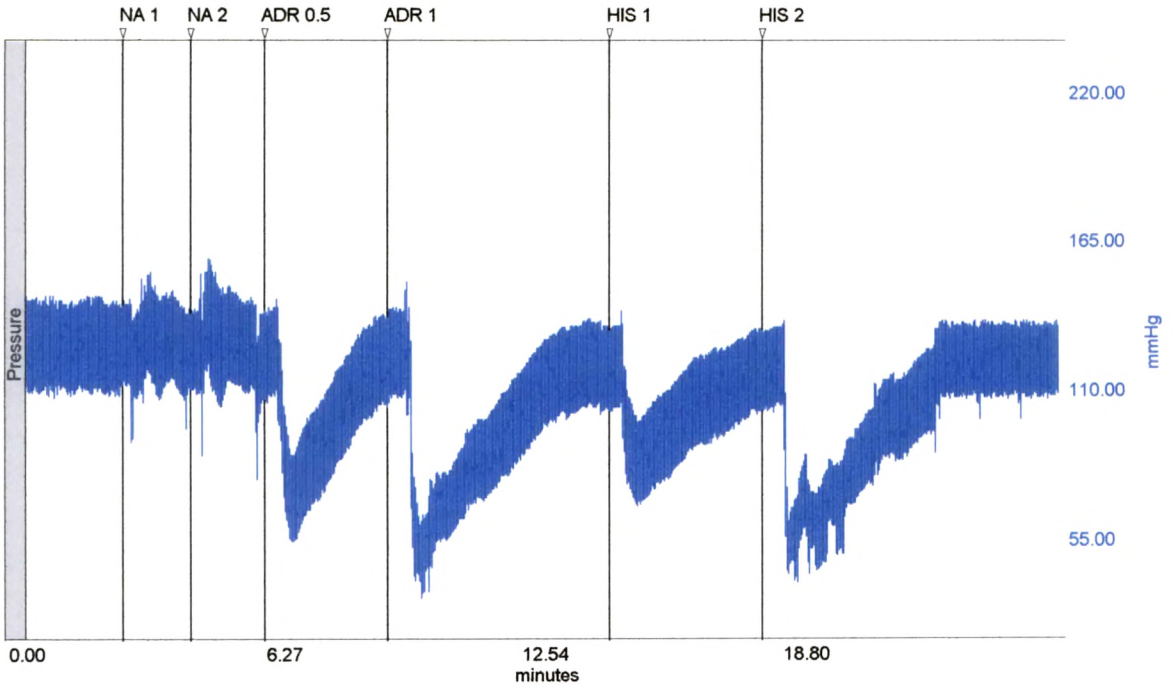

mean change in arterial blood pressure (mmHg)
50
40
30
20
10
0
-10
-20
-30
***
***
***
***
NS
NS
NS
NS
NA1
NA2
ADR 0.5
ADR 0.5
ADR1
ADR1
HIS1
HIS2
control
1
Dose of various agonists (mcg/kg i.v.)

Figure 14 B: Tracing of blood pressure response after acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats after oral administration of **1** (10 mg/kg).

Abscissa indicates time in minutes and ordinate indicates change in blood pressure (mm Hg).

Tracing of blood pressure response (Figure 14 B) clearly showed the *adrenaline reversal* response (Dale's vasomotor reversal) after oral treatment of **1** which indicates the α-adrenoceptor blocking activity of test compound.

Figure 14B


NA 1
NA 2
ADR 0.5
ADR 1
HIS 1
HIS 2
Pressure
220.00
165.00
110.00
55.00
mmHg
0.00
6.27
12.54
minutes
18.80

B.1.1.3.2 Effect of various agonists after the oral administration of 26 (10 mg/kg) on Rat Blood Pressure

Figure 15 A: Mean change in arterial blood pressure (mm Hg) produced by acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (Adr 0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats after oral administration of **26** (10 mg/kg).

Abscissa indicates doses of agonists like noradrenaline, adrenaline, and histamine (μg/kg i.v.) and ordinate indicates mean change in arterial blood pressure (mm Hg).

Vertical line on bars represents SEM (n = 6).

NS = insignificant

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

Figure 15A

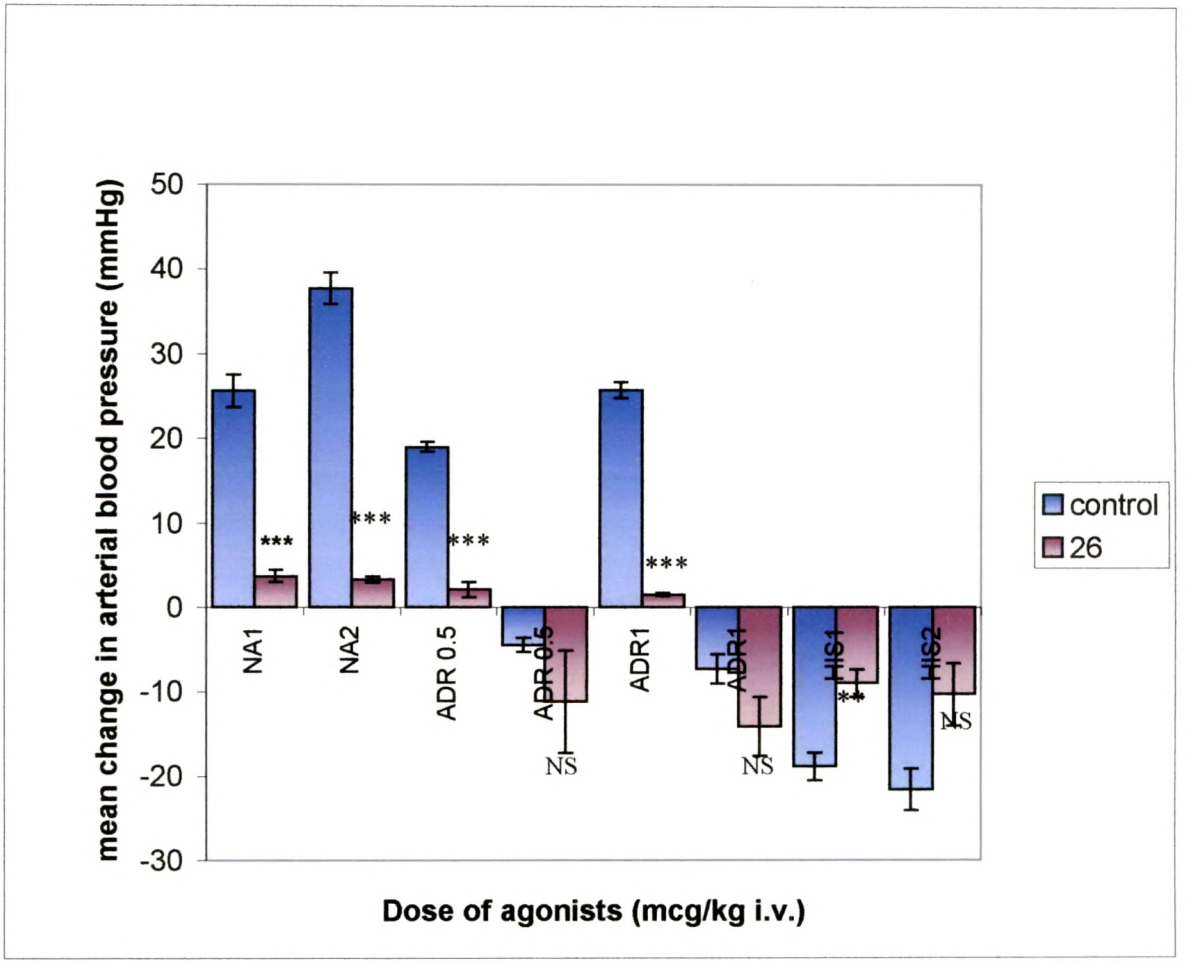
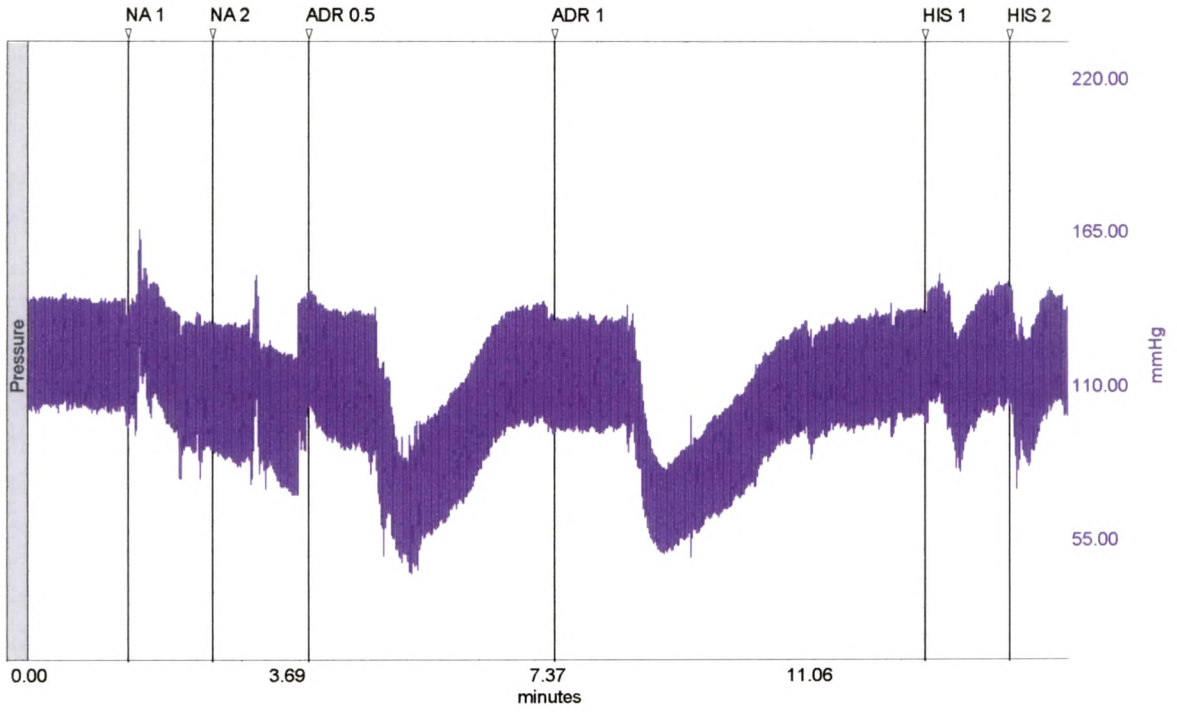

mean change in arterial blood pressure (mmHg)
50
40
30
20
10
0
-10
-20
-30
***
***
***
***
NS
NS
**
NS
NA1
NA2
ADR 0.5
ADR 0.5
ADR1
ADR1
HIS1
HIS2
control
26
Dose of agonists (mcg/kg i.v.)

Figure 15 B: Tracing of blood pressure response after acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats after oral administration of **26** (10 mg/kg).

Abscissa indicates time in minutes and ordinate indicates change in blood pressure (mm Hg).

Tracing of blood pressure response (Figure 15 B) clearly showed the *adrenaline reversal* response (Dale's vasomotor reversal) after oral treatment of **26** (10 mg/kg) indicating the α-adrenoceptor blocking activity of test compound.

Figure 15B


NA 1
NA 2
ADR 0.5
ADR 1
HIS 1
HIS 2
220.00
165.00
110.00
55.00
mmHg
Pressure
0.00
3.69
7.37
11.06
minutes

B.1.1.3.3 Effect of various agonists after the oral administration of 27 (10 mg/kg) on Rat Blood Pressure

Figure 16 A: Mean change in arterial blood pressure (mm Hg) produced by acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (Adr 0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats after oral administration of **27** (10 mg/kg).

Abscissa indicates doses of agonists like noradrenaline, adrenaline, and histamine (μg/kg i.v.) and ordinate indicates mean change in arterial blood pressure (mm Hg).

Vertical line on bars represents SEM (n = 6).

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

Figure 16A

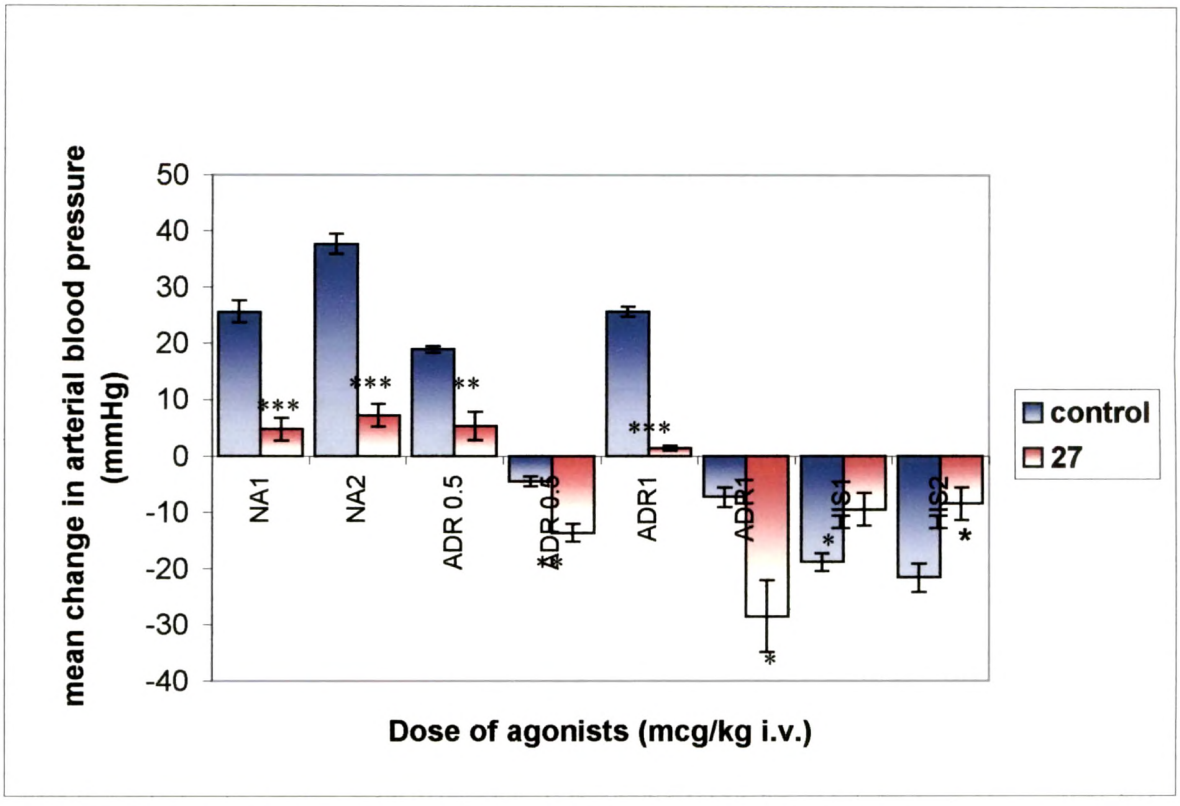
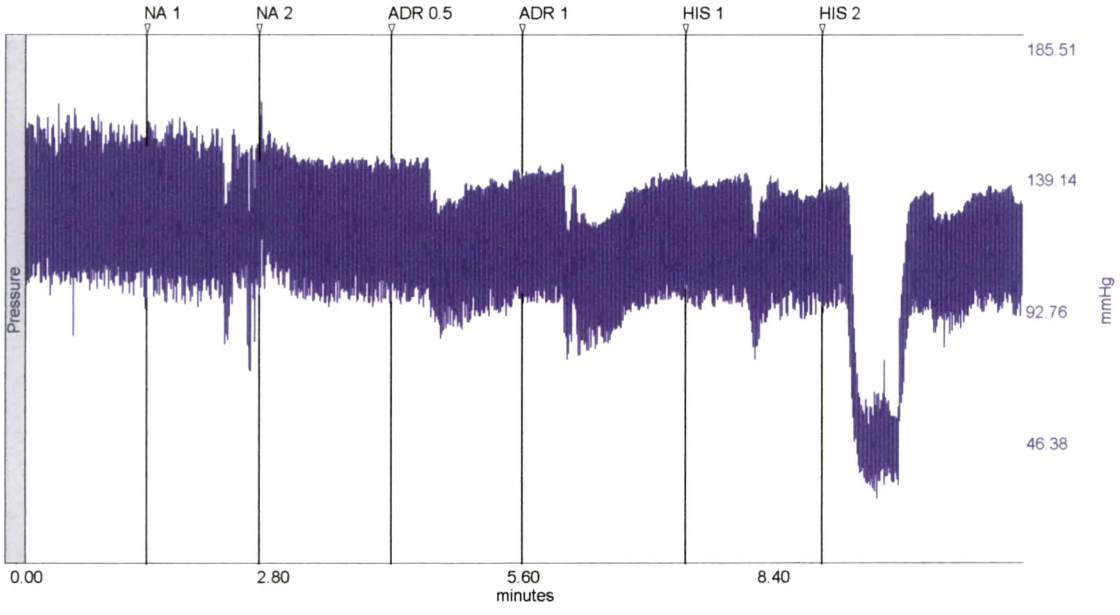

mean change in arterial blood pressure (mmHg)
50
40
30
20
10
0
-10
-20
-30
-40
NA1
NA2
ADR 0.5
ADR 0.5
ADR1
ADR1
HIS1
HIS2
***
***
**
***
*
*
*
*
control
27
Dose of agonists (mcg/kg i.v.)

Figure 16 B: Tracing of blood pressure response after acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats after oral administration of **27** (10 mg/kg).

Abscissa indicates time in minutes and ordinate indicates change in blood pressure (mm Hg).

Tracing of blood pressure response (Figure 16 B) clearly showed the *adrenaline reversal* response (Dale's vasomotor reversal) after oral treatment of **27** (10 mg/kg) indicating the α-adrenoceptor blocking activity of test compound.

Figure 16B


NA 1
NA 2
ADR 0.5
ADR 1
HIS 1
HIS 2
185.51
139.14
92.76
46.38
mmHg
Pressure
0.00
2.80
5.60
8.40
minutes

B.1.1.3.4 Effect of various agonists after the oral administration of 8 (10 mg/kg) on Rat Blood Pressure

Figure 17 A: Mean change in arterial blood pressure (mm Hg) produced by acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (Adr 0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats after oral administration of **8** (10 mg/kg).

Abscissa indicates doses of agonists like noradrenaline, adrenaline, and histamine (μg/kg i.v.) and ordinate indicates mean change in arterial blood pressure (mm Hg).

Vertical line on bars represents SEM (n = 6).

NS = insignificant

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

Figure 17A

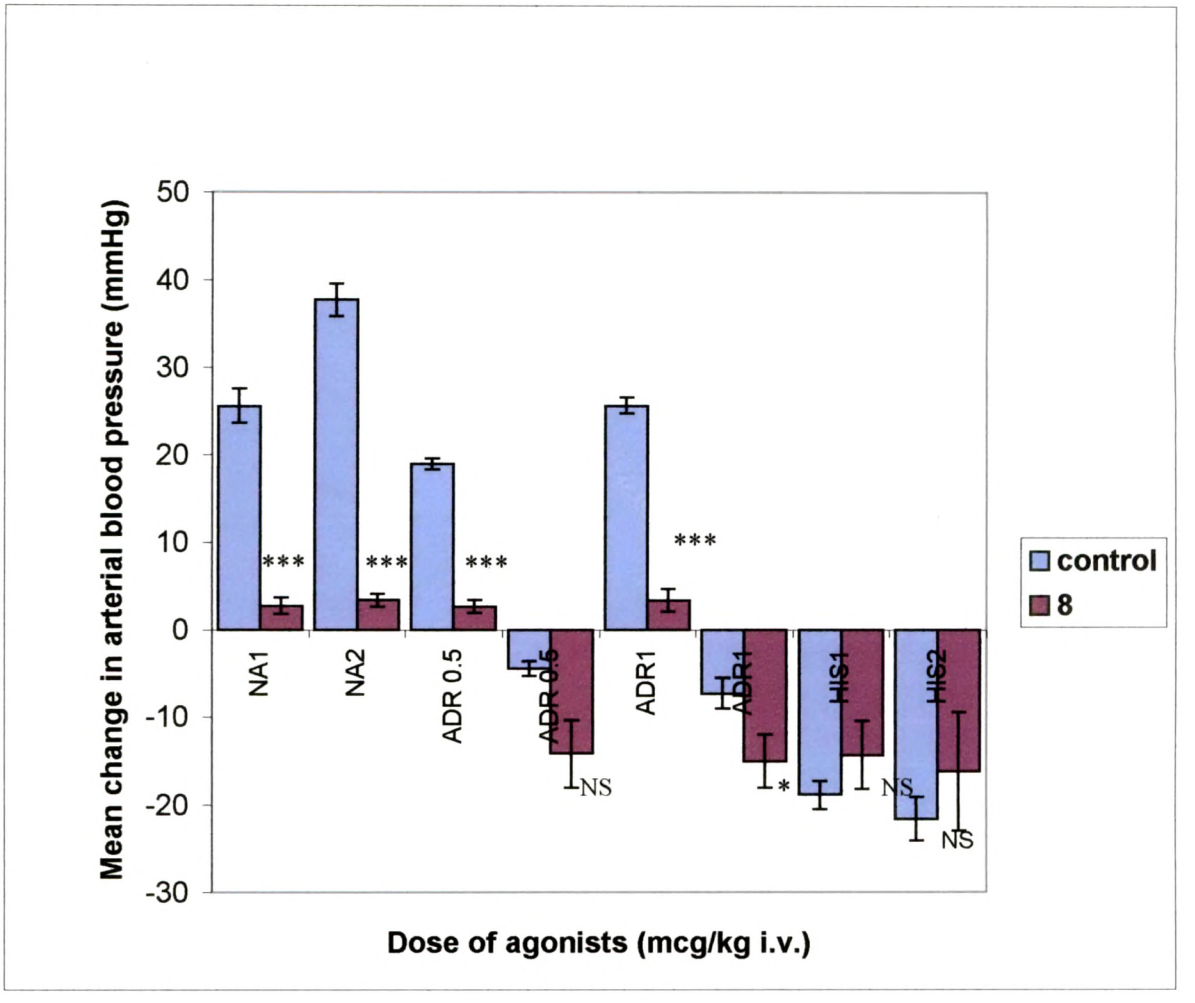
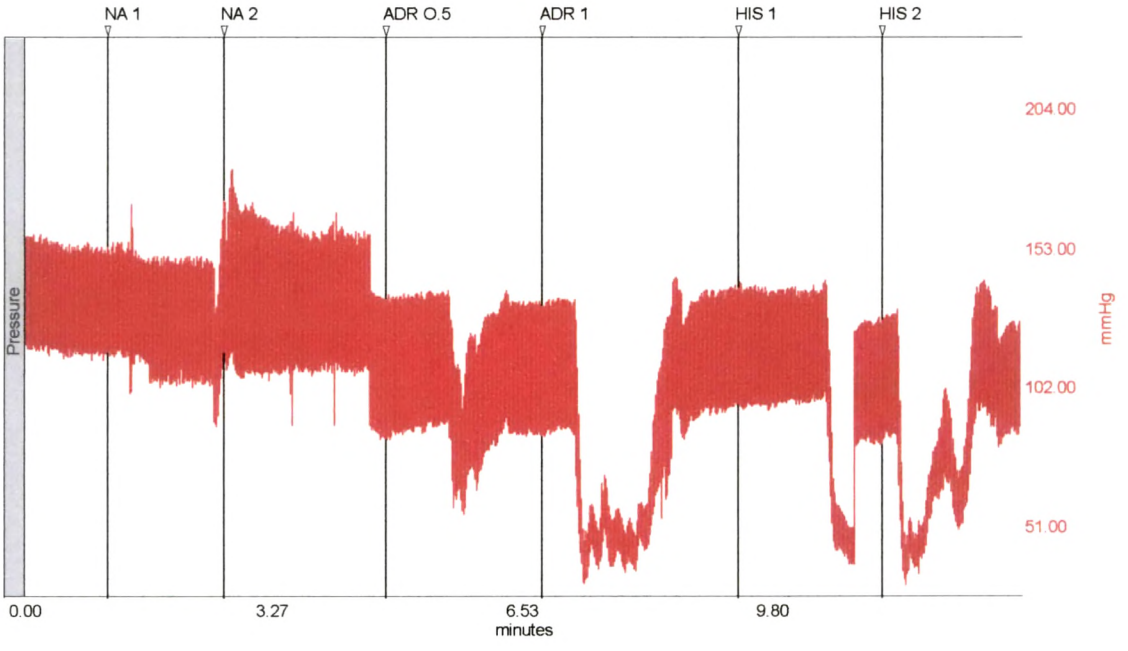

Mean change in arterial blood pressure (mmHg)
50
40
30
20
10
0
-10
-20
-30
***
***
***
***
NS
*
NS
NS
NA1
NA2
ADR 0.5
ADR 0.5
ADR1
ADR1
HIS1
HIS2
control
8
Dose of agonists (mcg/kg i.v.)

Figure 17B: Tracing of blood pressure response after acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (0.5, and 1 μg/kg) and histamine (His 1 and 2 μg/kg) in anesthetized rats after oral administration of **8** (10 mg/kg).

Abscissa indicates time in minutes and ordinate indicates change in blood pressure (mm Hg).

Tracing of blood pressure response (Figure 17 B) clearly showed the *adrenaline reversal* response (Dale's vasomotor reversal) after oral treatment of **8** (10 mg/kg) which indicates the α-adrenoceptor blocking activity of test compound.

Figure 17B


NA 1
NA 2
ADR 0.5
ADR 1
HIS 1
HIS 2
Pressure
204.00
153.00
102.00
51.00
mmHg
0.00
3.27
6.53
9.80
minutes

B.1.1.3.5 Effect of Various Agonists Before And After Oral Administration of 21 on Rat Blood Pressure

The experiment with the test compound was carried out different doses (10 and 20 mg/kg). After 1.5 h from the dosing time of **21** (10 and 20 mg/kg) agonists such as adrenaline (1 µg/kg, iv), noradrenaline (1 µg/kg, iv), angiotensin-II (100 ng/kg, iv) and histamine (1 µg/kg, iv) were administered. A significant blockade ($p<0.01$) response of adrenaline and noradrenaline was observed (fig 18, 19) at 10 mg/kg dose of **21** the degree of blockade of 10 mg/kg dose of **21** to adrenaline and noradrenaline was significantly less as compared to 20 mg/kg doses of **21** (Fig 18, 19). After 3 h from the dosing of test compound, there was complete blockade of responses of adrenaline and noradrenaline. Adrenaline reversal responses was also produced by both the doses (10, 20 mg/kg) of **21** (Fig 23) Reponse to angiotensin-II and histaine were not affected by any of the dose studied (Fig. 20, 21). The effects of vehicle (5% sodium carboxymethyl cellulose) and **21** on normal blood pressure were studied in anestheized rats. Vehicle did not change the normal blood pressure but **21** was significantly ($p< 0.01$) reduced the normal blood pressure (fig 22).

Figure 18: Mean change in blood pressure by acute intravenous injection of adrenaline in anesthetized rats after oral administration of **21** (10(Fig 18A) and 20 (Fig 18B) mg/kg)). Abscissa indicates adrenaline doses and ordinate indicates mean change in blood pressure (mm Hg). Vertical line represents SEM (n = 5)

Fig : 18A

At 1.5 h (NS)

At 3 h ** $p<0.01$ as compared with corresponding control

Fig: 18B

At 1.5 h (NS)

At 3 h *** $p< 0.001$ as compared with corresponding control

Figure18A

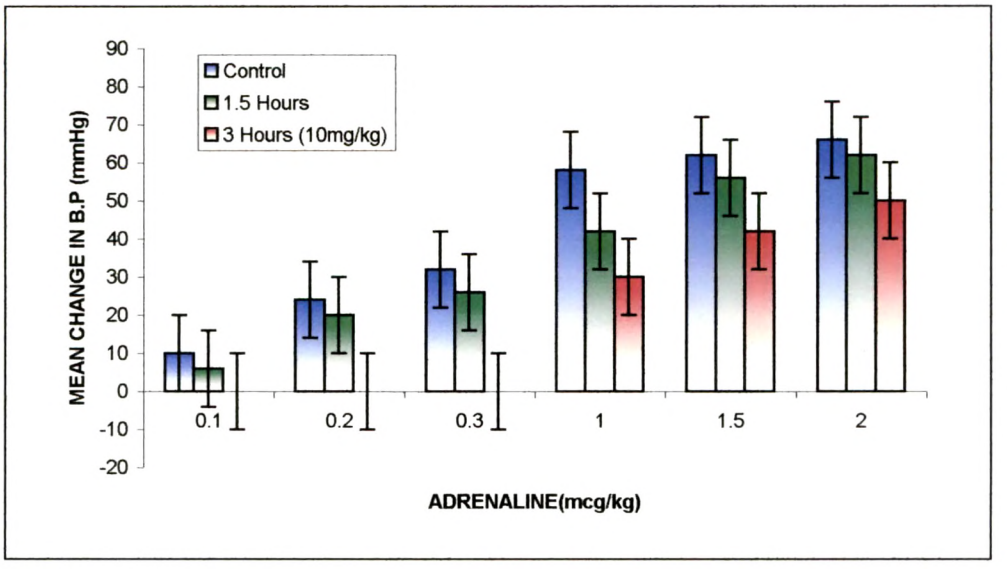


Figure 18B

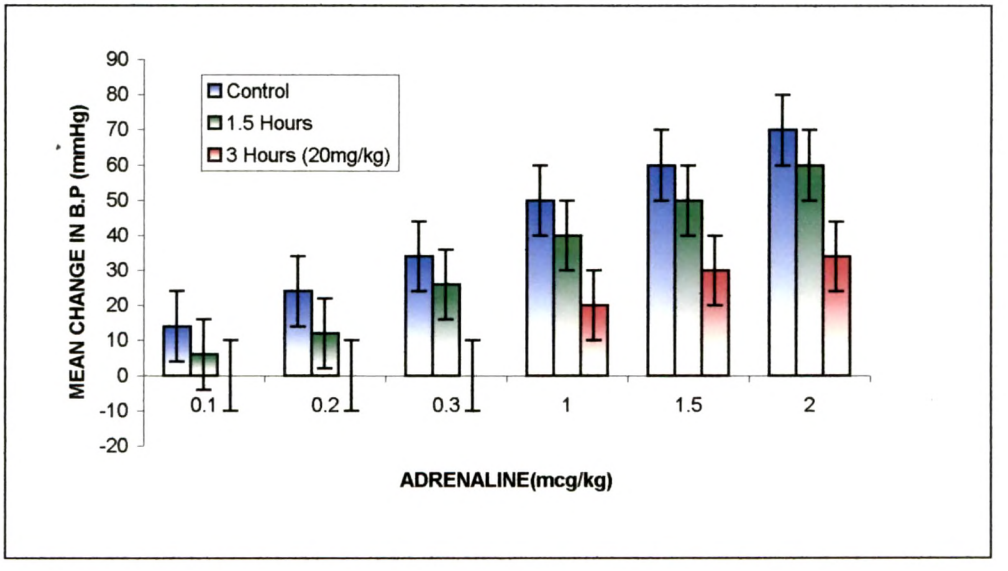


Figure 19: Mean change in blood pressure by acute intravenous injection of noradrenaline in anesthetized rats after oral administration of **21** (10 (Fig19A) and 20 (Fig 19B) mg/kg)). Abscissa indicates noradrenaline doses and ordinate indicates mean change in blood pressure (mm Hg). Vertical line represents SEM (n = 5)

Fig : 19A

At 1.5 h (NS)

At 3 h ** $p<0.01$ as compared with corresponding control

Fig: 19B

At 1.5 h (NS)

At 3 h *** $p< 0.001$ as compared with corresponding control

Figure 19A

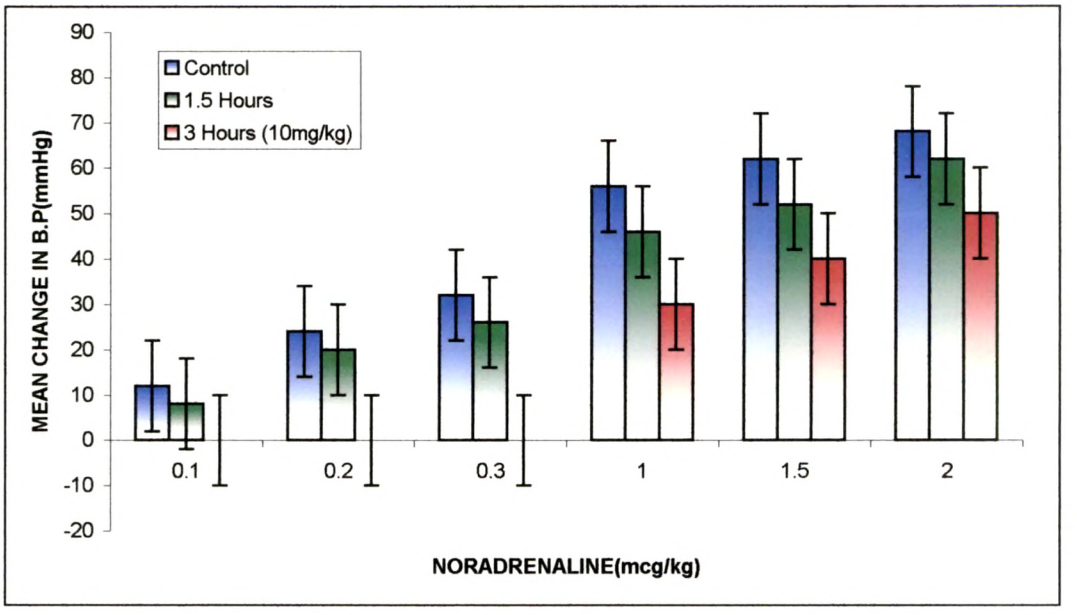


Figure 19B

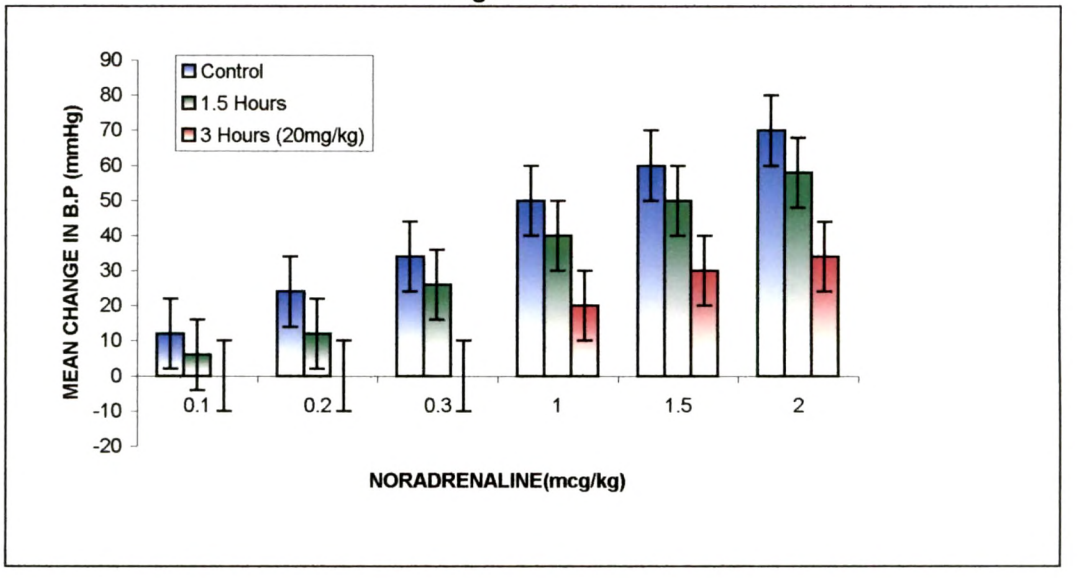


Figure 20: Mean change in blood pressure by acute intravenous injection of angiotensin-II in anesthetized rats after oral administration of **21** (10Fig (20A) and 20 (Fig 20B) mg/kg)). Abscissa indicates angiotensin-II doses and ordinate indicates mean change in blood pressure (mmHg). Vertical line represents SEM (n = 5)

Figure 20A

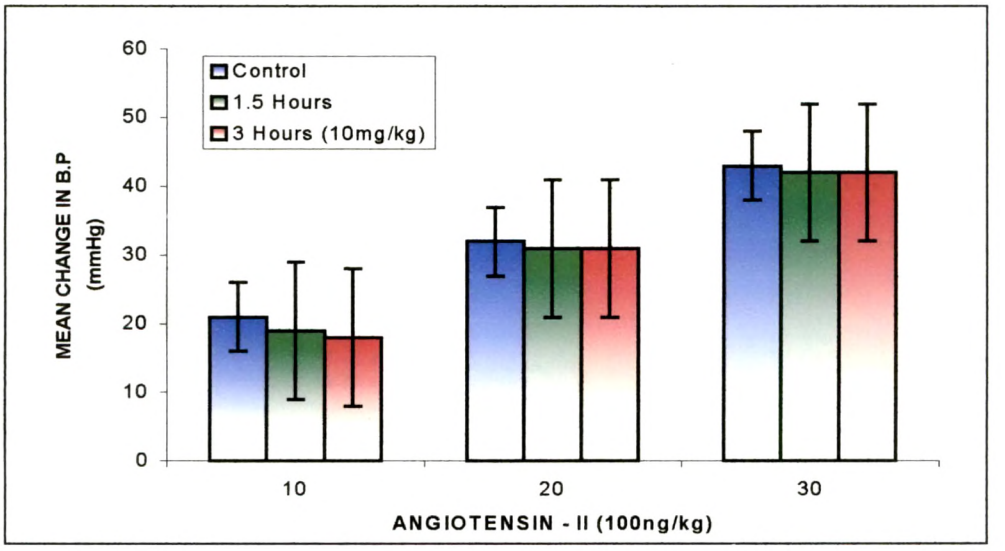


Figure 20B

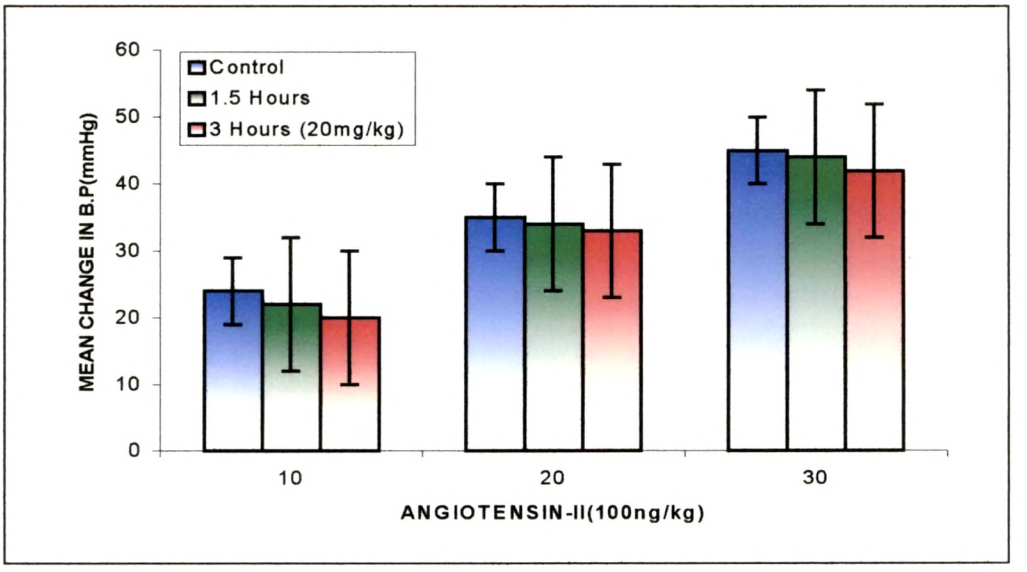


Figure 21: Mean change in blood pressure by acute intravenous injection of histamine in anesthetized rats after oral administration of **21** (10 (Fig (21A) and 20 (Fig 21B) mg/kg)). Abscissa indicates histamine and ordinate indicates mean change in blood pressure (mm Hg). Vertical line represents SEM (n = 5)

Figure 21 A

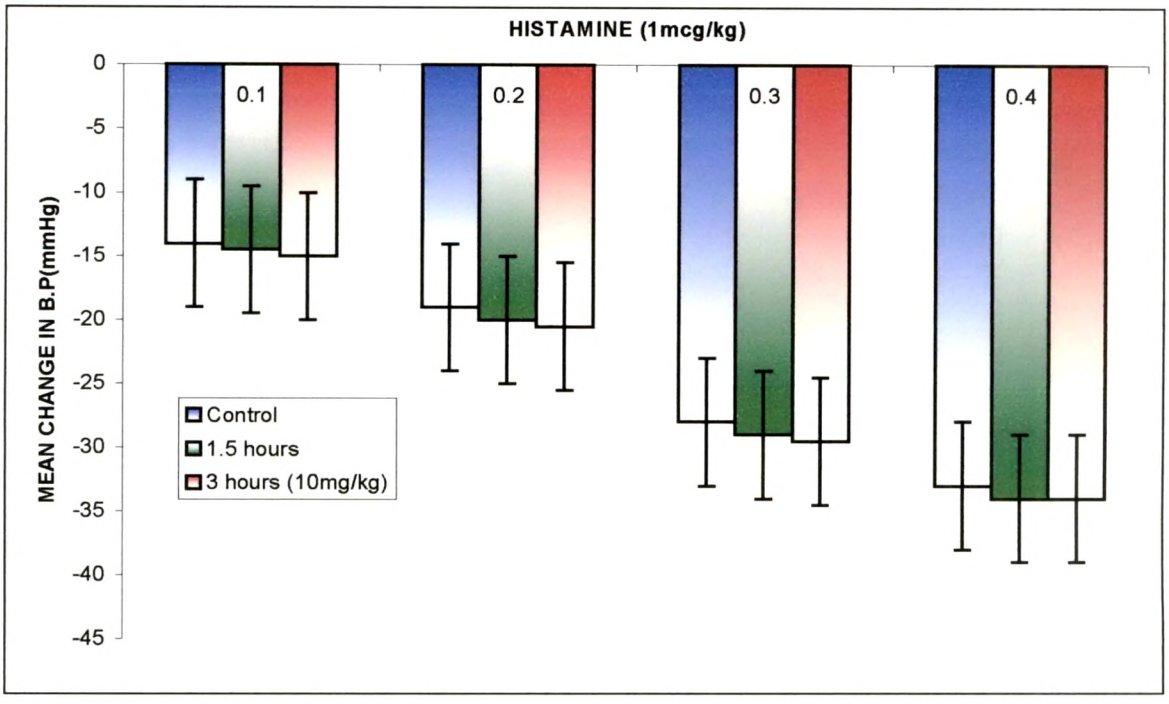


Figure 21B

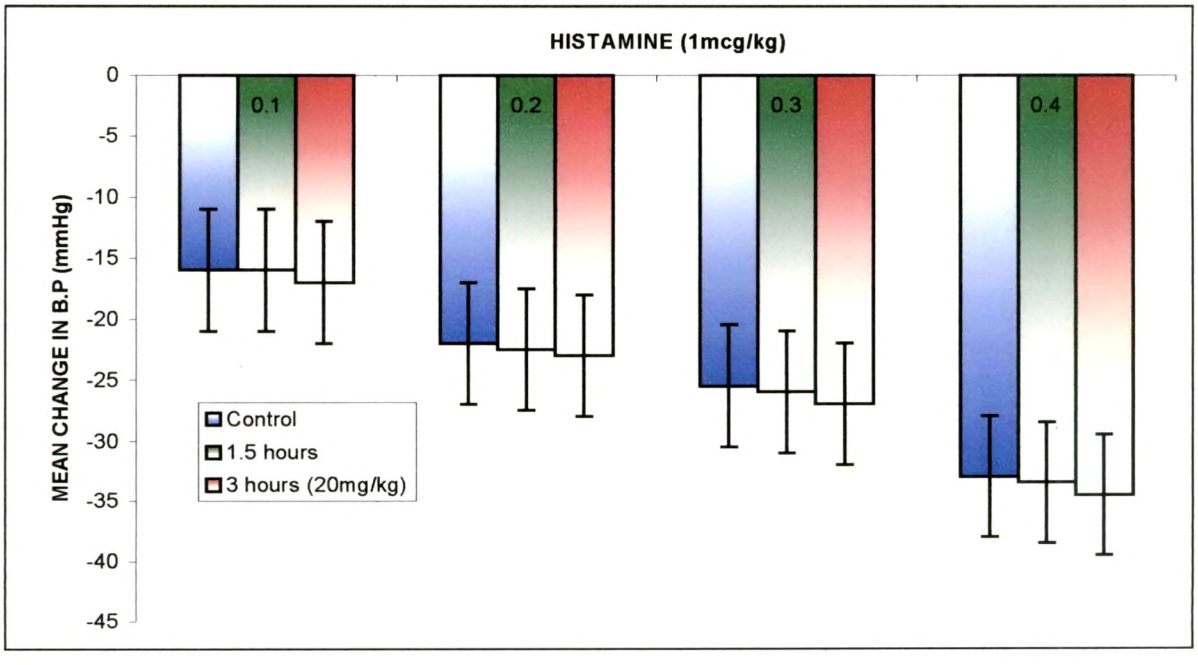


Figure 22: Mean change in normal systolic blood pressure after oral administration of vehicle (5% sodium CMC) and various doses of **21.** Vertical line on histograms represents SEM (n = 5)

A vs B = ** $p<0.01$

A vs C = ** $p< 0.001$ as compared to control

Figure 22

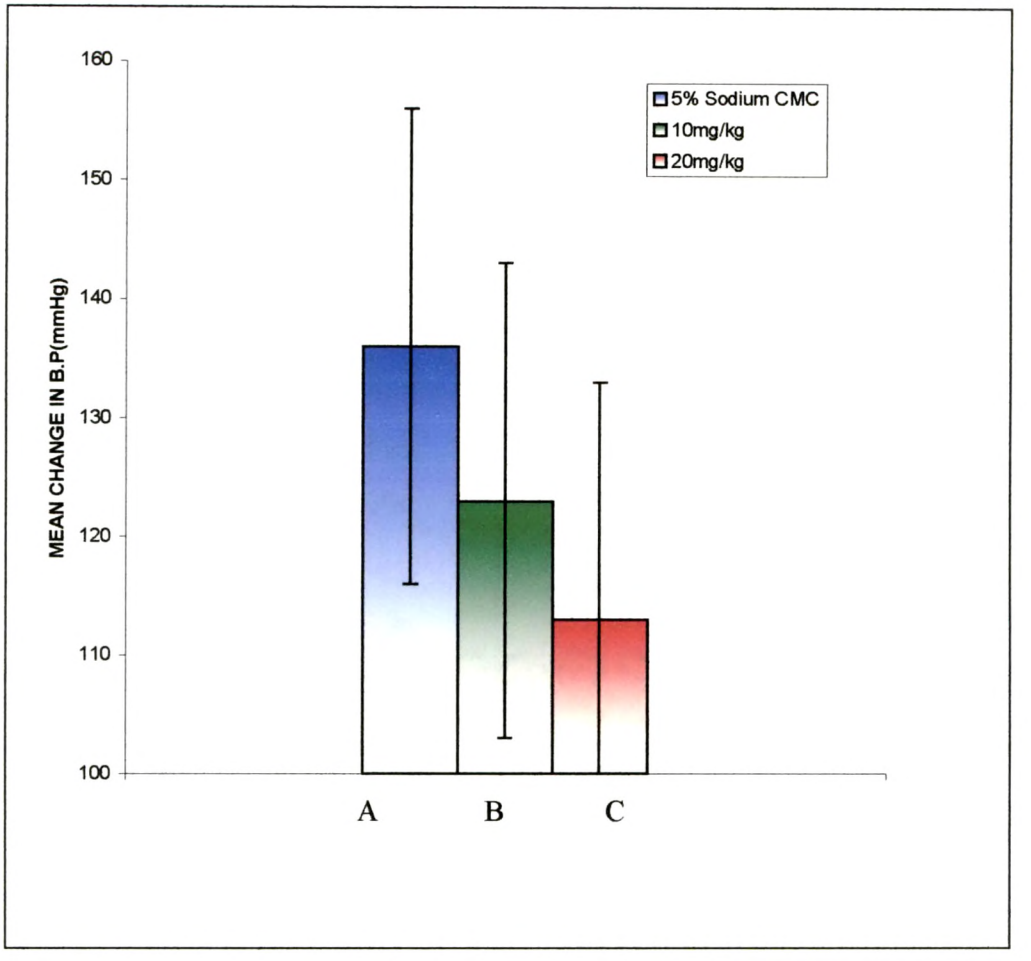
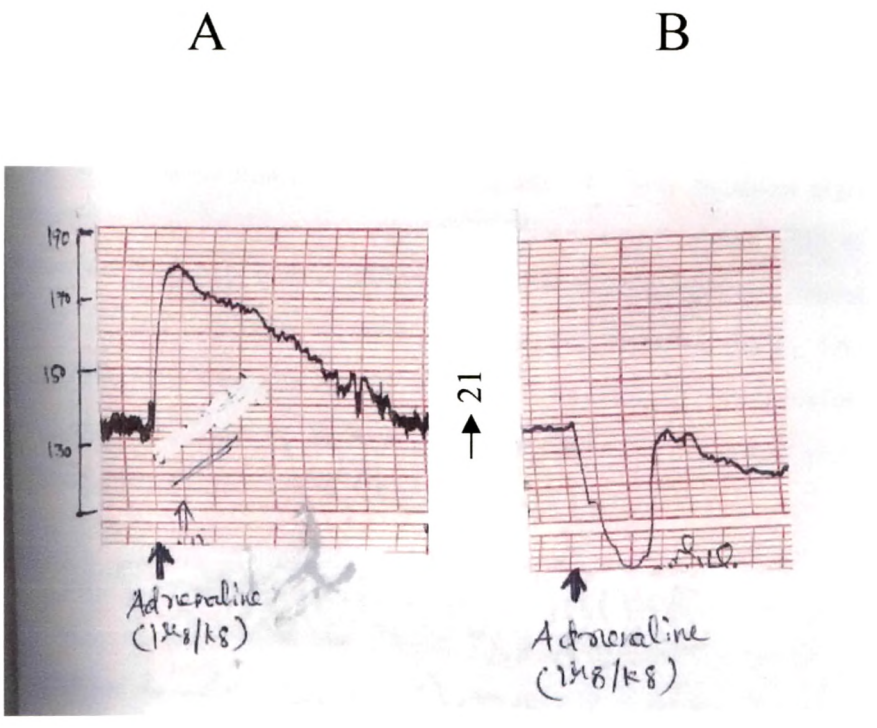

160
150
140
130
120
110
100
MEAN CHANGE IN B.P(mmHg)
5% Sodium CMC
10mg/kg
20mg/kg
A
B
C

Figure 23: Tracing of adrenaline reversal of **21** on anesthetized (Urethane (120 mg/kg, ip) rats Abscissa indicates doses of adrenaline and ordinate indicates the blood pressure (mmHg).

A –Control adrenaline response (1 µg/kg)

B – Adrenaline (1 µg/kg) response in 21 treated animals

Figure 23



B.1.1.3.6 Effect of various agonists after the oral administration of Prazosin (500 μg/kg) on Rat Blood Pressure

Figure 24 A: Mean change in arterial blood pressure (mm Hg) produced by acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (Adr 0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats after oral administration of Prazosin (500 μg/kg).

Abscissa indicates doses of agonists like noradrenaline, adrenaline, and histamine (μg/kg i.v.) and ordinate indicates mean change in arterial blood pressure (mm Hg).

Vertical line on bars represents SEM (n = 6).

NS = insignificant

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

Figure 24A

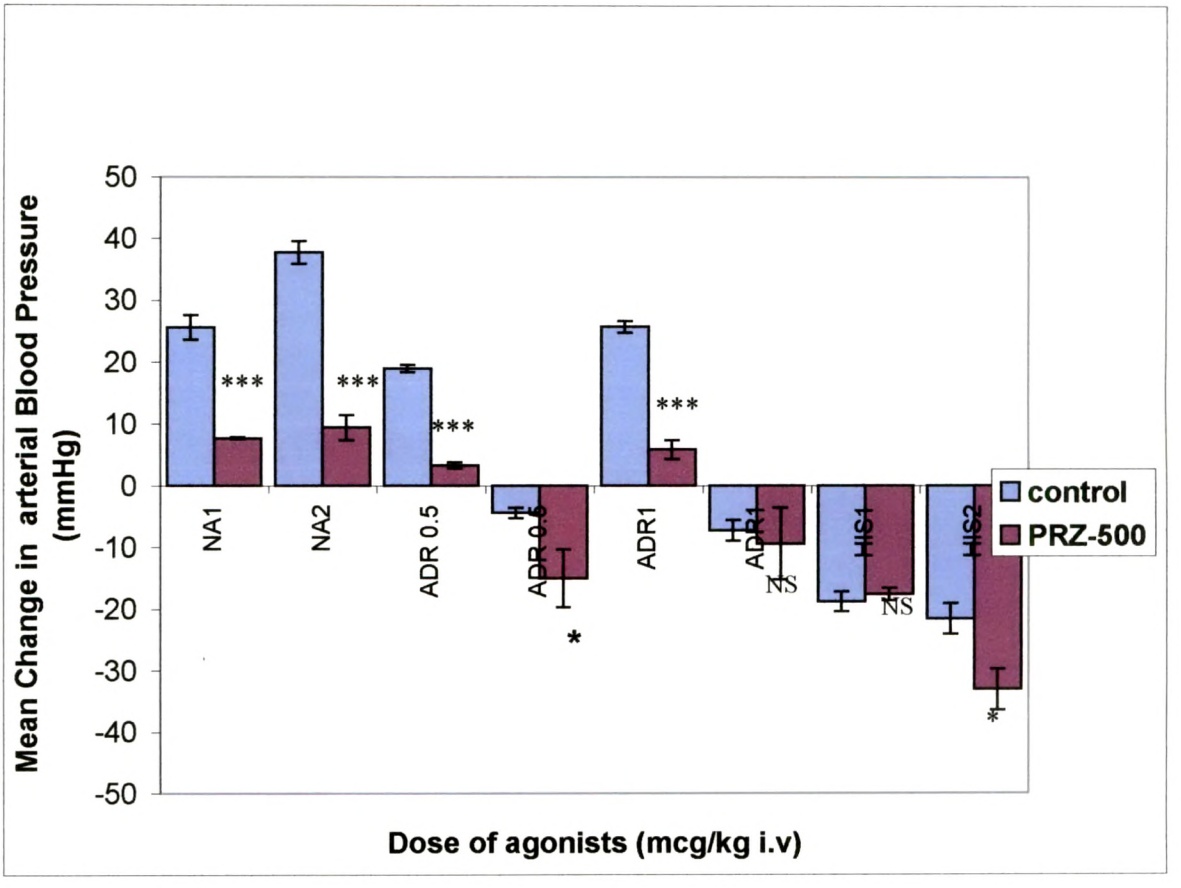
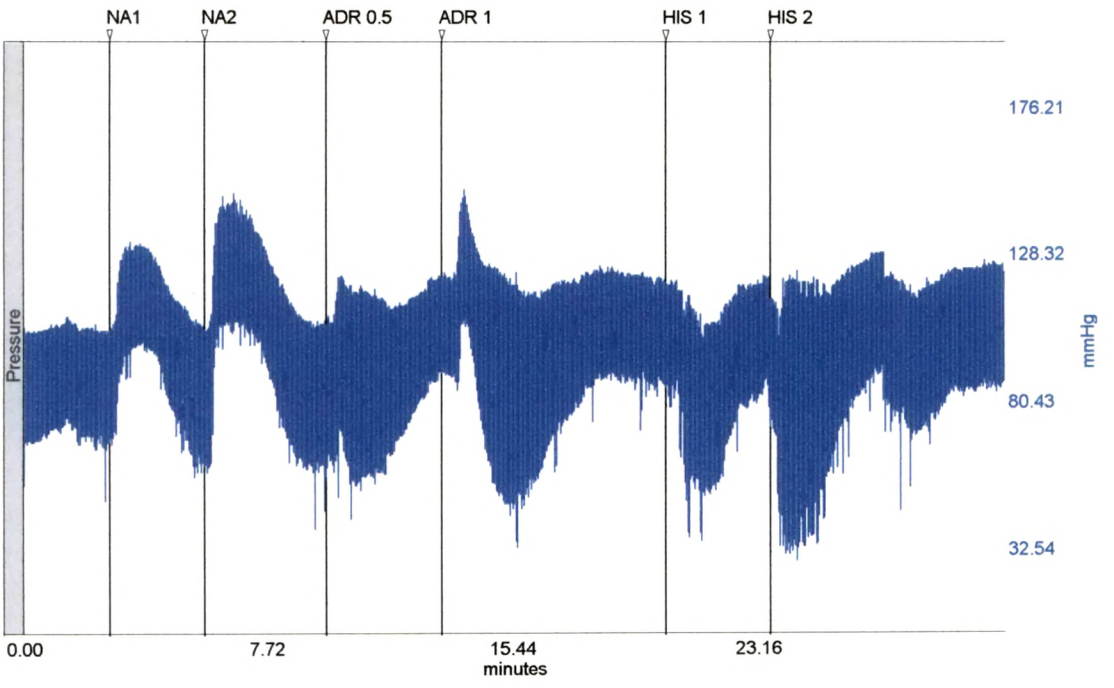

Mean Change in arterial Blood Pressure (mmHg)
50
40
30
20
10
0
-10
-20
-30
-40
-50
***
***
***
***
*
NS
NS
*
NA1
NA2
ADR 0.5
ADR 0.5
ADR1
ADR1
HIS1
HIS2
control
PRZ-500
Dose of agonists (mcg/kg i.v)

Figure 24 B: Tracing of blood pressure response after acute intravenous injections of noradrenaline (NA 1, and 2 μg/kg), adrenaline (0.5, and 1 μg/kg) and histamine (His 1, and 2 μg/kg) in anaesthetized rats after oral administration of Prazosin (500 μg/kg).

Abscissa indicates time in minutes and ordinate indicates change in blood pressure (mm Hg).

Figure 24B


NA1
NA2
ADR 0.5
ADR 1
HIS 1
HIS 2
176.21
128.32
80.43
32.54
mmHg
Pressure
0.00
7.72
15.44
23.16
minutes

B.1.1.3.7 Effect of Phenylephrine (i.v. administration by femoral vein) on rat blood pressure after oral administration of Test Compounds

Figure 25: The α_1-adrenergic receptor blocking action after oral administration of test compounds (Triazolo Quinazolone derivatives) using Phenylephrine i.v. as selective α_1- adrenoceptor agonists.

Abscissa indicates doses of Phenylephrine (µg/kg, i.v.) and ordinate indicates Mean Change in Arterial Blood Pressure (mm Hg).

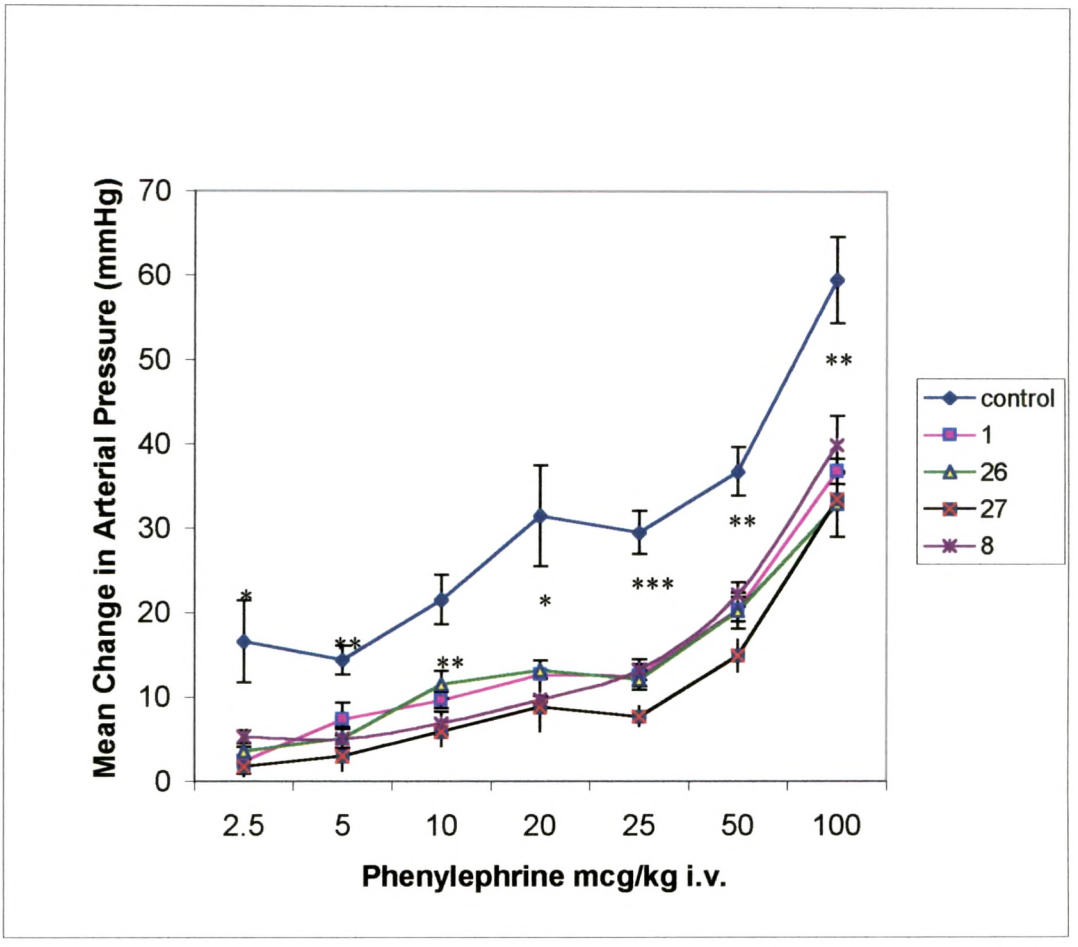
Vertical line on bars represents SEM (n = 6).

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

Figure 25


Mean Change in Arterial Pressure (mmHg)
70
60
50
40
30
20
10
0
2.5
5
10
20
25
50
100
Phenylephrine mcg/kg i.v.
control
1
26
27
8

B.1.2 *In vitro* Study

The *in vitro* study showed rightward shift of phenylephrine concentration Response Curve (CRC) in **1**, **26**, **27**, and **8** treated rat aorta as compared to control.

Figure 26 A: the Concentration Response Curve (CRC) of isolated rat aorta from control and **1** (10 mg/kg, p.o.) & **26** (10 mg/kg, p.o.) treated rats.

Abscissa indicates Log dose of PE in nM (nanomolar) concentration and ordinate indicates % maximum contraction of aortic strip.

Vertical bars on graphical lines represent SEM (n = 4)

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

Figure 26A

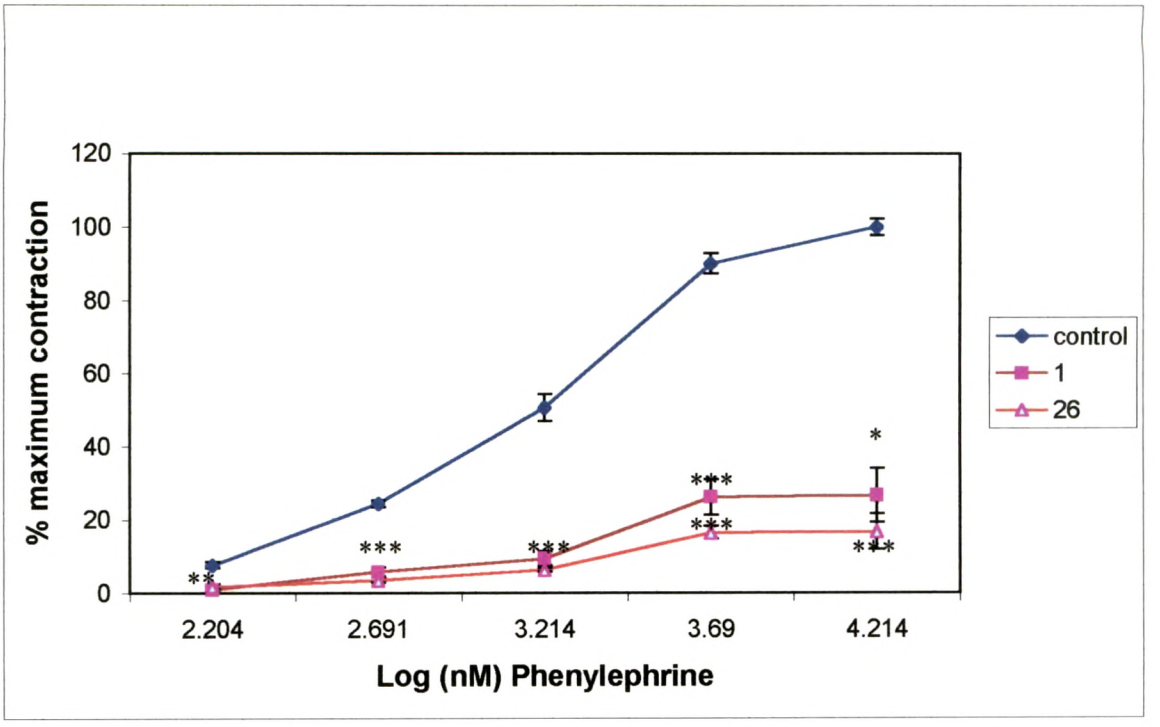

120
100
80
60
40
20
0
% maximum contraction
2.204
2.691
3.214
3.69
4.214
Log (nM) Phenylephrine
control
1
26
**
***
***
***
***
*
**

Figure 26 B 1 -tracing of aortic strip of control rats.

Figure 26 B-2 -tracing of aortic strip of **1** & **26** (10 mg/kg per oral) treated rats.

Figure 27 B represents the tracing of aortic strip of **27** & **8** (10 mg/kg per oral) treated rats.

PE = Phenylephrine

Figure 26 B-1

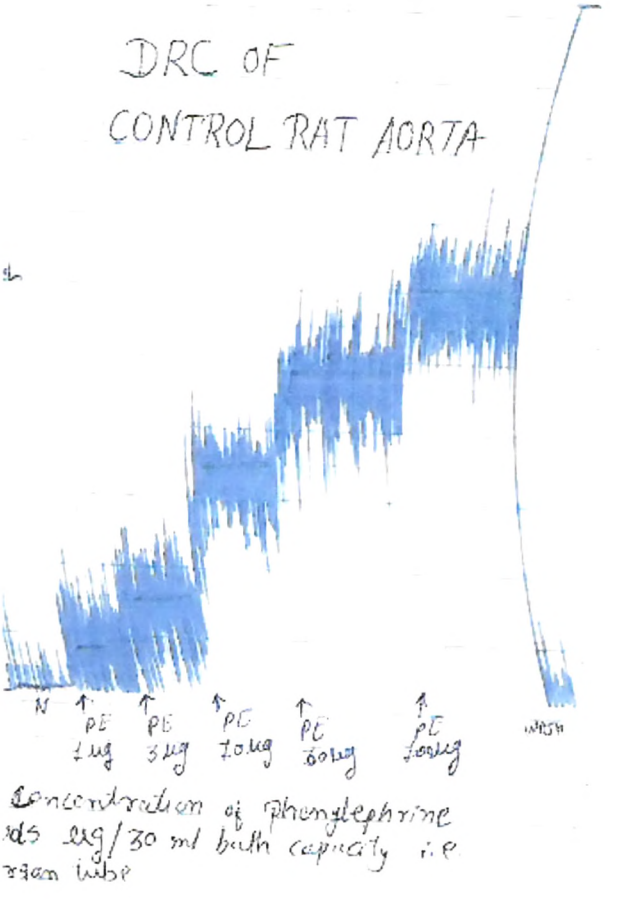

DRC OF
CONTROL RAT AORTA
N
PE 1ug
PE 3ug
PE 10ug
PE 30ug
PE 100ug
WASH
Concentration of phenylephrine
ads 2ug/30 ml bath capacity i.e
organ tube

Figure 26 B-2

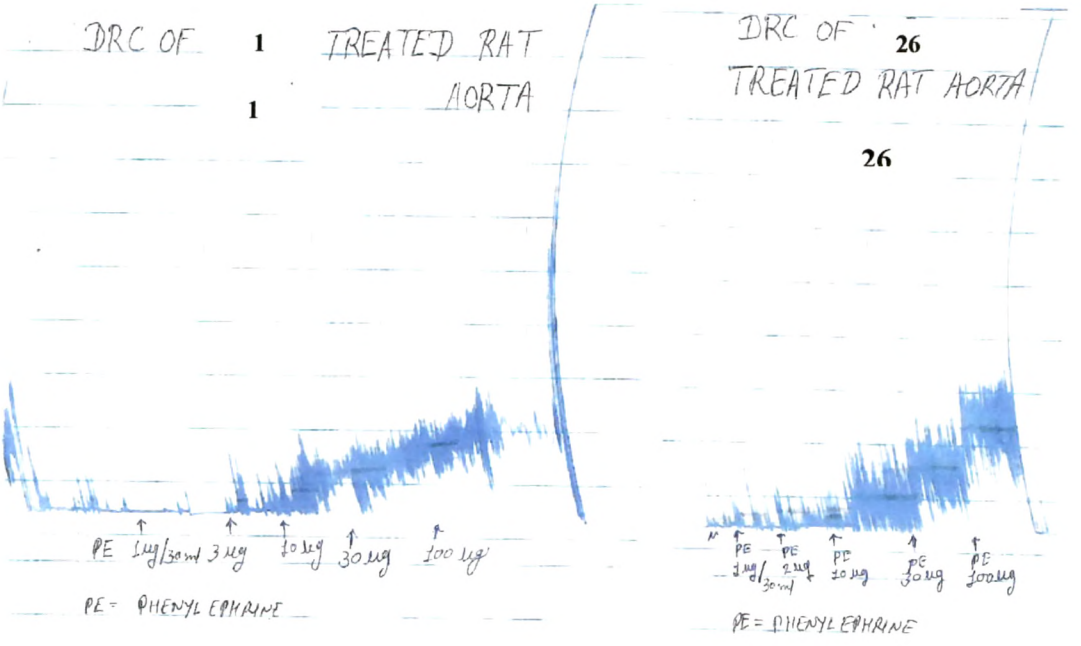

DRC OF 1 TREATED RAT AORTA
1
PE 1µg/30ml 3µg 10µg 30µg 100µg
PE = PHENYL EPHRINE
DRC OF 26
TREATED RAT AORTA
26
PE 1µg/30ml PE 2µg PE 10µg PE 30µg PE 100µg
PE = PHENYL EPHRINE

Figure 27 A : The Concentration Response Curve (CRC) of isolated rat aorta from control and **27** (10 mg/kg p.o.) & **8** (10 mg/kg p.o.) treated rats.

Abscissa indicates Log dose of PE in nM (nanomolar) concentration and ordinate indicates % maximum contraction of aortic strip.

Vertical bar on graphical lines represents SEM (n = 4).

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

Figure 27A

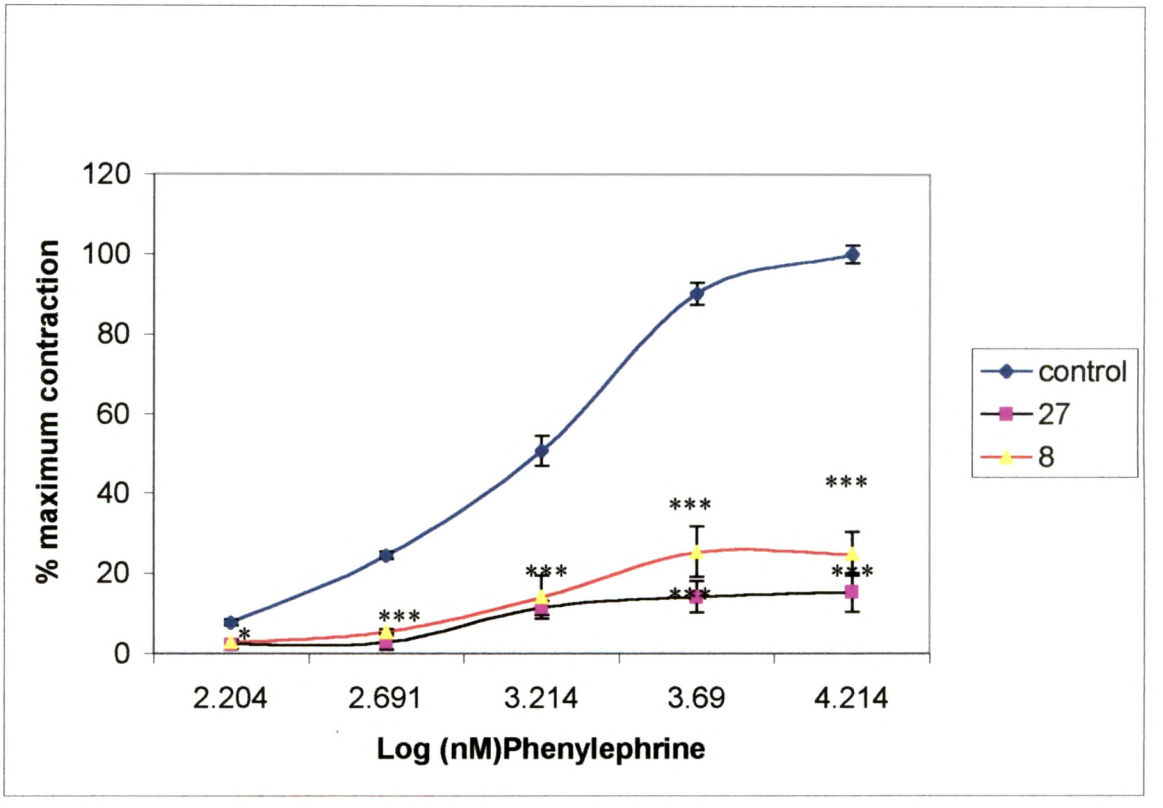
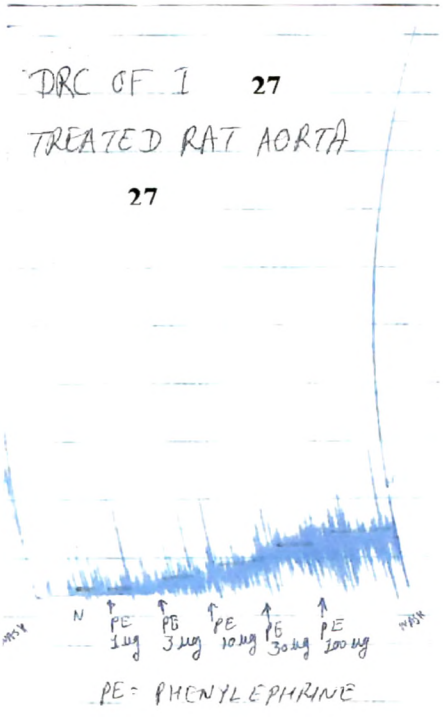
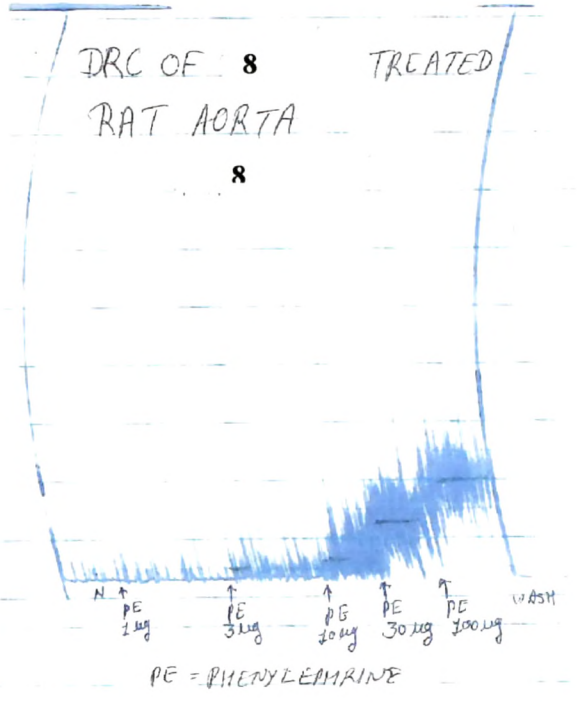

120
100
80
60
40
20
0
% maximum contraction
2.204
2.691
3.214
3.69
4.214
Log (nM)Phenylephrine
control
27
8
*
***
***
***
***
***

Figure 27- B

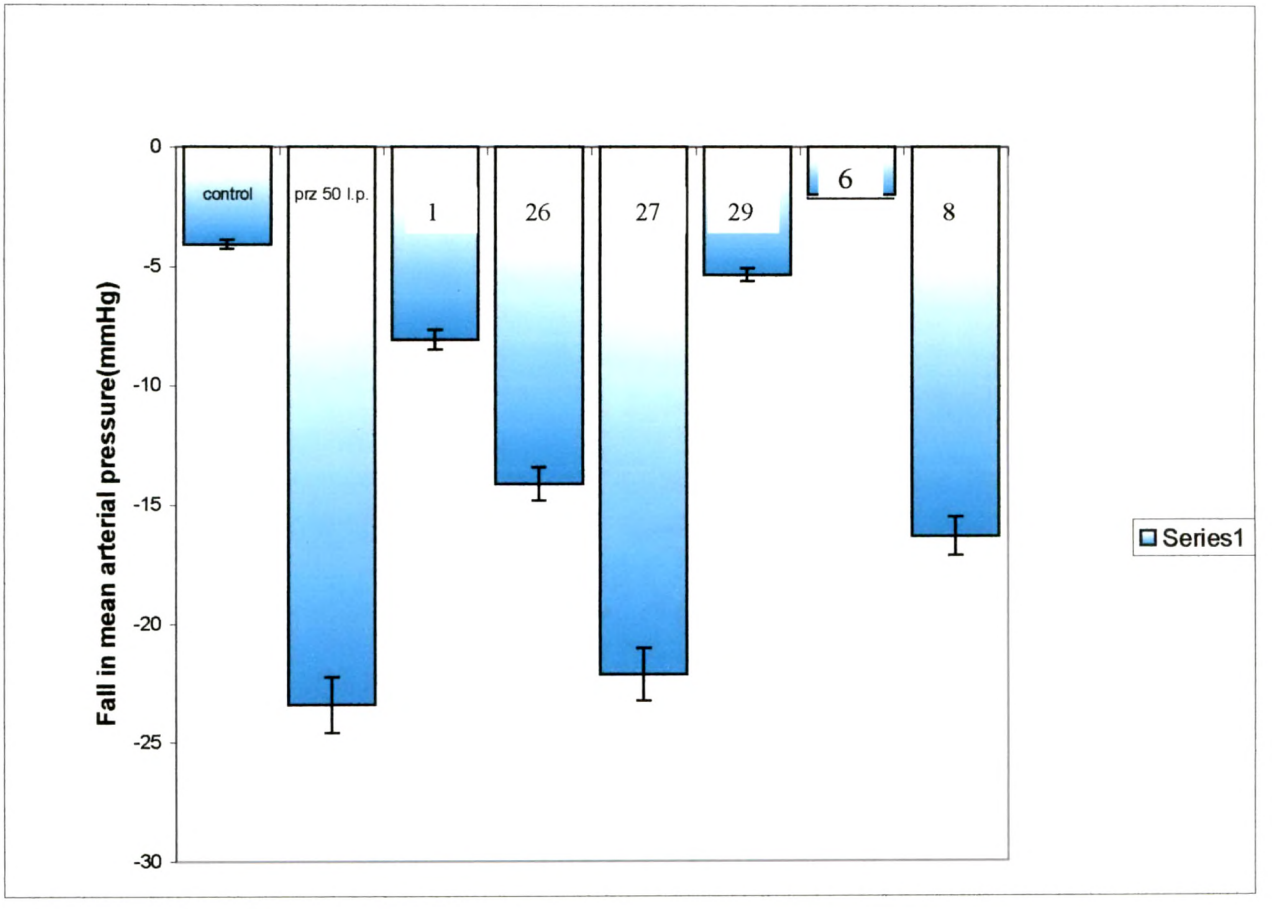




B.1.3 Comparison of Hypotensive Action of Test Compounds After Intraperitoneal Injection

Figure 28: Comparison of hypotensive action after i.p. administration of test compounds and it shows that **27** had the maximum hypotensive action at the dose of 5 mg/kg i.p. as compared to control where as Prazosin showed almost similar hypotension after i.p. at the dose of 50 μg/kg.

Figure 28


Fall in mean arterial pressure(mmHg)
0
-5
-10
-15
-20
-25
-30
control
prz 50 l.p.
1
26
27
29
6
8
Series1

B.2. Antihistaminic activity

Thirty compounds containing 1,4-disubstituted-1,2,4-triazoloquinazoline ring system have been evaluated for their in *vivo* antihistaminic activity. Protection against histamine induced bronchospasm on conscious guinea pigs method was adopted to determine the antihistaminic potential of the test compounds. All of them have been found to exhibit good antihistaminic activity (Table 1). Percentage protection data showed that all compounds of the series show significant protection in the range of 68-72%.

Structure activity relationship (SAR) studies indicated that different substituents on the N-4 aromatic ring, exerted varied biological activity. The electronic nature of the substituent group of N-4 aromatic ring led to a significant variation in antihistaminic activity. For example electron withdrawing group (chloro and methoxy substituents) enhanced the biological activity, whereas electron releasing groups (methyl) made the compounds less active, and the order of activity was p-chlorophenyl > pyridyl > p-methoxyphenyl > phenyl > tolyl. SAR studies also indicated that different alkyl substituents over the first position of triazoloquinazoline ring exerted varied biological activity. It has been found that presence of methyl group showed better activity over the unsubstituted compounds. When chain length increased from methyl to ethyl and propyl the activity decreased. Replacement of a proton of the methyl group by chloro showed further decrease in activity. While the order of activity was methyl > ethyl > unsubstituted > propyl >

chloromethyl. Among the series, 1-methyl-4-(4-chlorophenyl)-*s*-triazoloquinazolin-4(3*H*)-one (**22**) was the most potent with the percentage protection of 72.71 which is equipotent with that of standard chlorpheniramine maleate (percentage protection 71.00) but less potent than standard certirizine (percentage protection 78.95) and aminophylline (percentage protection 90.29).

As the test compounds could not be converted to water soluble form, *in vitro* evaluation for antihistaminic activity could not be performed.

Table 1 : *In vivo* Antihistaminic Activity of 1,4-disubstituted-*s*-triazoloquinazolines.

Drug Code	Mean ± SEM (Time of onset of Convulsion in Sec.)	% Protection
1	385± 7.51*	69.87
2	396 ± 6.47*	70.70
3	387 ± 7.56*	70.03
4	377 ± 7.96*	69.23
5	370 ± 5.24*	68.65
6	372 ±2.43*	68.82
7	383±8.99*	69.71
8	376±8.36*	69.15
9	369±2.79*	68.56
10	368±5.07*	68.48
11	371±3.97*	68.73
12	382±7.13*	69.63
13	372±2.42*	68.82
14	371±4.33*	68.73
15	367±3.70*	68.39
16	384±4.73*	69.79
17	400±5.86*	71.00
18	389±7.75*	70.18
19	373±5.08*	68.90
20	371±4.24*	68.73
21	396±6.20*	70.71
22	425±12.25*	72.71
23	394±6.79*	70.56
24	389±8.20*	70.18

25	382±8.46*	69.63
26	388±9.03*	70.10
27	406±11.03*	71.43
28	390±7.14*	70.26
29	386±8.23*	69.95
30	381±7.64*	69.55
Control	116±2.02*	----
Chlorpheniramine maleate	400±29.50*	71.00
Cetirizine	551±16.89*	78.95
Aminophylline	1193±35.49*	90.29

Each value represents the mean ± SEM (n=6). Significance levels: *p<0.001 as compared with the respective control.

Dose of test compounds : 10 mg/kg

Figure 29: Comparision of *Invivo* Antihistaminic Activity of 1,4-disubstituted-*s*-triazoloquinazolines

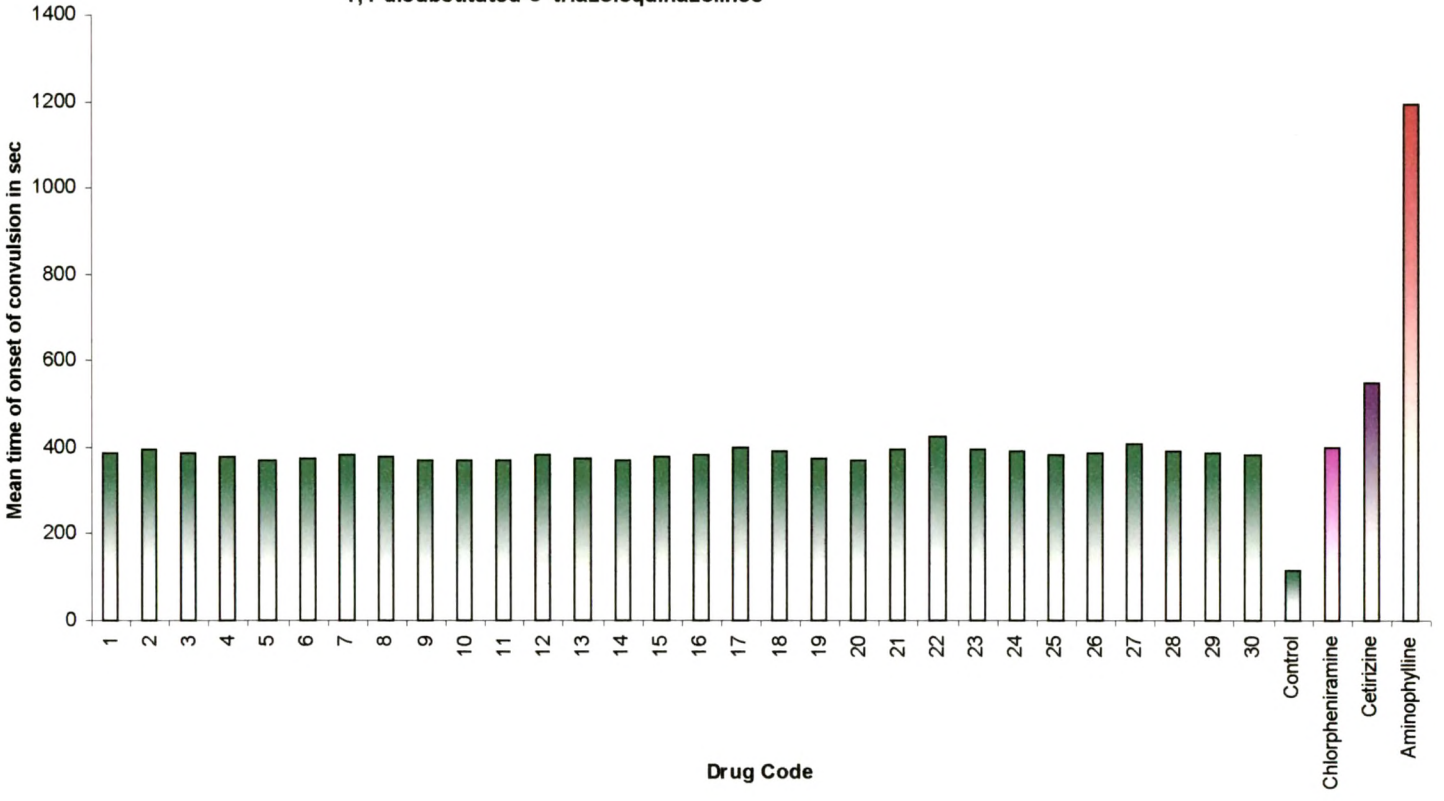
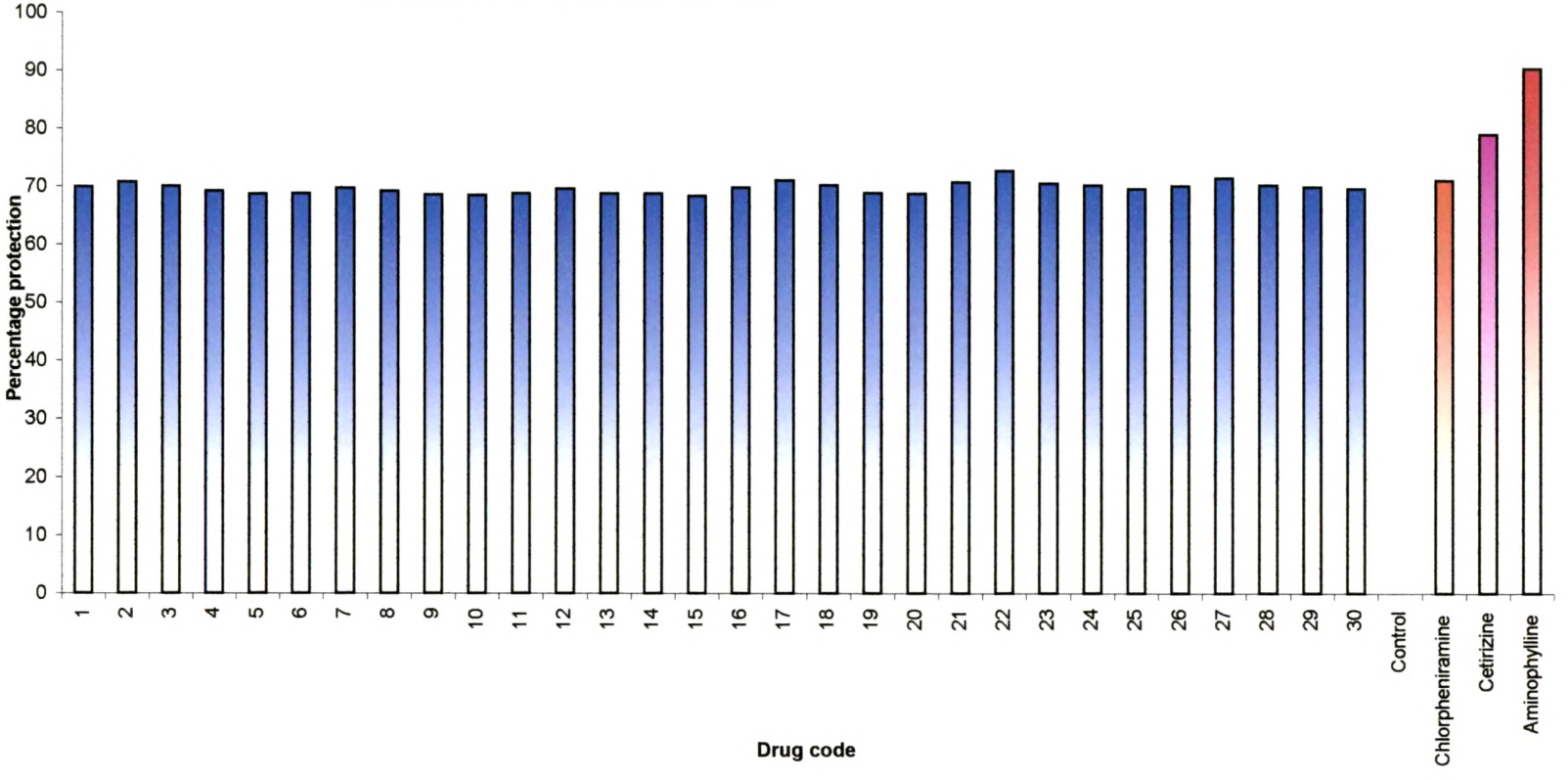


Figure 30 : Comparision of *in vivo* Antihistaminic Activity (% protection) of 1,4-disubstituted-*s* -triazoloquinazolines



B.3. Sedative-hypnotic activity

As sedation is one of the major side effects associated with antihistamines, the test compounds were also evaluated for their sedative potentials.

Sedative-hypnotic activity was determined by measuring the reduction in motor activity. The results of this study (Table 2 & 3) showed that almost all the test compounds were found to exhibit mild activity (less than 10 %), except for the compounds **3**, **4**, **6**, **7**, **8**, **9**, **12**, **13**, **14**, **19**, **28** and **29** which to exhibited moderate activity (10-15 %). Cetirizine and chlorpheniramine maleate as references showed 9% and 26% sedation, respectively.

Table 2 : Sedative–hypnotic activity of 1,4-disubstitued-*s*-triazoloquinazolines

Drug Code	Mean Basal Score	Score after drug treatment			
		30 min	1 h	2h	3h
1	32.0	30.4 ± 1.21*	29.0 ± 1.05*	28.0 ± 1.05**	29.8 ± 0.97*
2	30.4	29.0 ± 1.26*	27.2 ± 1.49**	26.4 ± 1.63***	27.4 ± 0.98**
3	32.4	30.6 ± 1.60*	28.4 ± 1.40 *	28.0 ± 1.64*	28.8 ± 1.98*
4	31.2	28.8 ± 0.58***	26.8 ± 1.08***	26.0 ± 1.09****	27.4 ± 1.29**
5	29.8	28.2 ± 0.80***	27.4 ± 1.12***	27.0 ± 0.89***	27.6 ± 1.03**
6	32.2	30.2 ± 1.02*	28.8 ± 1.02*	27.2 ± 2.01**	27.8 ± 2.06**
7	30.2	27.4 ± 0.68****	25.2 ± 1.07*****	25.0 ± 0.77*****	27 ± 1.26***
8	31.4	28.4 ± 1.50**	27.0 ± 1.58***	25.4 ± 1.50****	27.4 ± 1.12*
9	29.0	26.0 ± 1.22*****	24.0 ± 1.51*****	23.6 ± 1.39*****	25.2 ± 1.85***
10	32.0	30.2 ± 1.49*	29.2 ± 1.35*	28.4 ± 1.25*	29.8 ± 1.16*
11	34.0	32.4 ± 0.68 NS	31.0 ± 0.45*	29.8 ± 0.73*	30.6 ± 0.75*
12	31.0	28.8 ± 1.02**	26.0 ± 1.05****	26.0 ± 1.26****	27.8 ± 1.42**
13	32.8	29.4 ± 1.69*	28.4 ± 1.03**	27.4 ± 1.88**	28.8 ± 1.07*
14	32.0	28.2 ± 1.53**	26.4 ± 1.16 ****	26.2 ± 1.16****	28.0 ± 1.61 *
15	31.2	29.2 ± 1.02**	28.2 ± 1.35**	27.4 ± 1.21***	28.8 ± 1.20*
16	32.0	30.6 ± 1.50*	29.8 ± 1.39 *	28.6 ± 1.57*	29.4 ± 1.43*
17	31.2	29.4 ± 1.47*	28.0 ± 1.41**	26.8 ± 1.39***	28.4 ± 1.63*
18	29.2	27.4 ± 1.57***	25.6 ± 1.72***	25.2 ± 1.82***	27.0 ±1.76**
19	32.8	30.6 ± 1.93*	28.8 ± 1.59*	28.0 ± 1.41**	28.8 ± 1.24*
20	32.4	30.8 ± 1.93*	30.0 ± 1.87*	29.4 ± 1.60 *	30.2 ± 1.59*
21	31.0	30.0 ± 1.64*	28.0 ± 1.73*	27.6 ± 1.94*	28.6 ±1.63 *
22	31.8	30.4 ± 1.86 *	28.8 ± 1.85*	27.8 ± 2.08 *	29.2 ± 2.56*
23	30.4	28.8 ± 1.93 *	27.0 ± 1.87**	26.4 ± 1.5 ***	27. 2 ± 1.90**
24	30.0	28.0 ± 2.28 *	27.2 ± 1.89**	26.8 ± 1.74**	27. 4 ± 1.91*
25	29.2	27.8 ± 1.24**	26.6 ± 1.21***	25.2 ± 1.32****	27. 2 ± 1.28 **
26	33.2	31.8 ± 1.82 NS	30.4 ± 1.94*	29.4 ± 1.47*	31.2 ± 1.91 NS

27	31.6	30.4 ± 2.25*	28.0 ± 1.87*	27.4 ± 1.72**	29.2 ± 2.20*
28	34.4	33.0 ± 1.18 ^{NS}	30.4 ±1.21*	30.0 ± 1.51*	31.4 ± 1.21 ^{NS}
29	29.2	27.6 ± 1.72**	25.0 ± 1.40*****	25.4 ± 1.30*****	25.8 ± 1.65****
30	30.4	29.4 ± 1.88*	28.0 ± 1.92 *	27.4 ± 2.01*	28.4 ± 2.13*
Control	33.2	32.6 ± 1.50	32.2 ± 1.60	32.2 ± 1.68	32.4 ± 1.96
Chlorpheniramine	32.4	28.6 ± 1.96*	21.2 ± 1.82*****	22.0 ± 1.73*****	25.2 ± 1.98***
Cetirizine	31.0	29.6 ± 1.36*	28.0 ± 1.10 **	27.4 ± 1.08***	28.4 ± 1.03*

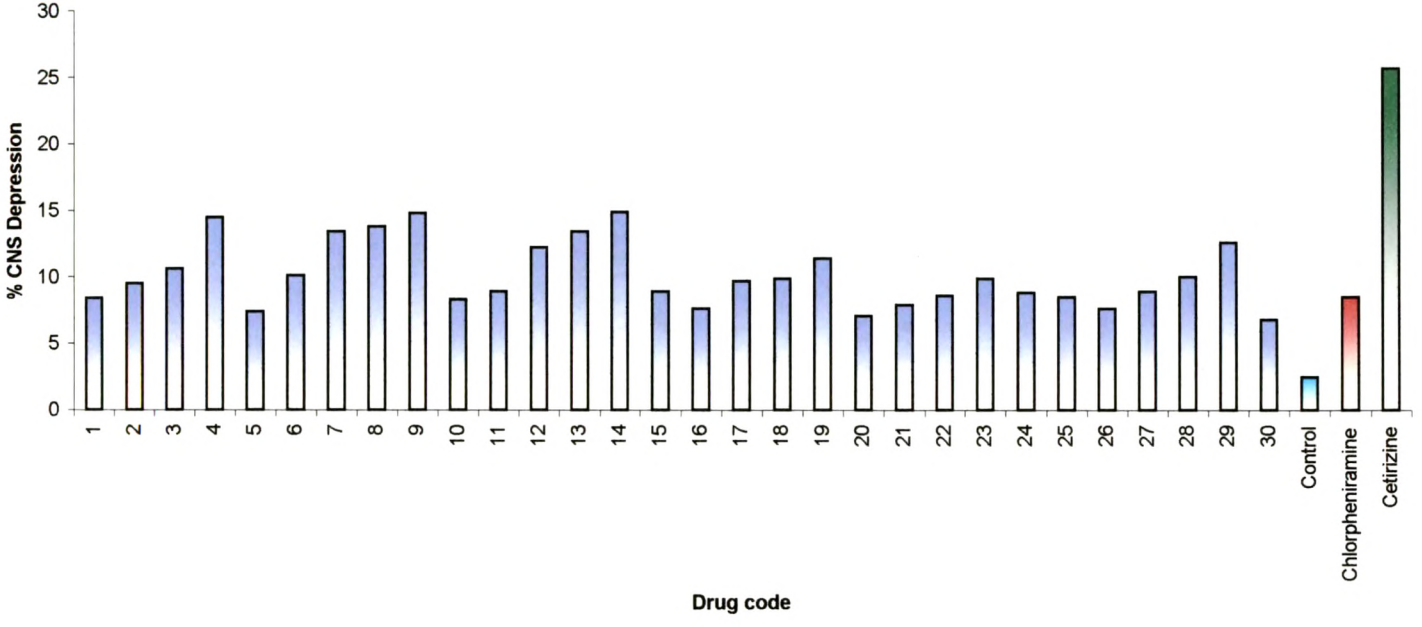
Each value represents the mean ± SEM (n=5). Significance levels *p<0.5, **p<0.1, ***p<0.05, ****p<0.01 and *****p<0.001 as compared with the respective control.

Dose of test compounds : 10 mg/kg , Cetirizine 5 mg/kg, Chlorpheniramine maleate 5 mg/kg.

Table 3 : Percent CNS depression of 1,4-disubstitued-*s*-triazoloquinazolines

Drug Code	Percent CNS Depression				
	30 min	1 h	2 h	3 h	Average
1	5.0	9.3	12.5	6.9	8.4
2	4.6	10.5	13.1	9.7	9.5
3	5.5	12.3	13.5	11.1	10.6
4	7.6	14.1	16.6	12.1	14.5
5	5.3	8.0	9.4	6.7	7.4
6	6.2	10.5	15	8.6	10.1
7	9.2	16.5	17.2	10.5	13.4
8	9.5	14.0	19.1	12.7	13.8
9	10.3	17.2	18.6	13.1	14.8
10	5.6	8.7	12.1	6.8	8.3
11	4.7	8.8	12.3	10.0	8.9
12	8.3	14.1	16.1	10.3	12.2
13	10.3	13.4	17.6	12.1	13.4
14	11.8	17.5	18.1	12.5	14.9
15	6.4	9.6	12.1	7.6	8.9
16	4.3	6.8	11.2	8.1	7.6
17	5.7	10.2	14.1	8.9	9.7
18	6.1	12.1	13.6	7.5	9.9
19	6.7	12.1	14.6	12.1	11.4
20	4.9	7.4	9.2	6.7	7.1
21	3.2	9.6	10.9	7.7	7.9
22	4.4	9.4	12.5	8.1	8.6
23	5.2	11.1	13.1	10.5	9.9
24	6.6	9.3	10.6	8.6	8.8
25	4.7	8.9	13.6	6.8	8.5
26	4.2	8.4	11.4	6.2	7.6
27	3.7	11.3	13.2	7.5	8.9
28	6.9	11.6	12.8	8.7	10.0
29	8.2	14.3	16.4	11.6	12.6
30	3.2	7.8	9.8	6.5	6.8
control	1.8	3.0	3.0	2.4	2.5
Cetirizine	4.5	9.6	11.6	8.3	8.5
Chlorpheni ramine	11.7	37.0	32.0	22.2	25.7

Figure 31 : Percent CNS Depression of 1,4-disubstituted-*s*-triazoloquinazolines



Considering that quinazolines have been reported in the literature to possess a wide variety of biological activities, it was further planned to evaluate the synthesized compounds for certain other types of biological activities as given below:

- Anticancer activity.
- Anti HIV activity
- Antibacterial activity
- Antitubercular activity
- Analgesic activity and
- Antiinflammatory activity

B.4. Anticancer activity

Selected compounds (**1**, **4**, **IVb**, **6** and **9**) were screened for anticancer activity in drug–screening programme at the National Cancer Institute (NCI, USA).

Among the compounds tested for primary anticancer assay against a panel of 3 cell lines i.e. lung, breast and CNS cancer (Table 4), the compounds **IVb** shown 31%, 9% and 43% inhibition of growth against lung, breast and CNS cancer respectively, where as the other compounds shown less than 62% growth inhibition in all the 3 cell lines tested. Hence, the compound **IVb** was evaluated against the full panel of 60 human tumor cell lines, at a minimum of 5 concentrations at 10 fold dilutions. The results are expressed as GI_{50} values (Concentration

required to inhibit the growth of 50 % cells) at micromolar concentration. The results are presented in Table 5.

The results of anticancer activity studies (Table 5) indicate that the compound **IVb** has shown good cytotoxiciy against HL-60 (TB), leukemia (GI_{50} 2.58 μm), and strong cytotoxicity against UO-31 renal cancer (GI_{50} 0.364 μm), however it has shown weak cytotoxicity against the rest of cancer cell line tested.

Table 4 : Anticancer Activity (Percentage growth of cells)

Drug Code	Conc. (μm)	Lung Cancer NCI-H460	Breast Cancer MCF7	CNS Cancer SF-268	Activity
1	1.00	95	63	99	Inactive
4	1.00	84	89	97	Inactive
IV b	1.00	31	9	43	**Active**
6	1.00	111	83	108	Inactive
9	1.00	103	83	109	Inactive

Figure 32: Anticancer Activity (Percentage growth of cells)

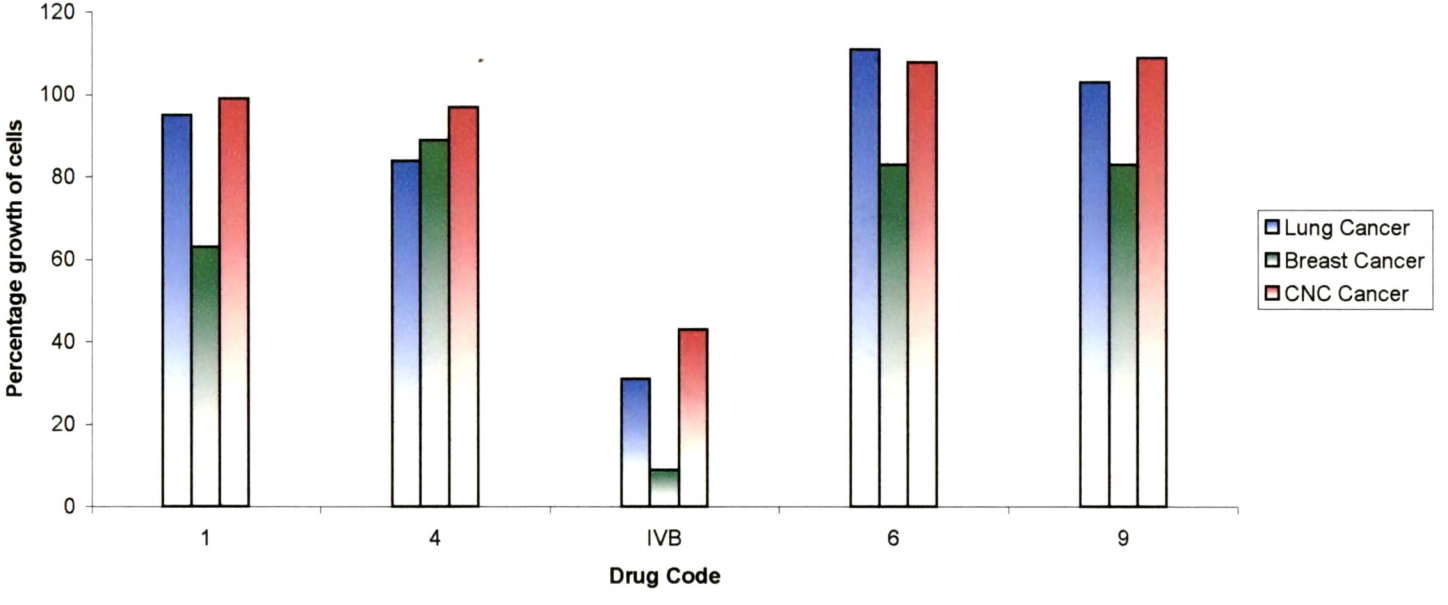


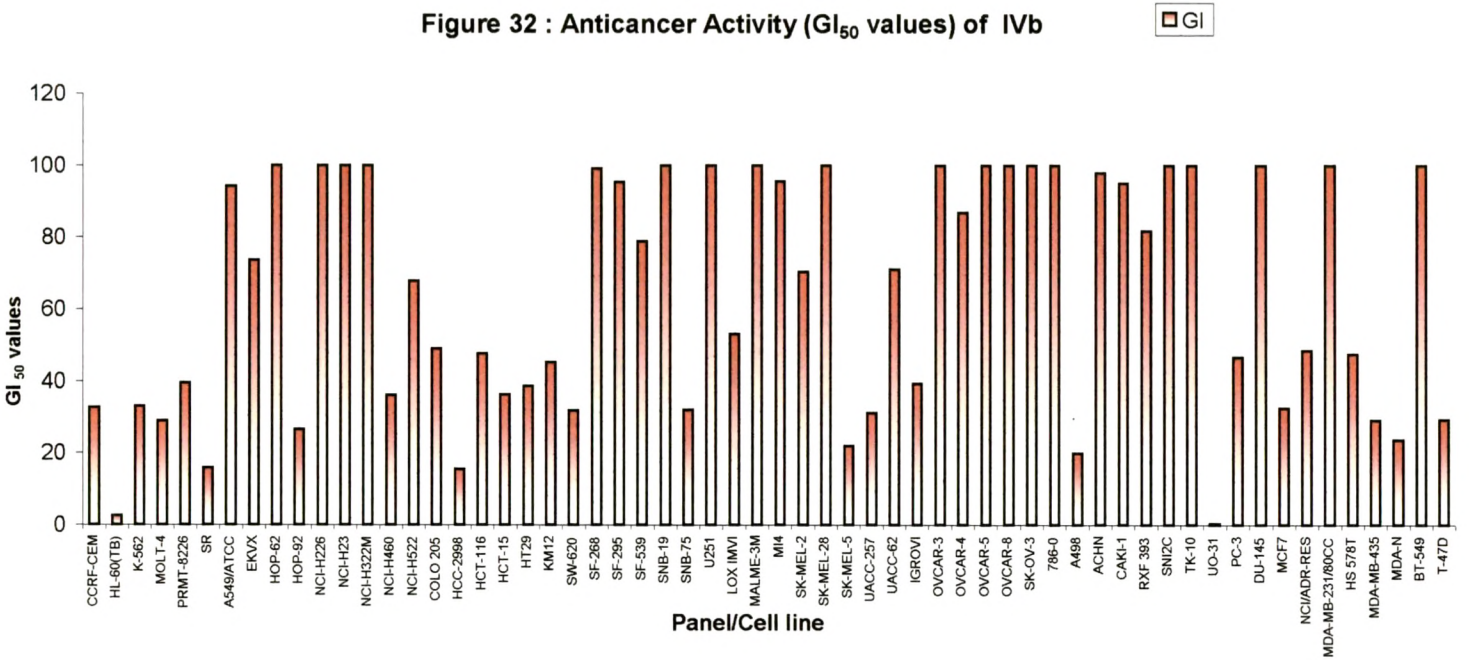
Table 5 : Anticancer Activity (GI_{50} values) of IV b

Panel/Cell Line	[a]GI_{50} (μm)
Leukemia	
CCRF-CEM	32.6
HL-60(TB)	**2.58**
K-562	33.0
MOLT-4	28.9
PRMT-8226	39.4
SR	15.8
Non-Small Cell Lung Cancer	
A549/ATCC	94.2
EKVX	73.6
HOP-62	>100.0
HOP-92	26.5
NCI-H226	>100.0
NCI-H23	>100.0
NCI-H322M	>100.0
NCI-H460	36.0
NCI-H522	67.8
Colon Cancer	
COLO 205	48.9
HCC-2998	15.6
HCT-116	47.7
HCT-15	36.2
HT29	38.6
KM12	45.3
SW-620	31.9
CNS Cancer	
SF-268	99.1
SF-295	95.4
SF-539	78.9
SNB-19	>100.0
SNB-75	32.0
U251	>100.0
Melanoma	
LOX IMVI	53.1
MALME-3M	>100.0
M14	95.5
SK-MEL-2	70.4
SK-MEL-28	>100.0
SK-MEL-5	21.9
UACC-257	31.1
UACC-62	71.0

Panel/Cell Line	[a]GI_{50} (μm)
Ovarian Cancer	
IGROVI	39.2
OVCAR-3	>100.0
OVCAR-4	86.8
OVCAR-5	>100.0
OVCAR-8	>100.0
SK-OV-3	>100.0
Renal Cancer	
786-0	>100.0
A498	19.95
ACHN	97.9
CAKI-1	95.0
RXF 393	81.8
SNI2C	>1.00.0
TK-10	>1.00.0
UO-31	**0.364**
Prostate Cancer	
PC-3	46.6
DU-145	>1.00.0
Breast Cancer	
MCF7	32.5
NCI/ADR-RES	48.4
MDA-MB-231/ATCC	>1.00.0
HS 578T	47.4
MDA-MB-435	29.1
MDA-N	23.6
BT-549	>1.00.0
T-47D	29.2

[a]I_{50} values at micromolar concentration.

Figure 32 : Anticancer Activity (GI_{50} values) of IVb



B.5. AntiHIV activity

The synthesized compounds were evaluated for their inhibitory effect of the replication of HIV-1 and HIV-2 in human MT-4 cells.

The results of anti HIV activity (Table 6), shows that the compound **Vb** exhibited maximum 113 % protection against HIV-1 (IIIB) [EC_{50} 10.1 μg/ml] and 136 % protection against HIV-2 (ROD) [EC_{50} 7.85 μg/ml] ; The compound **26** exhibited maximum 50% protection against HIV-1 (IIIB) [EC_{50} 4.94μg/ml] and 25% protection against HIV- 2 (ROD) [EC50 > 19.9 μg/ml]; The compound **Vf** exhibited maximum 23% protection against HIV-1 (IIIB) [EC_{50} >125 μg/ml] and 47% protection against HIV-2 (ROD) [EC_{50} >125 μg/ml]. While the rest of compounds exhibited mild protection at its subtoxic concentration.

Table 6 : AntiHIV Activity of test compounds.

Compound Code	Strain	EC_{50}[a] (µg/ml)	CC_{50}[b] (µg/ml)	SI[c]	Maximum Protection %
III a	IIIB	>12.3 >12.3	=12.3 =12.3	<1 <1	2 0
	ROD	>14.4	=14.4	<1	1
IV a	IIIB	>125 >12.1	>125 =12.1	X1 <1	2 0
	ROD	>15.9	=15.9	<1	0
IV b	IIIB	>33.9 >23.5	=33.9 =23.5	<1 <1	0 0
	ROD	>66.6	=66.6	<1	0
IV f	IIIB	>125 >75.1	>125 =75.1	X1 <1	1 2
	ROD	>125	>125	X1	9
V a	IIIB	>13.7 >12.4	=13.7 =12.4	<1 <1	3 2
	ROD	>10.5	=10.5	<1	0
V b	IIIB	=10.1 >64.6	=77.6 =64.6	=8 <1	**113** **33**
	ROD	=7.85 =19.4	=85.2 =63	=11 =3	**136** **59**
V c	IIIB	>125 >125	>125 >125	X1 X1	3 4
	ROD	>125	>125	X1	0
V d	IIIB	>12.5 >12	=12.5 =12	<1 <1	13 2
	ROD	>13.9	=13.9	<1	0
V f	IIIB	>125 >125	>125 >125	X1 X1	23 3
	ROD	>125 >125	>125 >125	X1 X1	47 16
1	IIIB	>69.1 >65	=69.1 =65	<1 <1	2 0

	ROD	>72.3	=72.3	<1	0
2	IIIB	>125	>125	X1	2
		>125	>125	X1	0
	ROD	>125	>125	X1	0
3	IIIB	>125	>125	X1	2
		>103	=103	<1	0
	ROD	>125	>125	X1	1
4	IIIB	>13	=13	<1	2
		>6.79	=6.79	<1	1
	ROD	>10.2	=10.2	<1	0
5	IIIB	>101	=101	<1	2
		>117	=117	<1	0
	ROD	>125	>125	X1	0
6	IIIB	>125	>125	X1	1
		>125	>125	X1	2
	ROD	>125	>125	X1	0
7	IIIB	>13.6	=13.6	<1	2
		>25	>25	X1	0
	ROD	>25	>25	X1	0
8	IIIB	>125	>125	X1	2
		>14.1	=14.1	<1	0
	ROD	>11.5	=11.5	<1	6
9	IIIB	>86.8	=86.8	<1	1
		>125	>125	X1	0
	ROD	>125	>125	X1	0
10	IIIB	>125	=125	X1	0
		>125	=125	X1	1
	ROD	>125	>125	X1	0
11	IIIB	>125	>125	X1	1
		>125	>125	X1	0
	ROD	>125	>125	X1	0
12	IIIB	>23.4	=23.4	<1	3
		>12.7	=12.7	<1	1
	ROD	>19.8	=19.8	<1	0
13	IIIB	>125	>125	X1	2
		>125	>125	X1	1
	ROD	>125	>125	X1	0
14	IIIB	>125	>125	X1	1
		>125	>125	X1	0
	ROD	>125	>125	X1	4

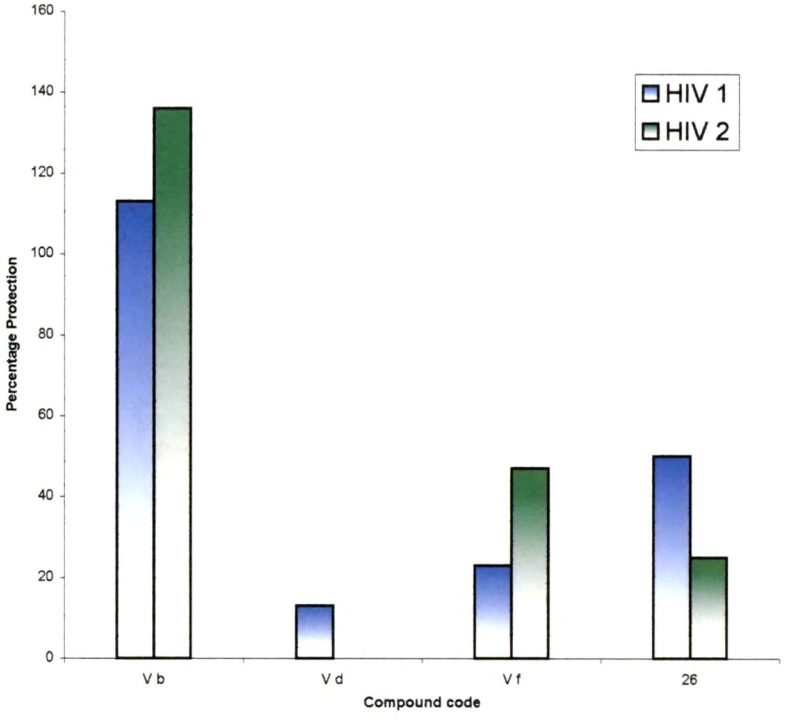
15	IIIB	>84 >125	=84 >125	<1 X1	2 2
	ROD	>125	>125	X1	0
16	IIIB	>125 >125	>125 >125	X1 X1	1 0
	ROD	>125	>125	X1	0
17	IIIB	>125 >125	>125 >125	X1 X1	1 1
	ROD	>125	>125	X1	0
19	IIIB	>125 >95.6	>125 =95.6	X1 <1	2 0
	ROD	>125	>125	X1	4
20	IIIB	>104 >98.8	=104 =98.8	<1 <1	3 1
	ROD	>125	>125	X1	0
21	IIIB	>108 >104	=108 =104	<1 <1	4 0
	ROD	>125	>125	X1	1
22	IIIB	>125 >107	>125 =107	X1 <1	3 5
	ROD	>125	>125	X1	0
24	IIIB	>14.8 >12.8	=14.8 =12.8	<1 <1	2 1
	ROD	>13.5	=13.5	<1	0
26	IIIB	>15 =4.94	=15 >25	<1 >5	9 50
	ROD	>19.9 >18.2	=19.9 =18.2	<1 <1	25 0
27	IIIB	>125 >125	>125 >125	X1 X1	3 0
	ROD	>125	>125	X1	1

[a] Effective concentration of compound, achieving 50 % protection of MT-4 cells against the cytopathic effect of HIV.

[b] Cytotoxic concentration of compound required to reduce the viability of normal uninfected MT-4 cells by 50 % .

[c] Selective index or ratio of CC_{50} to EC_{50}.

Figure 34: Anti HIV Activity (Percentage Protection) of Test Compounds



B.6. Antibacterial activity

The antibacterial activity of title compounds was studied by agar cup-plate method, the results are expressed in the terms of zone of inhibition in mm and presented in Table 7.

The results of antibacterial activity depicted in Table 7 indicate that none of the compounds was equipotent to ciprofloxacin, a standard drug employed in the investigation. Almost all the compounds exhibited mild antibacterial activity against *S.typhi*, however the test compounds did not inhibits the growth of *E.coli* (except **25** and **28**) at the concentration tested, while the rest of compounds showed mild to moderate antibacterial activity against some of the bacteria tested.

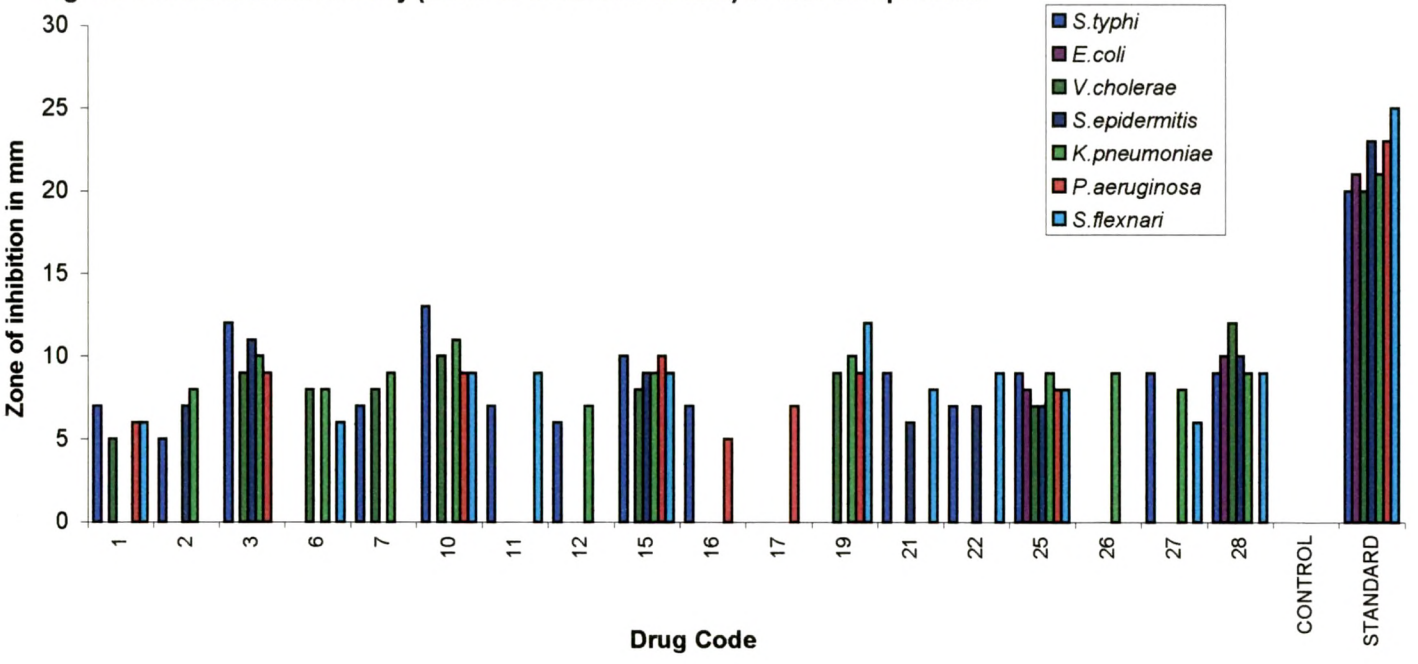
Table 7 : Antibacterial Activity (Zone of inhibition in mm) of Test compounds

Drug Code	*S. typhi.*	*E.Coli*	*V. cholerae*	*S.Epidermitis*	*K.Pneumoniae*	*P.aeruginosa*	*S.flexnari*
1	07	-	05	-	-	O6	06
2	05	-	-	07	08	-	-
3	12	-	09	11	10	09	-
6	-	-	08	-	08	-	06
7	07	-	08	-	09	-	-
10	13	-	10	-	11	09	09
11	07	-	-	-	-	-	09
12	06	-	-	-	07	-	-
15	10	-	08	09	09	10	09
16	07	-	-	-	-	05	-
17	-	-	-	-	-	07	-
19	-	-	09	-	10	09	12
21	09	-	-	06	-	-	08
22	07	-	-	07	-	-	09
25	09	08	07	07	09	08	08
26	-	-	-	-	09	-	-
27	09	-	-	-	08	-	06
28	09	10	12	10	09	-	09
Control	-	-	-	-	-	-	-
Standard	20	21	20	23	21	23	25

(-) Indicates no activity.

Test concentration : 200 µg/ml, Standard Ciprofloxacin : 10 µg/ml

Fig 35 : Antibacterial Activity (Zone of inhibition in mm) of test compounds



B.7. Antitubercular activity

The synthesized compounds were screened for their *in vitro* antimycobacterial activity against *Mycobacterium tuberculosis* strain H_{37} Rv at the Tuberculosis Antimicrobial Acquisition & Coordinating Facility (TAACF, USA). The results are expressed in terms of Minimum Inhibitory Concentration (MIC) and percentage inhibition of growth.

The results of antimycobacterial activity activity depicted in Table 8 indicate that the test compounds inhibited the growth of mycobaterium in varying degree. Compound **27** was found to be the most potent agent and it showed 72% inhibition ; compound **IVd** exhibited 61% inhibition ; compound **IVc** exhibited 60% inhibition while, the rest of compounds exhibited less than 60% inhibition. The test compounds exhibited their antimycobacterial activity at the concentration of 6.25 μg/ml (MIC 6.25 μg/ml). Some of the comounds did not exhibited antimycobacterial activity at this concentration.

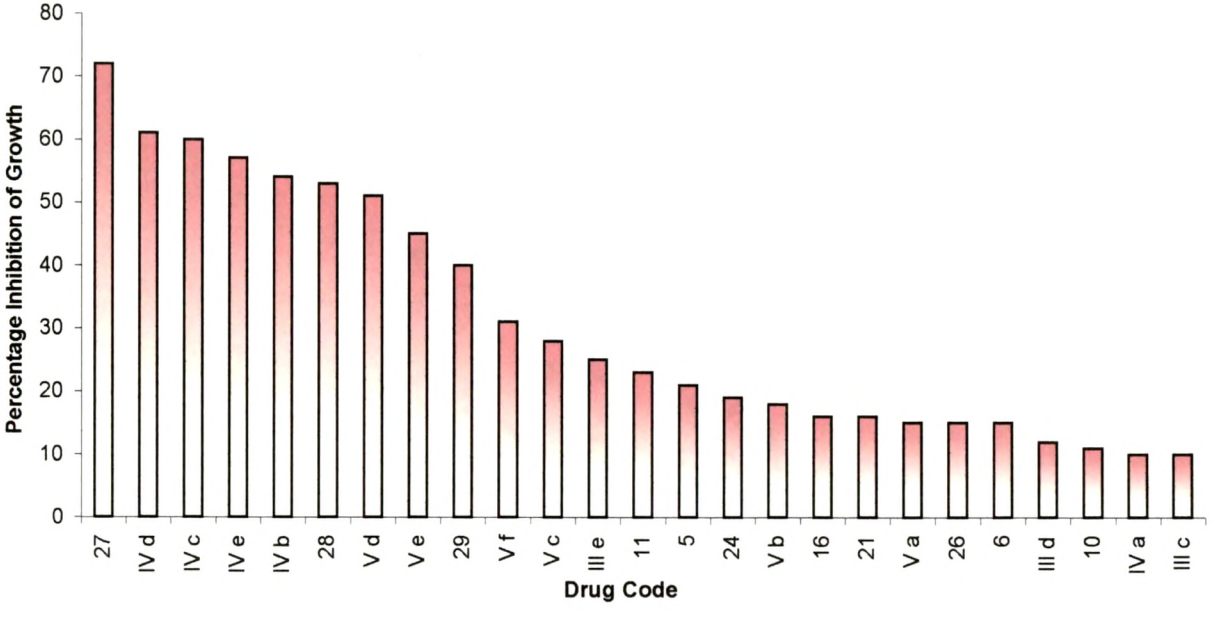
Table 8 : Antitubercular Activity of Test Compounds.

Drug Code	MIC (µg/ml)	Percent Inhibition
27	>6.25	72
IV d	>6.25	61
IV c	>6.25	60
IV e	>6.25	57
IV b	>6.25	54
28	>6.25	53
V d	>6.25	51
V e	>6.25	45
29	>6.25	40
V f	>6.25	31
V c	>6.25	28
III e	>6.25	25
11	>6.25	23
5	>6.25	21
24	>6.25	19
V b	>6.25	18
16	>6.25	16
21	>6.25	16
V a	>6.25	15
26	>6.25	15
6	>6.25	15

III d	>6.25	12
10	>6.25	11
IV a	>6.25	10
III c	>6.25	10
1	>6.25	09
19	>6.25	09
3	>6.25	09
III b	>6.25	07
9	>6.25	06
14	>6.25	05
20	>6.25	05
III f	>6.25	03
IV f	>6.25	01
18	>6.25	01
4	>6.25	00
30	>6.25	00
12	>6.25	00
15	>6.25	00
17	>6.25	00
7	>6.25	00
22	>6.25	00
25	>6.25	00
8	>6.25	00

13	>6.25	00
23	>6.25	00

Figure 36 : Antitubercular Activity of Test Compounds



B.8. Analgesic activity

The analgesic activity of selected title compounds was tested in mice by tail-flick technique and the results are recorded in Table 9.

Table 9 depicting the results of analgesic activity of title compounds indicate that the test compounds exhibited moderate activity while none of the test compounds was equipotent to pentazocine, a standard drug employed in the investigation.

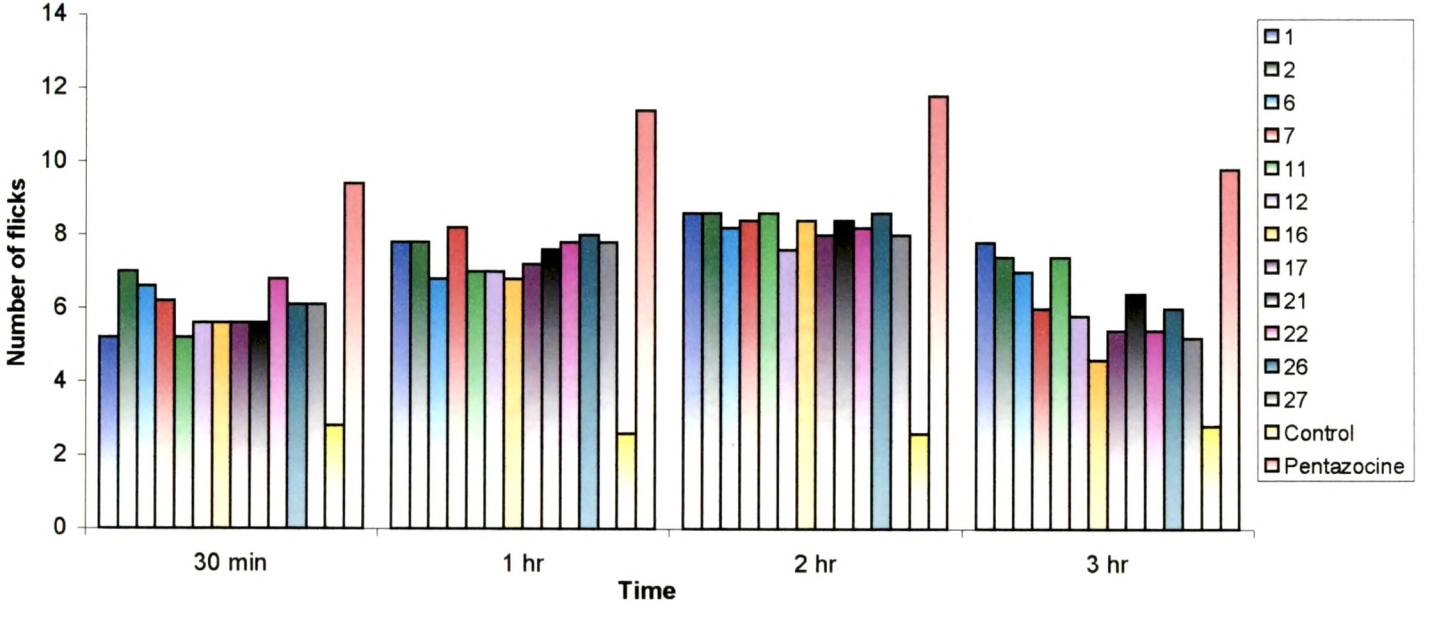
Table 9 : Analgesic activity (Number of flicks ± SEM) of Test compounds (Tail- flick technique)

Drug Code	Mean basal Reaction time(In sec.)	Mean reaction time after Drug treatment (In sec.)			
		30 min	1 h	2 h	3 h
1	2.25	5.2±0.37**	7.8± 0.58**	8.6± 0.24**	7.8± 0.58**
2	2.85	7.0±0.32**	7.8±0.37**	8.6±0.51**	7.4±0.51**
6	3.40	6.6±0.57**	6.8±0.53**	8.2±0.62**	7.0±0.32**
7	2.30	6.2±0.58**	8.2±0.37**	8.4±0.70**	6.0±0.55**
11	3.40	5.2±0.20**	7.0±0.32**	8.6±0.51**	7.4±1.03**
12	2.75	5.6±0.24**	7.0±0.45**	7.6±0.40**	5.8±0.37**
16	2.9	5.6±0.24**	6.8±0.49**	8.4±0.51**	4.6±0.51*
17	3.15	5.6±0.28**	7.2±0.58**	8.0±0.45**	5.4±0.24**
21	2.15	5.6±0.40**	7.6±0.68**	8.4±0.51**	6.4±0.75**
22	2.55	6.8±0.49**	7.8±0.71**	8.2±0.58**	5.4±0.24**
26	2.55	6.1±0.38**	8.0±0.32**	8.6±0.24**	6.0±0.45**
27	2.55	6.1±0.47**	7.8±0.37**	8.0±0.32**	5.2±0.62**
Control	2.30	2.8±0.37	2.6±0.40	2.6±0.40	2.8±0.37
Pentazocine	2.35	9.4±0.40**	11.4±0.40**	11.8±0.20**	9.8±0.37**

Each value represents the mean ± SEM (n=6). Significance levels *p<0.2 and **p<0.001 as compared with the respective control.

Dose of test compounds : 10 mg/kg

Figure 37 : Analgesic activity (Number of flicks) of test compounds



B.9. Antiinflammatory activity

The antiinflammatory activity of selected title compounds was carried in rats by carrageenan-induced rat paw oedema method and the results are recorded in Table 10.

The results of antiinflammatory activity (Table 10) indicate that all test compounds exhibited mild to moderate activity, however none of the test compounds showed equipotent activity with diclofenac sodium, a standard drug employed in the investigation.

Table 10 : Antiinflammatory activity (Paw volume ±SEM) of test compounds (Carrageenan induced Rat paw oedema method)

Drug Code	Paw volume in cm						
	0 min.	30 min	1 h	2 h	3 h	4 h	5 h
1	0.56	0.68±0.013*	0.71±0.011*	0.78±0.009***	0.81±0.011***	0.82±0.014***	0.80±0.013*
2	0.55	0.67±0.012*	0.68±0.010**	0.77±0.010****	0.77±0.010****	0.80±0.010****	0.75±0.010**
6	0.60	0.73±0.010*	0.76±0.010*	0.81±0.010**	0.83±0.010**	0.85±0.010**	0.79±0.010*
7	0.61	0.72±0.010*	0.75±0.010*	0.85±0.010*	0.86±0.010*	0.90±0.010*	0.84±0.010*
11	0.56	0.69±0.010*	0.71±0.010*	0.82±0.010**	0.83±0.020**	0.83±0.020**	0.76±0.020*
12	0.54	0.67±0.010*	0.70±0.010*	0.82±0.010**	0.84±0.010**	0.85±0.010**	0.77±0.010*
16	0.62	0.74±0.020	0.76±0.020*	0.85±0.020*	0.86±0.020*	0.87±0.020*	0.81±0.020*
17	0.56	0.67±0.010*	0.71±0.004*	0.79±0.010***	0.79±0.004***	0.81±0.010***	0.78±0.010*
21	0.58	0.71±0.010*	0.73±0.010*	0.82±0.010**	0.85±0.010**	0.86±0.010**	0.80±0.010*
22	0.55	0.69±0.010*	0.72±0.010*	0.83±0.010**	0.84±0.010**	0.88±0.010*	0.78±0.010*
26	0.58	0.70±0.010*	0.73±0.010*	0.81±0.020**	0.87±0.020*	0.91±0.020*	0.81±0.010*
27	0.58	0.70±0.010*	0.74±0.010*	0.82±0.010**	0.85±0.010**	0.86±0.010**	0.80±0.010*
Control	0.60	0.74±0.048	0.78±0.042	0.92±0.045	0.94±0.045	0.95±0.043	0.85±0.041
Diclofenac sodium	0.52	0.63±0.010**	0.64±0.010***	0.69±0.010*****	0.60±0.010*****	0.74±0.010*****	0.70±0.010****

Each value represents the mean ± SEM (n=5). Significance levels *p<0.5, **p<0.1, ***p<0.05, ****p<0.02 and *****p<0.01 as compared with the respective control. Dose of test compounds : 10 mg/kg

Figure 38 : Anti inflammatory activity (Paw volume) of test compounds