RESULTS

RESULTS
I. In vivo Experiments

(A) Chronic studies

I. Pharmacological results

1. Effect of moxonidine on arterial blood pressure in DOCA-salt hypertensive hypercholesterolemic rats.

- **Systolic arterial blood pressure:**

Intramuscular injection of rats of group II with DOCA (50 mg/kg / week) suspended in olive oil and drinking saline 1 % NaCl and 2% cholesterol for 6 weeks resulted in a significant rise ($P < 0.01$) of systolic blood pressure from 121.67 ± 4.77 mm Hg in control group (group I) to 165.83 ± 5.54 mm Hg. Treatment of hypertensive hypercholesterolemic rats of group III with moxonidine in a dose of 0.5 mg / kg, I.P for 2 weeks resulted in a significant ($P < 0.001$) decrease in systolic blood pressure to 96.67 ± 1.67 mm Hg (Table 1, Figs.1,4).

- **Mean arterial blood pressure:**

In control group (group I) the mean blood pressure was 83.33 ± 2.11 mm Hg increased significantly ($P < 0.01$) to 117.5 ± 2.81 mm Hg in group II. Moxonidine treatment of group III decreased the mean blood pressure significantly ($P < 0.001$) to 73.33 ± 1.67 mm Hg compared to group II (Table 1, Figs. 2, 4).

2. Effect of moxonidine on heart rate of DOCA-salt hypertensive hypercholesterolemic rats

The mean value of heart rate in control group (group I) was 205 ± 11.95 beat/minute decreased insignificantly ($P > 0.05$) to 200.5 ± 13.0 beat/minute in group II. Treatment of group III with moxonidine (0.5 mg/kg, I.P. for 2 weeks) significantly ($P < 0.05$) reduced heart rate to 153.33 ± 13.95 beat / minute compared to group II (Table 1, Fig. 3).

Table (1): Effect of moxonidine treatment on arterial blood pressure and heart rate of hypertensive hypercholesterolemic rats.

Groups / Parameters	Group I	Group II	Group III
Systolic blood pressure (mm Hg)	121.67 ± 4.77	165.83 ± 5.54 $P_1 < 0.01$*	96.67 ± 1.67 $P_2 < 0.001$*
Mean blood pressure (mm Hg)	83.33 ± 2.11	117.5 ± 2.81 $P_1 < 0.01$*	73.33 ± 1.67 $P_2 < 0.001$*
Heart rate (beat / min.)	205 ± 11.95	200.5 ± 13.0 $P_1 > 0.05$	153.33 ± 13.95 $P_2 < 0.05$*

Data represented as mean ± SEM of six experiments

Group I : Control group

Group II : DOCA-salt hypertensive hypercholesterolemic group.

Group III : DOCA-salt hypertensive hypercholestrolemic moxonidine (0.5 mg/kg I.P. for 2 weeks) treated group.

P_1: Comparing results of group II with that of group I

P_2: Comparing results of group III with that of group II

* Significant at $P < 0.05$.

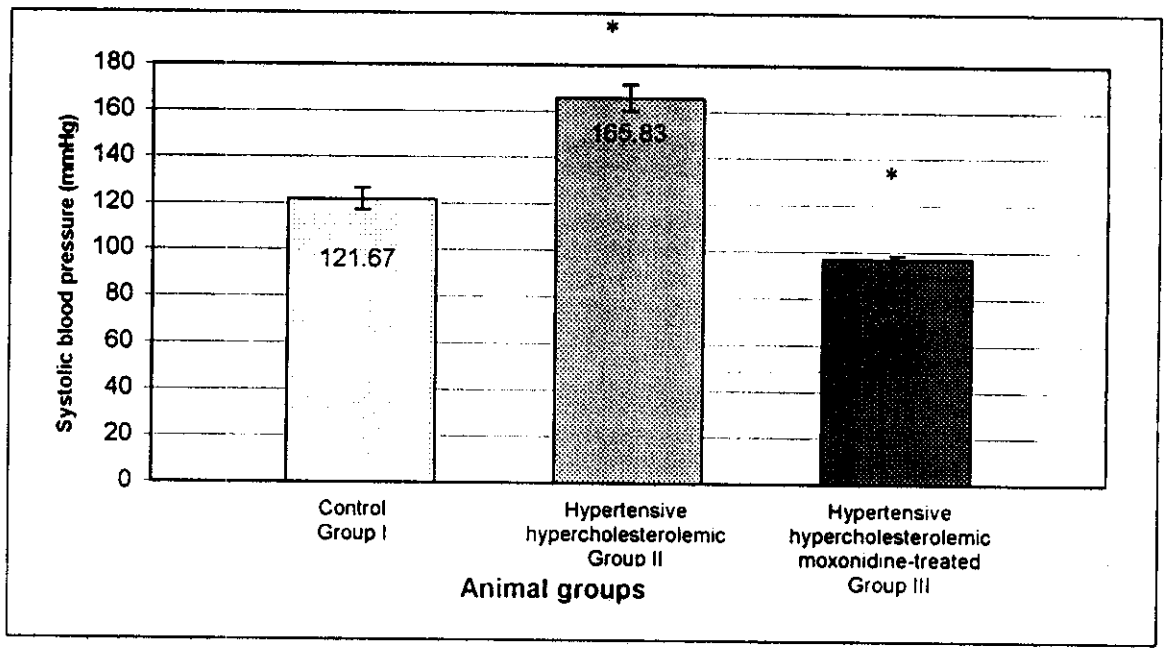


Figure (1): Histogram showing systolic arterial blood presssure in various groups.

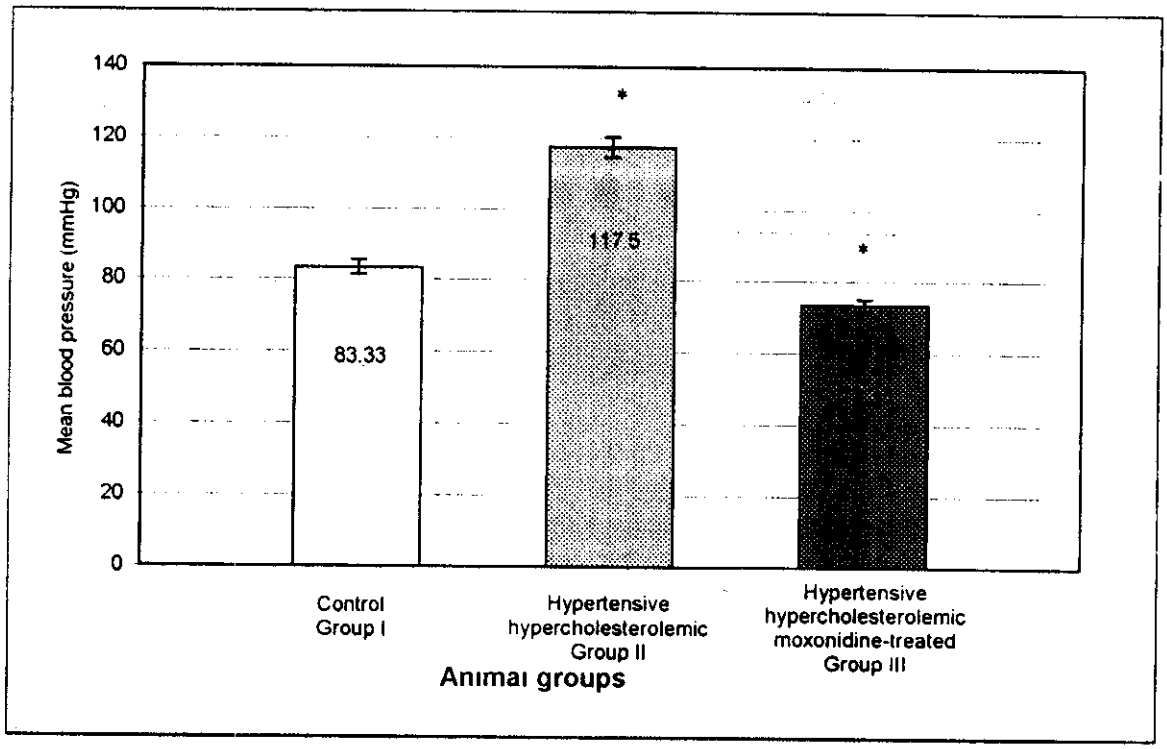


Figure (2): Histogram showing mean arterial blood presssure in various groups

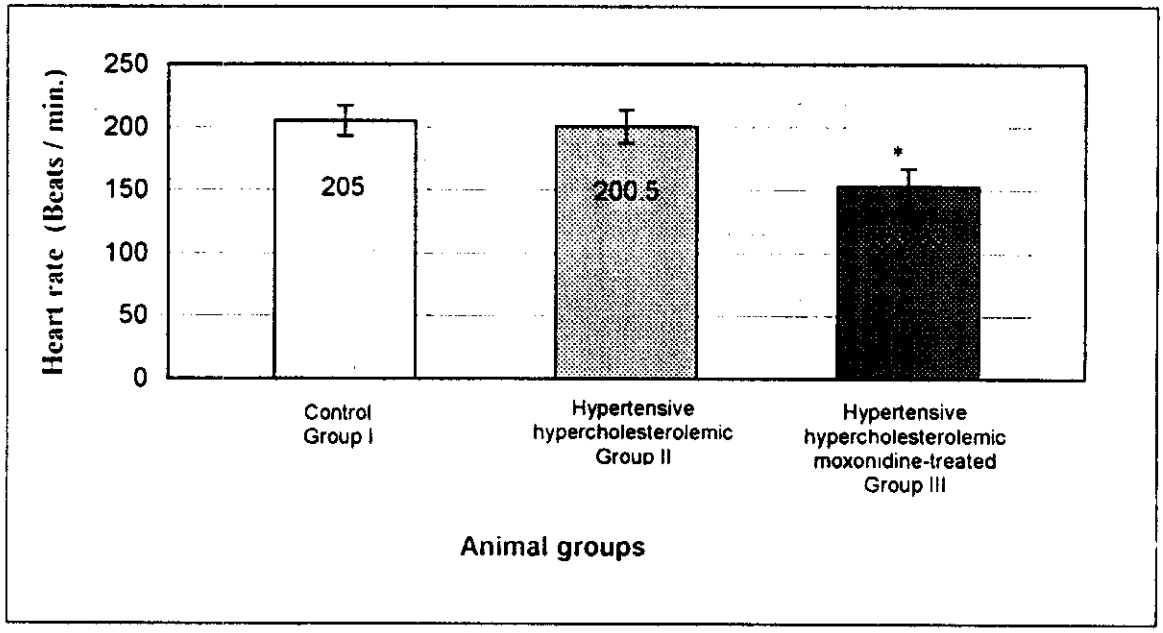


Figure (3): A histogram showing heart rate in various groups

(a)

(b)

(c)

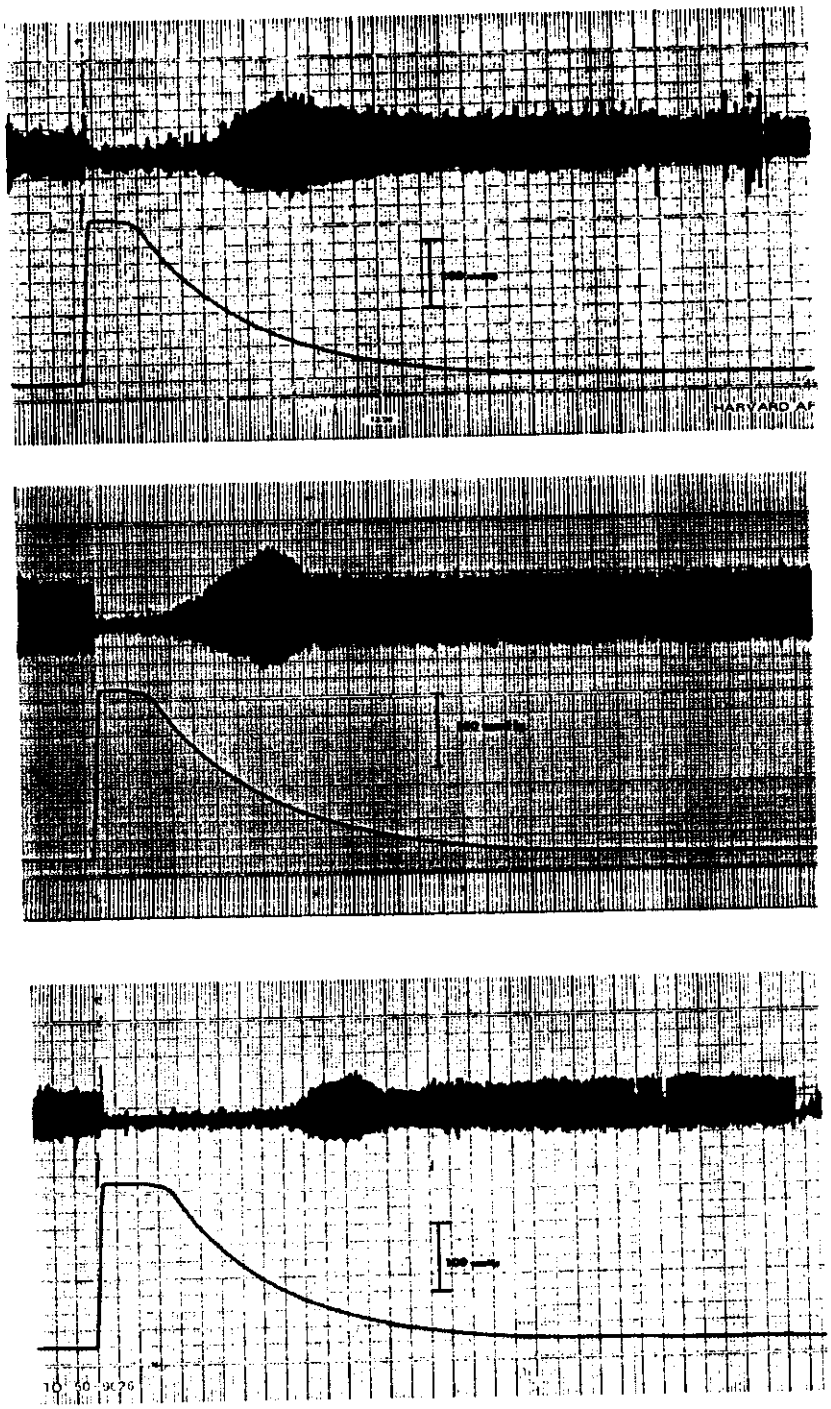


Fig (4) Diagrams showing arterial blood pressure in various groups.

(a) Control group (group I).

(b) DOCA -salt hypertensive hypercholesterolemic group (group II).

(c) Hypertensive hypercholesterolemic moxonidine -treated group (group III).

* Upper trace represents pulse blood flow.
* lower trace represents cuff pressure.

The systolic blood pressure measured at the start of pulsation and referenced to the pressure curve .

The mean blood pressure measured at stability of pulsation and refrenced to the the pressure curve .

3. Effect of moxonidine on renal and hepatic haemodynamics in hypertensive hypercholesterolemic rats:

- ***Renal hemodynamic parameters***

In control group (group I) the renal blood flow velocity was 4.22 ± 0.32 cm/s and decreased significantly ($P < 0.05$) in non-treated hypertensive hypercholesterolemic rats (group II) to 2.53 ± 0.55 cm/s. Treatment of hypertensive hypercholesterolemic rats (group III) with moxonidine (0.5 mg/kg. I.P) for 2 weeks increased renal blood flow velocity significantly ($P < 0.05$) to 3.8 ± 0.1 cm/s compared to group II (Table 2, Figs. 7, 9).

The value of renal artery resistance parameter in group I and group II were 0.97 ± 0.02 and 0.96 ± 0.01 respectively. There was no statistical significant change among the two groups. Meanwhile, the renal artery resistance parameter was significantly ($P < 0.01$) reduced to 0.85 ± 0.02 in moxonidine-treated group (group III) compared to group II (Table 2, Figs. 8,9).

In addition, the pulsatility index in control group (group I) was 2.83 ± 0.73 increased significantly ($P < 0.05$) to 6.63 ± 0.91 in hypertensive hypercholesterolemic non-treated rats (group II). Moxonidine treatment of group III reduced pulsatility index significantly ($P < 0.01$) to 1.503 ± 0.13 compared to group II (Table 2, Figs. 8, 9).

In hypertensive hypercholesterolemic rats (group II) the maximum systole in the renal artery was increased significantly ($P<0.01$) from a mean of 28.57 ± 2.58 cm/s in control group (group I) to 64.5 ± 3.51 cm/s. In moxonidine-treated group (group III), the maximum systole decreased

significantly ($P < 0.05$) to 21.57 ± 2.36 cm /s compared to group II (Table 2, Figs. 5, 9).

Moreover, the minimum systole increased significantly ($P < 0.05$) from 12.2 ± 2.09 cm/s in group I to 33.22 ± 5.83 cm/s in group II and decreased significantly ($P < 0.01$) in moxonidine-treated group (group III) to be 7.7 ± 0.93 cm/s compared to group II (Table 2, Figs. 5, 9).

On the other hand, the maximum and minimum diastole were 6.13 ± 0.75 and 2.57 ± 0.95 cm/s in group I insignificantly ($P > 0.05$) changed in group II to be 6.47 ± 0.54 and 3.1 ± 0.96 cm/s. Furthermore, in group III moxonidine treatment (0.5 mg/kg. I.P) for 2 weeks produced insignificant decrease ($P > 0.05$) in maximum and minimum diastole to be 5.92 ± 0.95 and 2.22 ± 0.43 cm/s respectively compared to group II (Table 2, Figs. 6, 9).

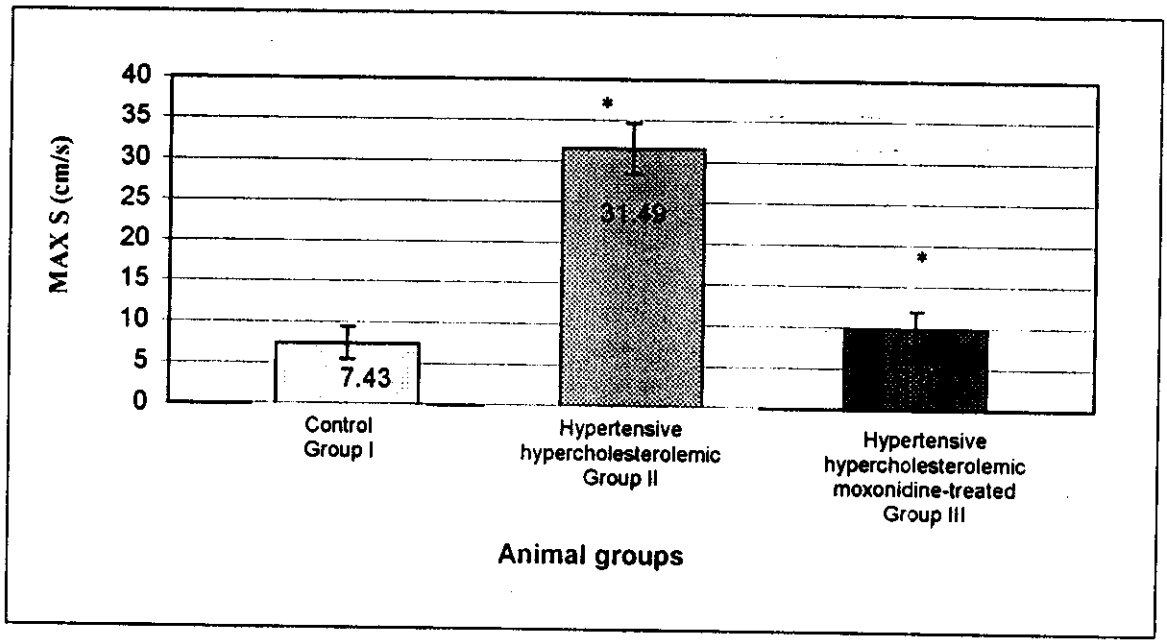
The systole / diastole ratio was 7.43 ± 1.95 in group I increased significantly ($P < 0.01$) to 31.49 ± 3.09 in group II and decreased significantly ($P < 0.01$) to 9.87 ± 2.05 in moxonidine-treated rats (group III) (Table 2, Figs. 7, 9).

Table (2): Effect of moxonidine treatment on renal hemodynamic parameters of hypertensive hypercholesterolemic rats.

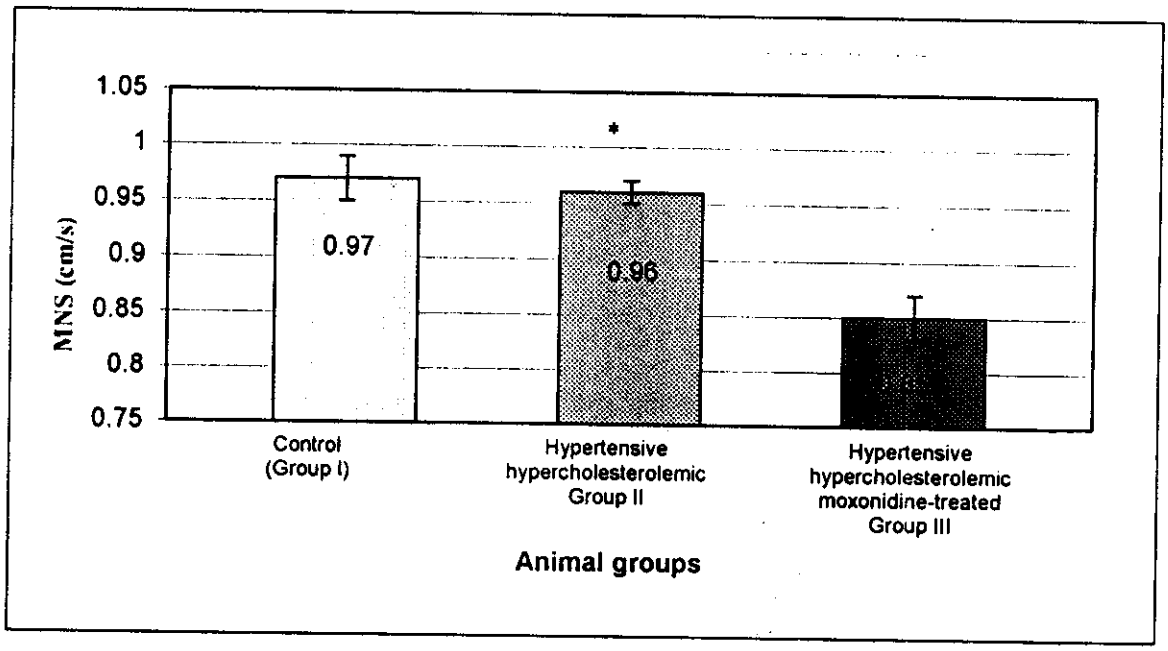
Parameters / Groups	MAX S cm/s	MN S cm/s	MAX D cm/s	MN D Cm/s	MN V Cm/s	S/D	RP	PI
Group I	28.57 ± 2.58	12.2 ± 2.09	6.13 ± 0.75	2.57 ± 0.95	4.22 ± 0.32	7.43 ± 1.95	0.97 ± 0.02	2.83 ± 0.73
Group II	64.5 ± 3.51	33.22 ± 5.83	6.47 ± 0.54	3.10 ± 0.96	2.53 ± 0.55	31.49 ± 3.09	0.96 ± 0.01	6.63 ± 0.91
P_1	<0.01*	<0.05*	>0.05	>0.05	<0.05*	<0.01*	>0.05	<0.05*
Group III	21.57 ± 2.36	7.7 ± 0.93	5.92 ± 0.95	2.22 ± 0.43	3.8 ± 0.11	9.87 ± 2.05	0.85 ± 0.02	1.503 ± 0.13
P_2	<0.05*	<0.01*	> 0.05	>0.05	<0.05*	<0.01*	<0.01*	<0.01*

Data represented as mean ± SEM of six experiments

Group I : Control group
Group II : DOCA-salt hypertensive hypercholesterolemic group.
Group III :DOCA-salt hypertensive hypercholestrolemic moxonidine (0.5 mg/kg, I.P. for 2 weeks) treated group.
P_1: Comparing results of group II with that of group I
P_2: Comparing results of group III with that of group II
* Significant at $P < 0.05$
MAX S: Maximum systole, **MAX D:** Maximum diastole, **MN S:** Minimum systole, **MN D:** Minimum diastole, **MN V:** Mean blood flow velocity, **S/D:** Systole/diastole ratio, **RP:** Resistance parameter in the renal artery, **PI:** Pulsatility index.

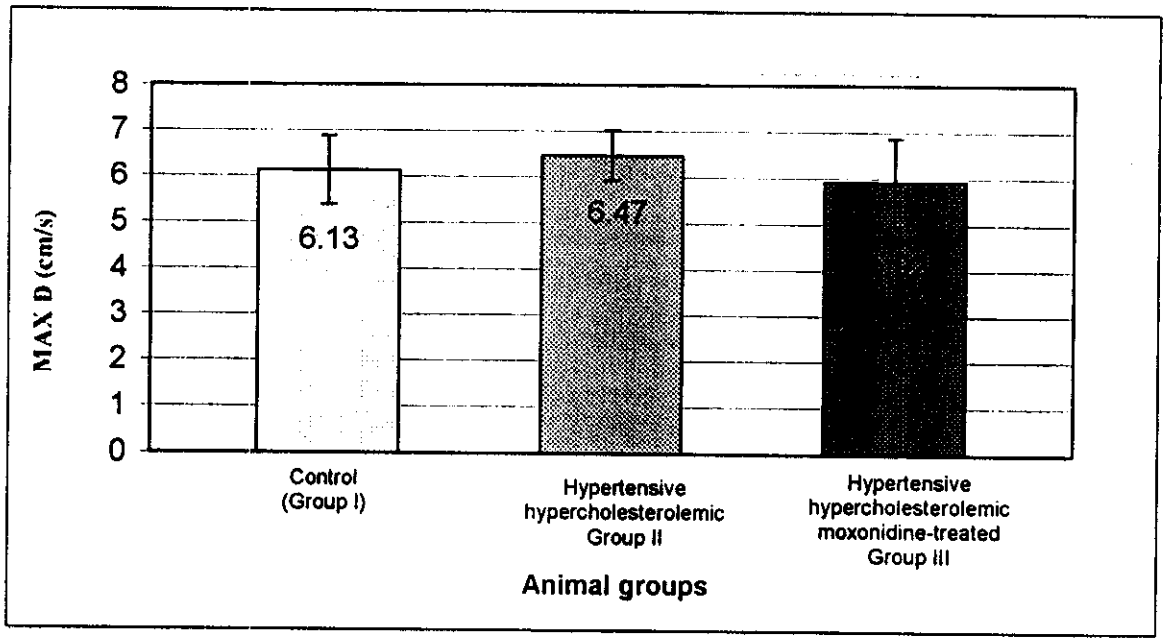


(Maximum Systole)

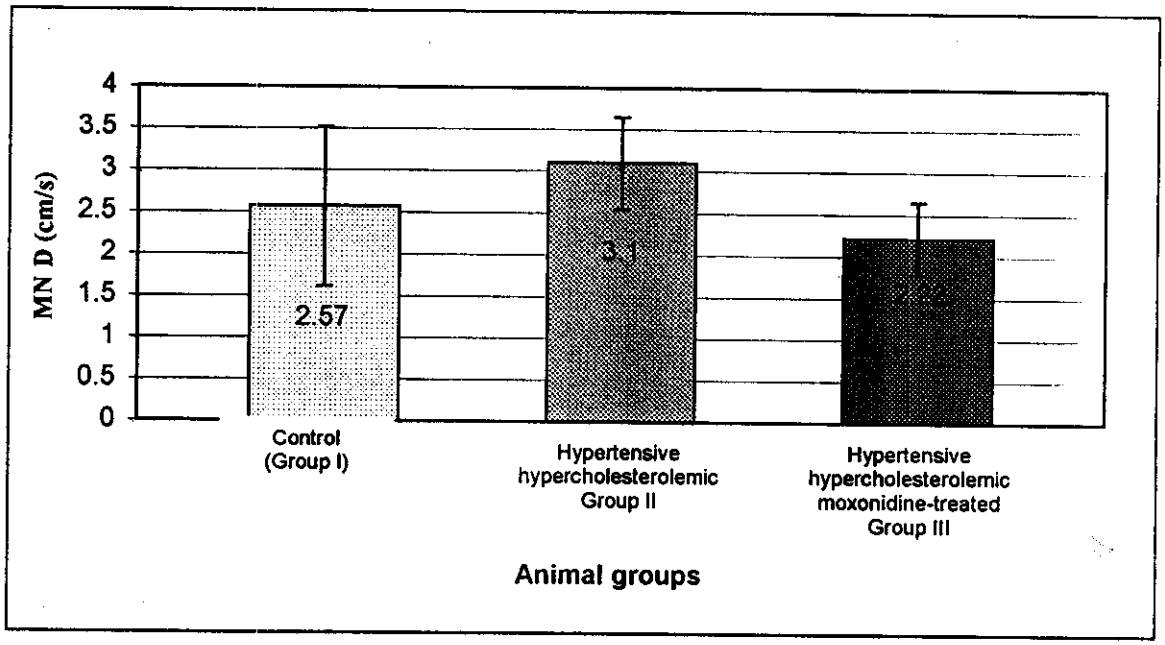


(Minimum Systole)

Figure (5): Histogram showing renal hemodynamics in various groups.

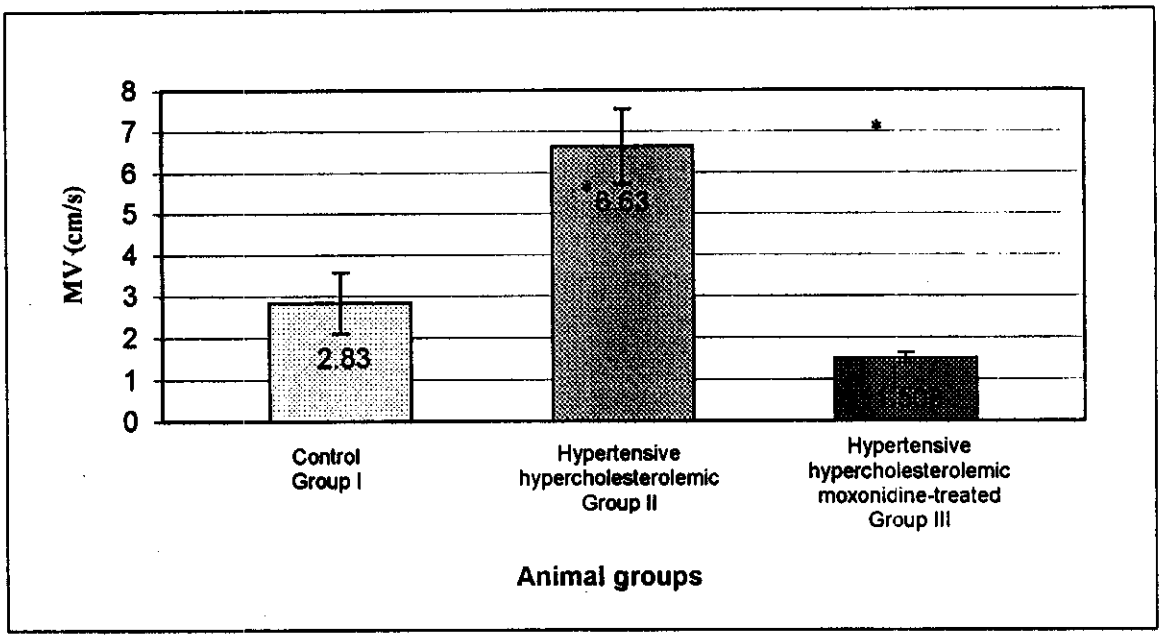


(Maximum Diastole)

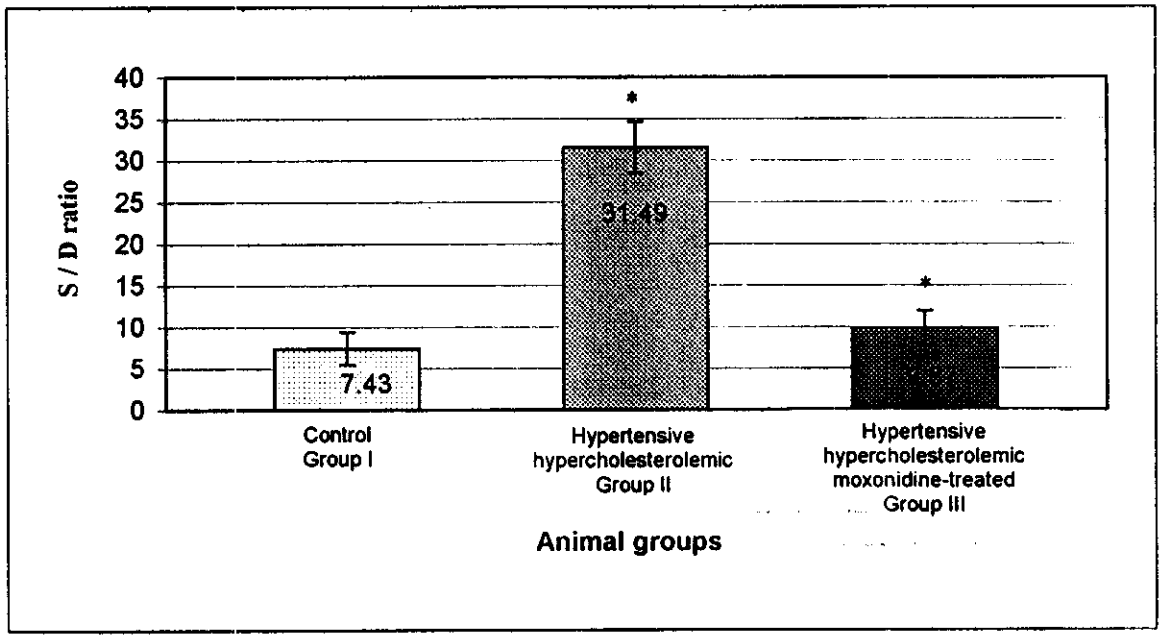


(Minimum Diastole)

Figure (6): Histogram showing renal hemodynamics in various groups.

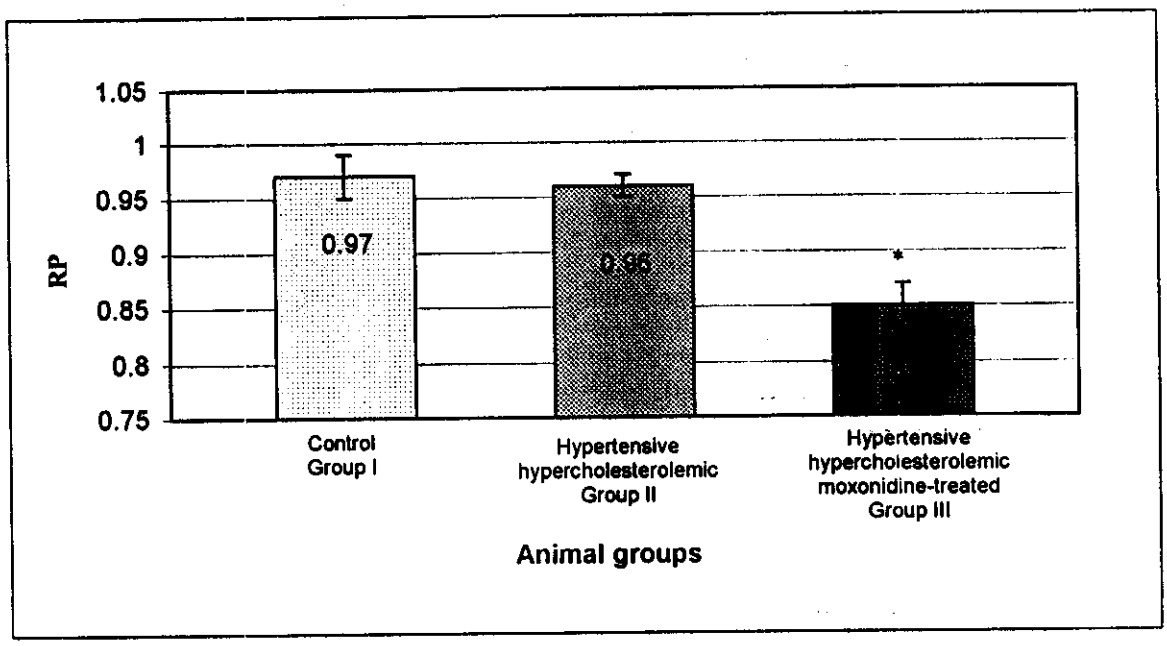


(Mean flow velocity)

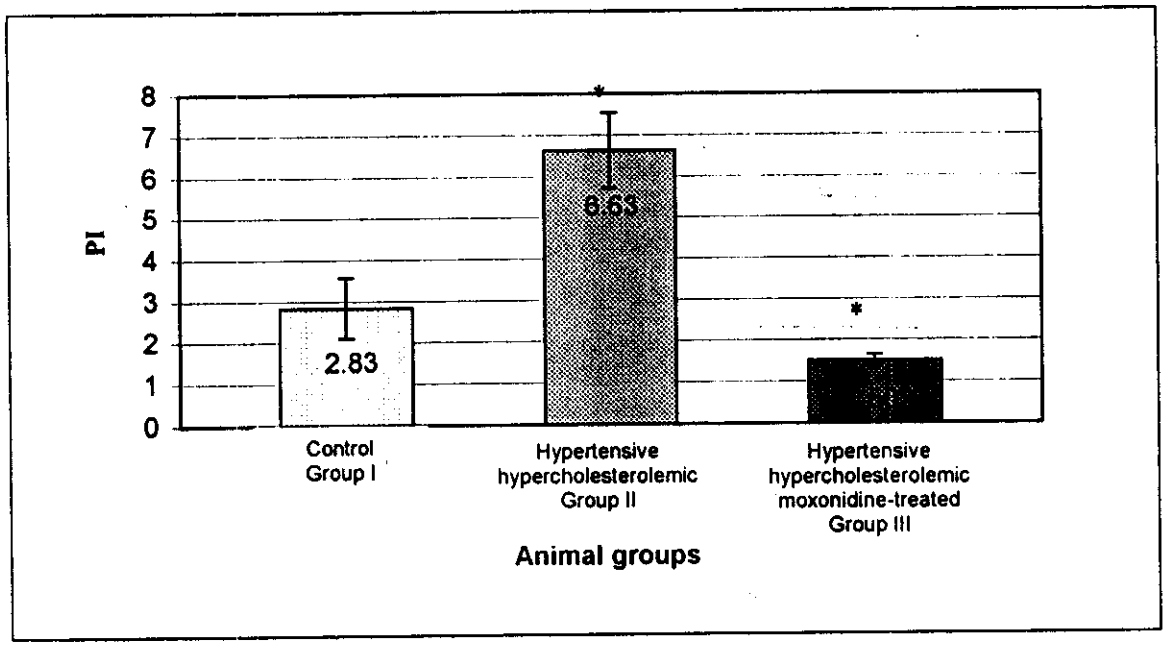


(Systole /Diastole ratio)

Figure (7): Histogram showing renal hemodynamics in various groups.



(Resistance Parameter)



(Pulsitility Index)

Figure (8): Histogram showing renal hemodynamics in various groups.

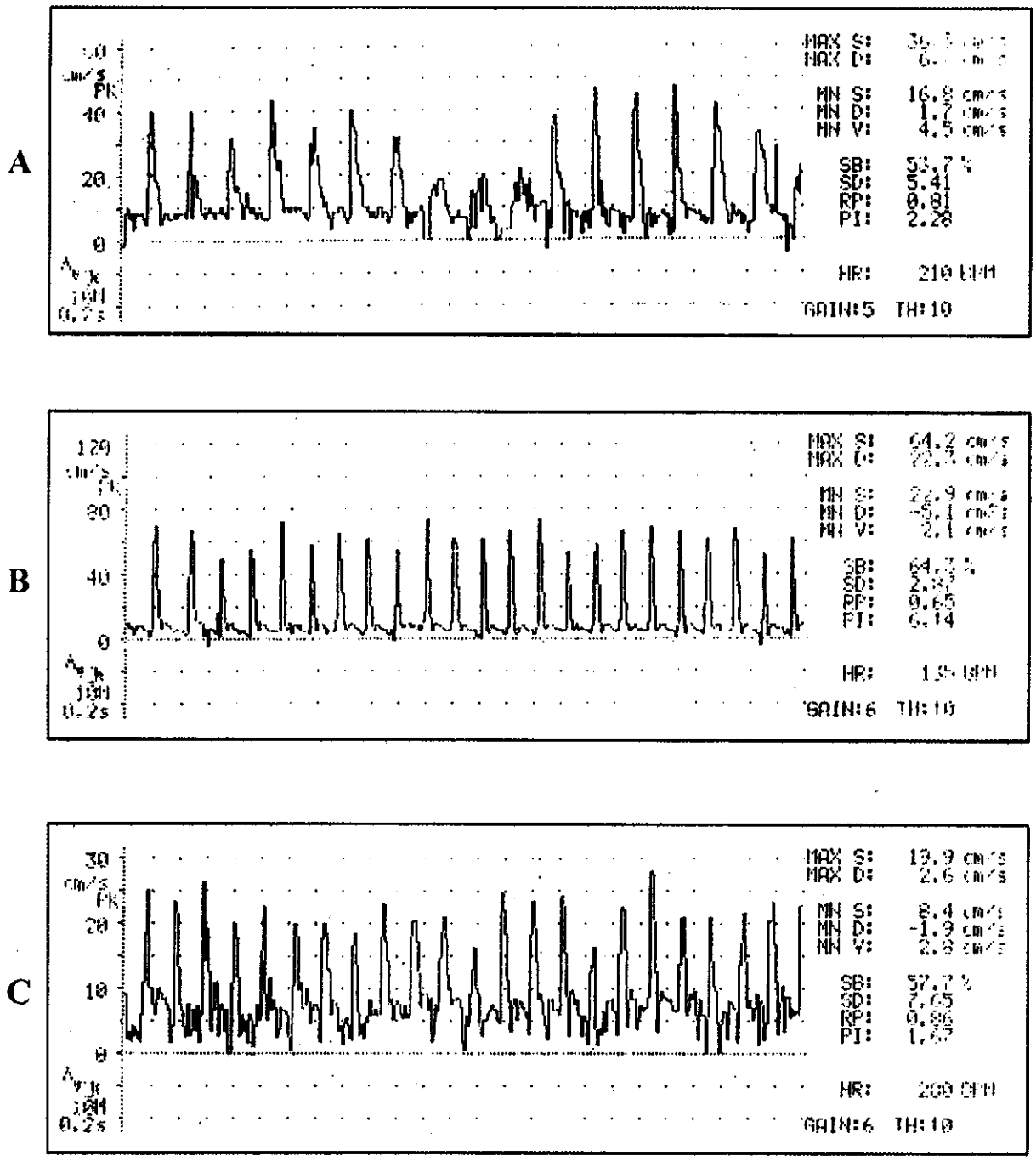


Fig. (9): Diagram showing renal hemodynamics in various groups

A: Control group.

B: DOCA-salt hypertensive hypercholesterolemic group.

C: DOCA- salt hypertensive hypercholesterolemic moxonidine (0.5 mg/kg, I.P. for 2 weeks)- treated group.

• ***Hepatic hemodynamic parameters***

The hepatic blood flow velocity in control group (group I) was 5.38 ± 0.35 cm/s. In hypertensive hypercholesterolemic non-treated rats (group II) the hepatic blood flow velocity was increased insignificantly ($P > 0.05$) to 8.55 ± 1.86 cm/s. Moxonidine treatment (0.5 mg/kg. I.P.) for 2 weeks in group III produced insignificant ($P > 0.05$) reduction of the hepatic blood flow velocity to 8.17 ± 0.99 cm/s compared to group II (Table 3, Figs. 12, 14).

The hepatic artery resistance parameters of the different groups (I, II and III) were 0.72 ± 0.08, 0.82 ± 0.05 and 0.89 ± 0.04 respectively. There were no statistical significant changes among the various groups (Table 3, Figs. 13, 14).

The pulsatility index in group I was 2.77 ± 0.52 increased significantly ($P < 0.05$) to 4.49 ± 0.47 in group II. Treatment of group III with moxonidine (0.5 mg/kg. I.P for 2 weeks) reduced the pulsatility index significantly ($P < 0.01$) to 1.99 ± 0.34 compared to group II (Table 3, Figs. 13, 14).

Moreover, the maximum and minimum systole in the hepatic artery increased significantly ($P < 0.05$) from 41.1 ± 5.43 and 20.87 ± 9.19 cm/s respectively to 91.67 ± 12.16 and 40.32 ± 5.49 cm/s respectively in hypertensive hypercholesterolemic rats (group II). In moxonidine-treated rats (group III) both maximum and minimum systole in hepatic artery decreased significantly ($P < 0.05$) to be 50.0 ± 5.601 and 23.83 ± 3.48 cm/s respectively compared to group II (Table 3, Figs. 10, 14).

The maximum and minimum diastole in group I were 13.42 ± 5.06 and 2.85 ± 0.65 cm/s respectively, insignificantly ($P > 0.05$) changed in other groups to be 14.58 ± 3.15 and 4.52 ± 0.71 cm/s in group II and 8.27 ± 1.68 and 3.57 ± 0.83 cm/s in moxonidine-treated group (group III) (Table 3, Figs. 11, 14).

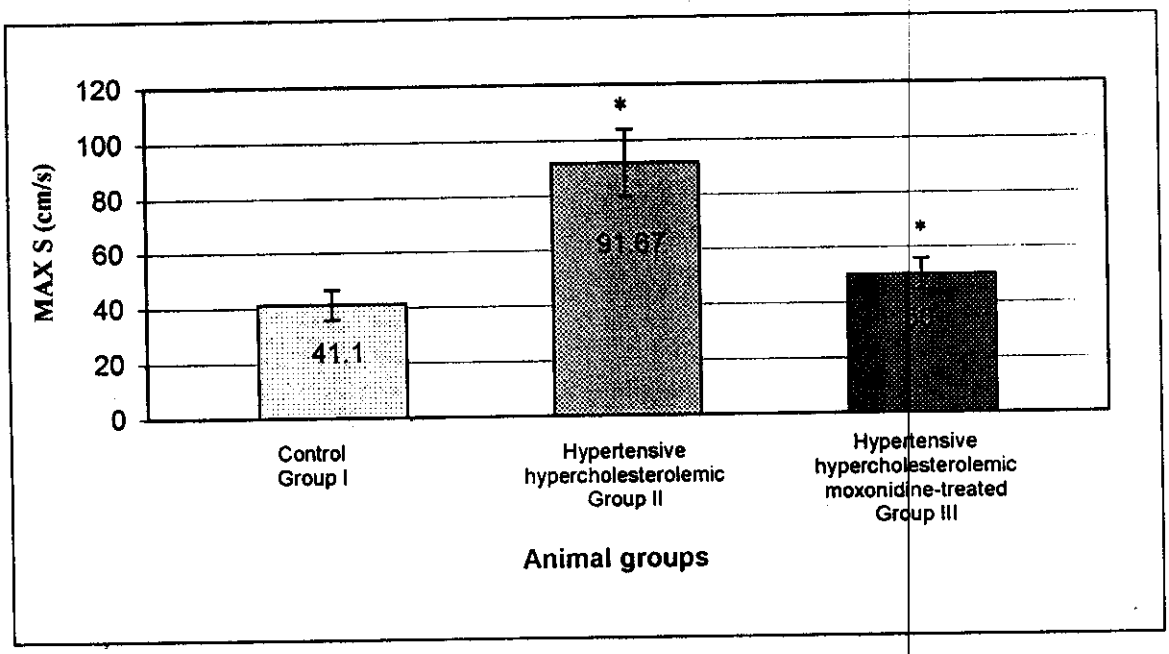
Furthermore, the systole/ diastole ratio was 4.58 ± 1.08 in group I, insignificantly ($P > 0.05$) increased to 7.64 ± 1.54 in group II and to 10.47 ± 3.06 in group III (Table 3, Figs. 12, 14).

Table (3): Effect of moxonidine treatment on hepatic hemodynamic parameters of hypertensive hypercholesterolemic rats.

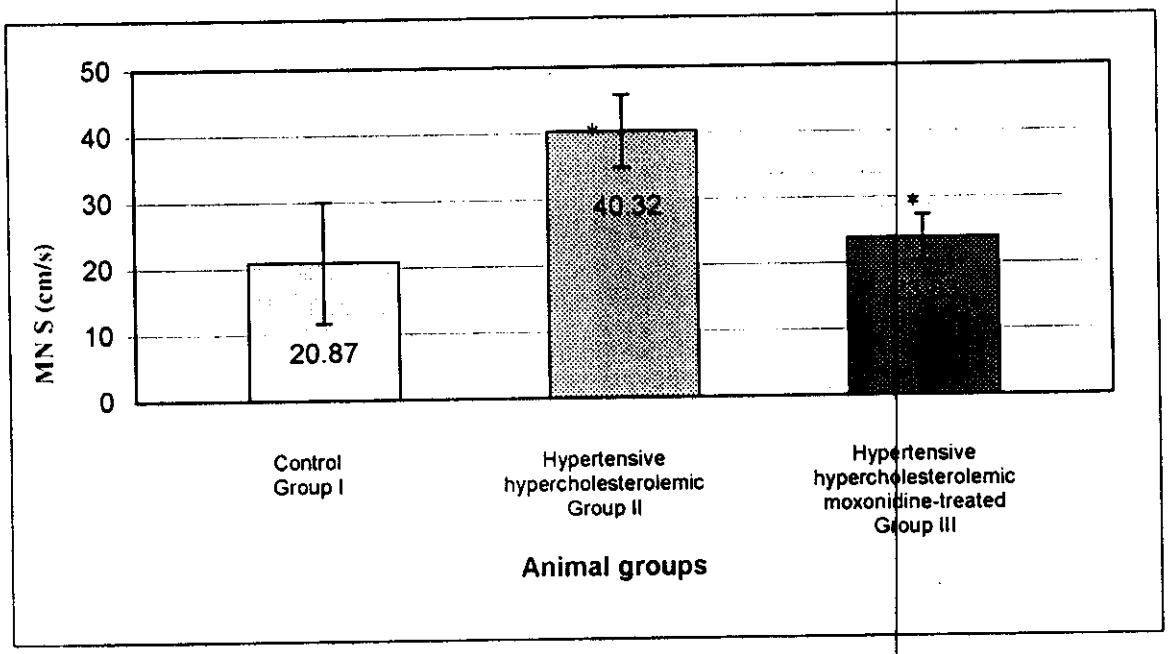
Parameters / Groups	MAX S cm/s	MN S cm/s	MAX D cm/s	MN D cm/s	MN V cm/s	S/D	RP	PI
Group I	41.1 ± 5.43	20.87 ± 9.19	13.42 ± 5.06	2.85 ± 0.65	5.38 ± 0.35	4.58 ± 1.08	0.72 ± 0.08	2.77 ± 0.52
Group II	91.67 ± 12.16	40.32 ± 5.49	14.58 ± 3.15	4.52 ± 0.71	8.55 ± 1.86	7.64 ± 1.54	0.82 ± 0.05	4.49 ± 0.47
P_1	<0.05*	<0.05*	>0.05	>0.05	>0.05	>0.05	>0.05	<0.05*
Group III	50.0 ± 5.6	23.83 ± 3.48	8.27 ± 1.68	3.57 ± 0.83	8.17 ± 0.99	10.47 ± 3.06	0.89 ± 0.04	1.99 ± 0.34
P_2	<0.05*	<0.05*	> 0.05	>0.05	>0.05	>0.05	>0.05	<0.01*

Data represented as mean ± SEM of six experiments

Group I : Control group
Group II : DOCA-salt hypertensive hypercholesterolemic group.
Group III :DOCA-salt hypertensive hypercholestrolemic moxonidine (0.5 mg/kg, I.P. for 2 weeks) treated group.
P_1: Comparing results of group II with that of group I
P_2: Comparing results of group III with that of group II
* Significant at $P < 0.05$.
MAX S: Maximum systole, **MAX D:** Maximum diastole, **MN S:** Minimum systole, **MN D:** Minimum diastole, **MN V:** Mean blood flow velocity, **S/D:** Systole/diastole ratio, **RP:** Resistance parameter in the renal artery, **PI:** Pulsatility index.

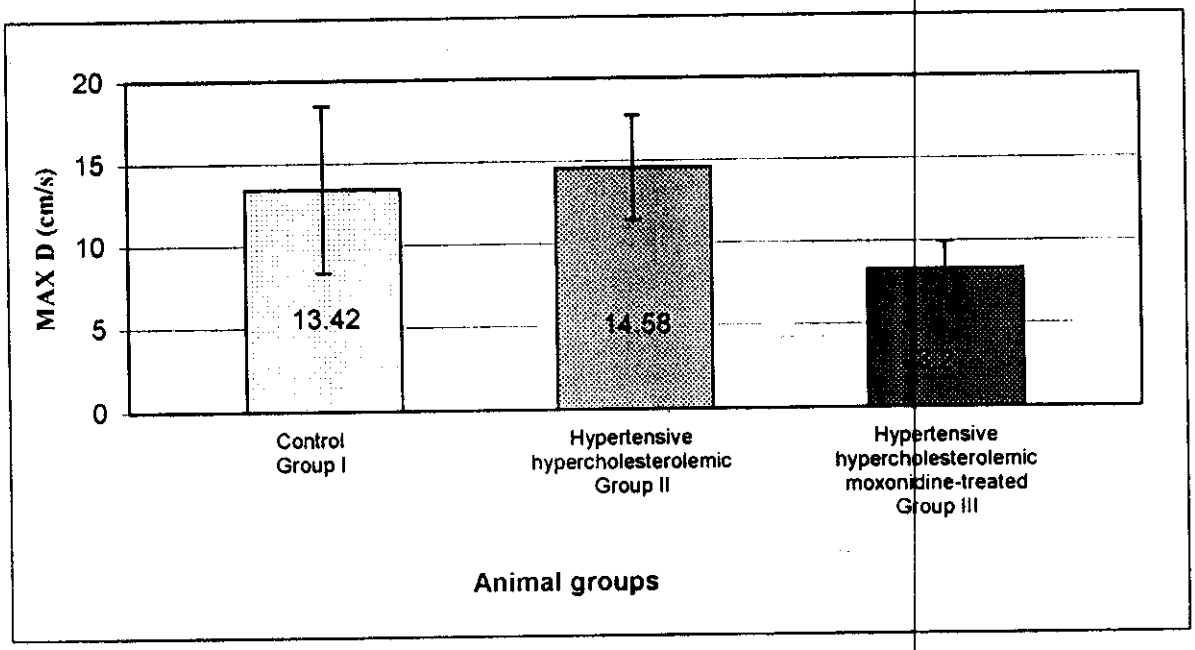


(Maximum Systole)

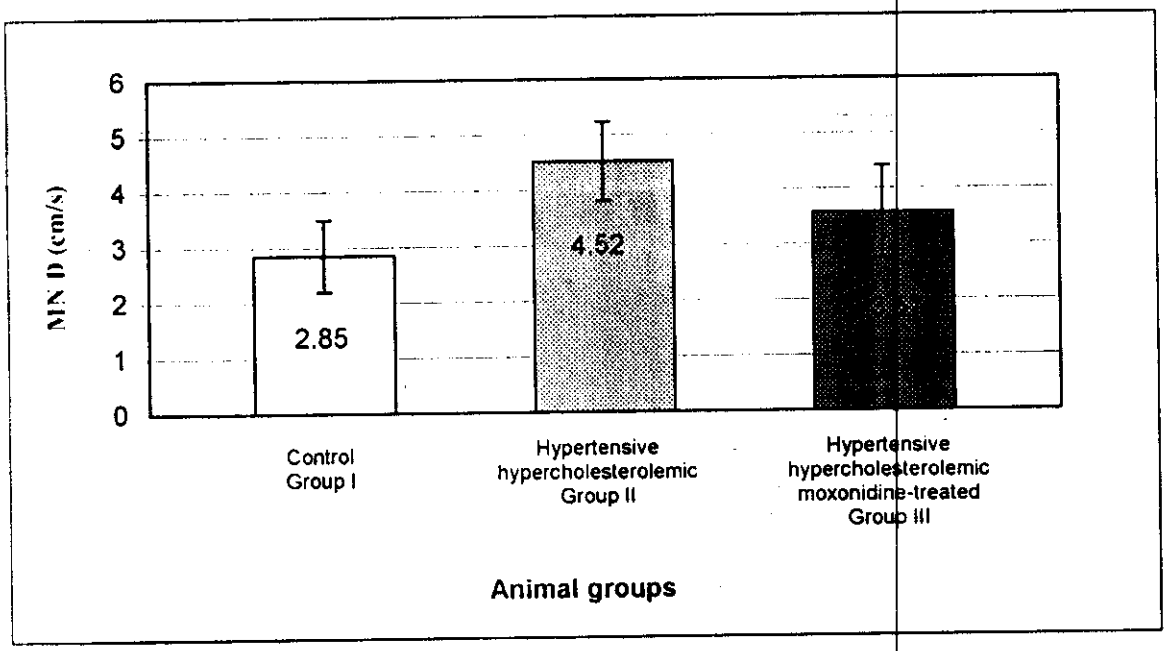


(Minimum Systole)

Figure (10): Histogram showing hepatic hemodynamics in various groups.

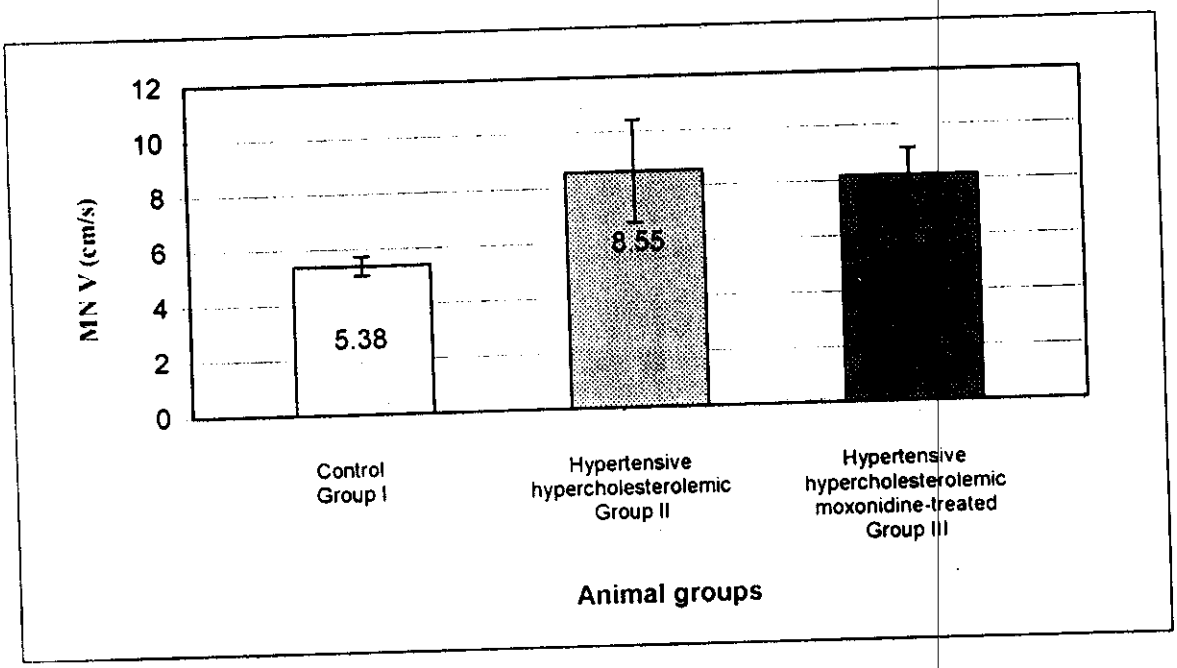


(Maximum Diastole)

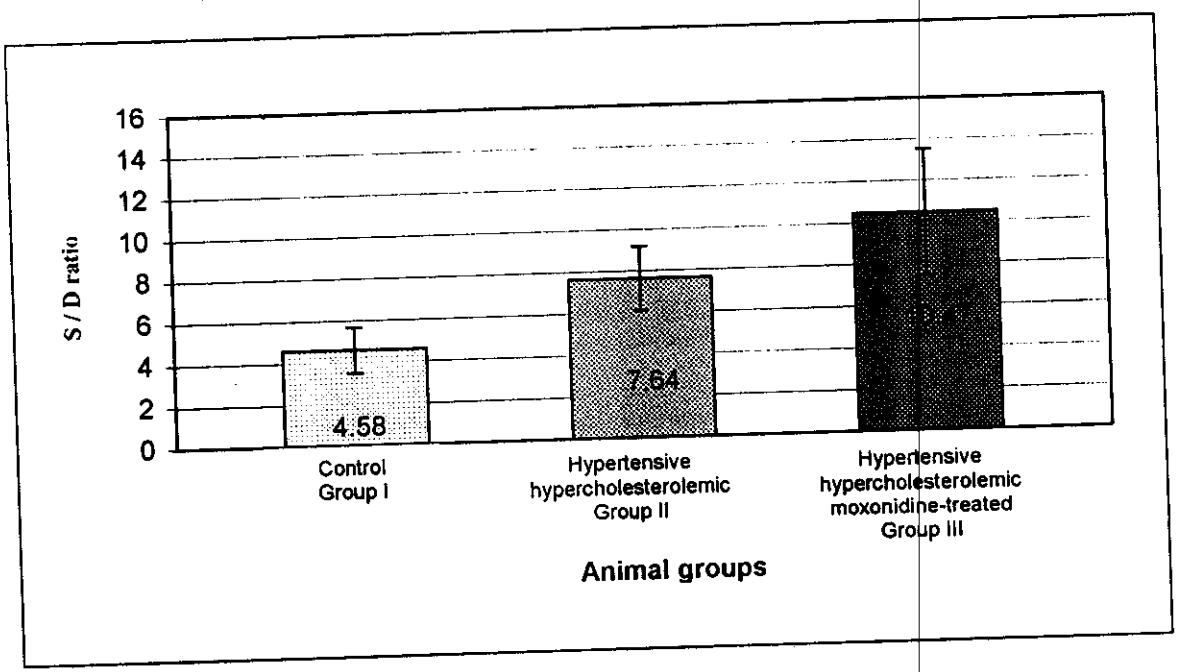


(Minimum Diastole)

Figure (11): Histogram showing hepatic hemodynamics in various groups.

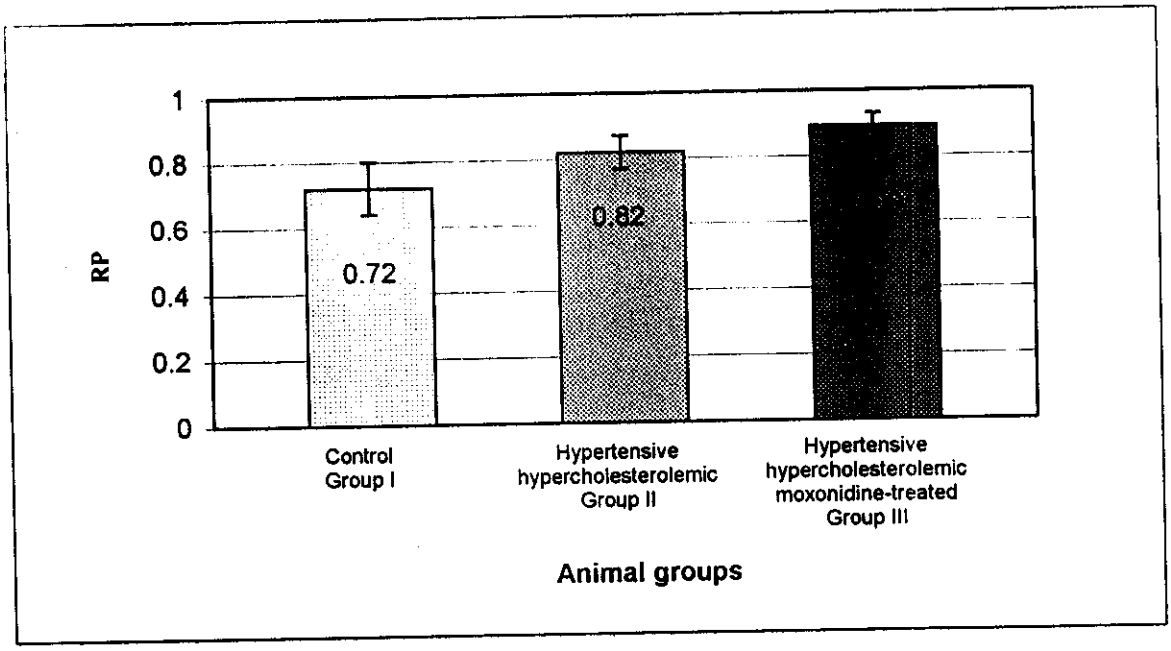


(Mean flow velocity)



(Systole / Diastole ratio)

Figure (12): Histogram showing hepatic hemodynamics in various groups.



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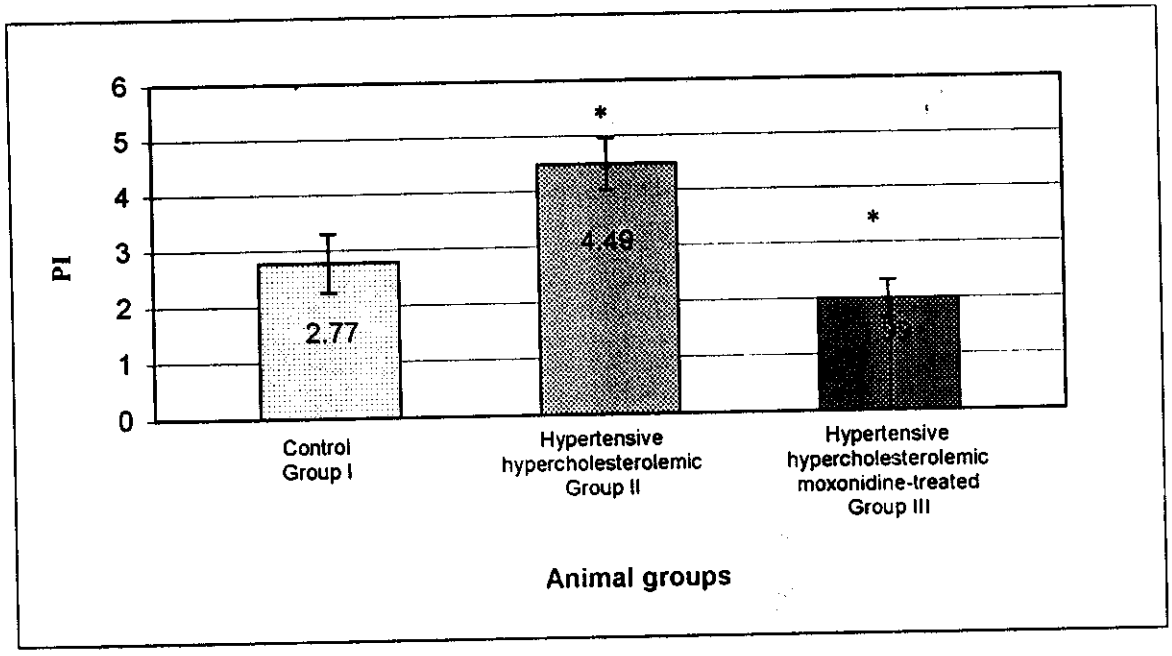


(Pulsitility Index)

Figure (13): Histogram showing hepatic hemodynamics in various groups.

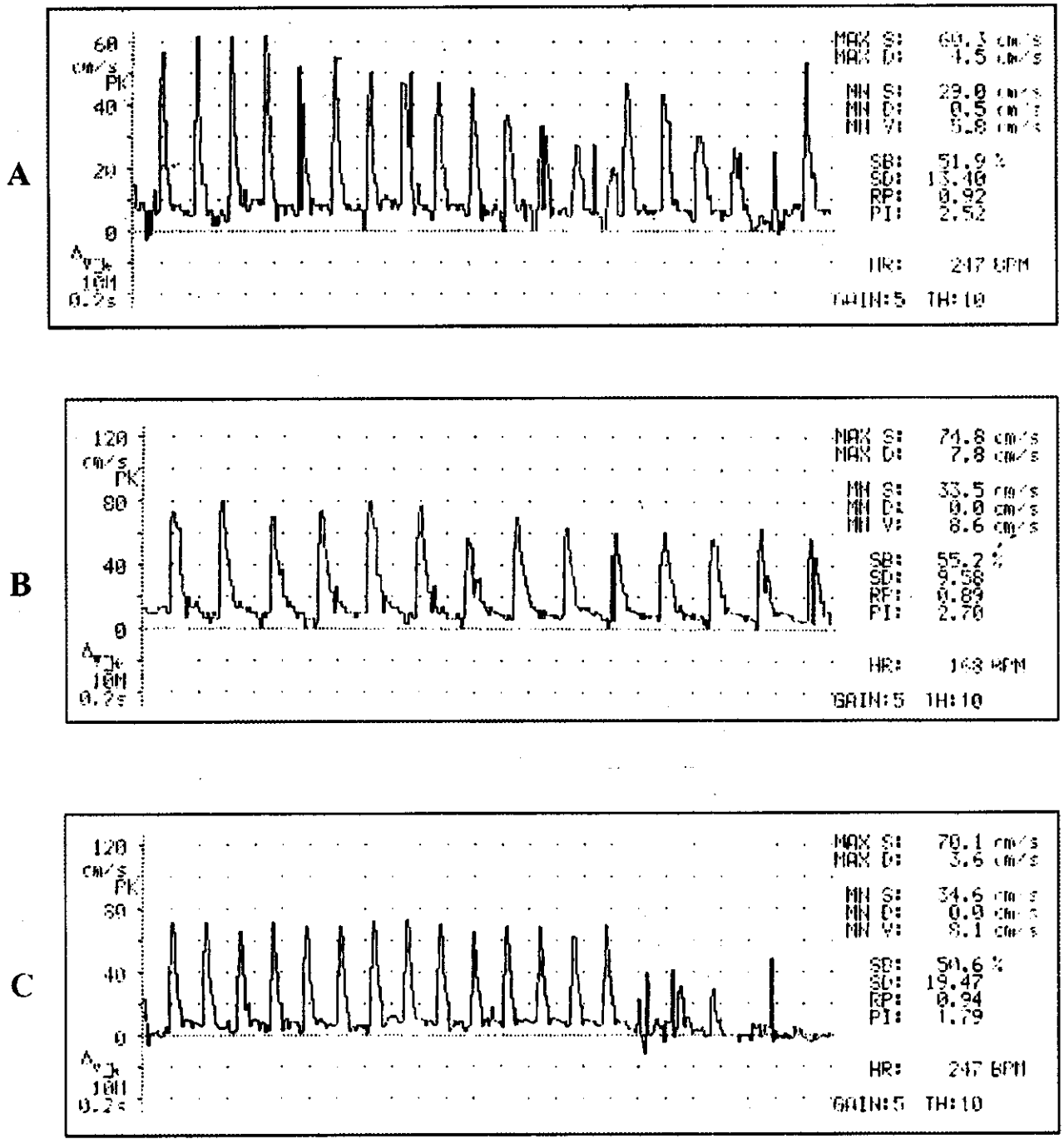


Fig. (14): Diagram showing hepatic hemodynamics in various groups.

A: Control group.

B: DOCA-salt hypertensive hypercholesterolemic group.

C: DOCA- salt hypertensive hypercholesterolemic moxonidine (0.5 mg/kg, I.P. for 2 weeks)- treated group.

II. Biochemical results:

- ***Effect of moxonidine on lipid profile of DOCA-salt, hypertensive hypercholesterolemic rats***

The serum cholesterol level was 92.5 ± 5.59 mg/dl in rats of control group receiving normal diet (group I). While, it was 241.5 ± 5.29 mg/dl in DOCA-salt rats receiving 2 % cholesterol-enriched diet (group II). So, the 2 % cholesterol diet given over 6 weeks significantly ($P < 0.001$) increased the serum cholesterol level to about 2.6 times the level in control group.

Moxonidine administration (0.5 mg / kg. I.P) for 2 weeks to hypertensive hypercholesterolemic rats (group III) induced significant decrease ($P < 0.001$) in serum cholesterol level to 145.5 ± 3.62 mg /dl compared to group II (Table 4, Fig. 15).

Regarding triglyceride level the mean value in normal diet fed rats of group I was 113.33 ± 4.72 mg/dl increased significantly ($P < 0.01$) in DOCA-salt rats fed on 2 % cholesterol-enriched diet of group II to be 139.67 ± 4.07 mg/dl compared to group I. Treatment of group III with moxonidine (0.5 mg/kg, I.P. for 2 weeks) produced significant ($P < 0.05$) reduction of triglyceride concentration to 125.83 ± 4.16 mg / dl compared to group II (Table 4, Fig. 16).

The serum HDL-cholesterol in rats on normal diet (group I) was 41.18 ± 1.38 mg /dl . However, 2 % cholesterol feeding for 6 weeks in group II insignificantly ($P > 0.05$) raised HDL-C to 43.14 ± 1.42 mg/dl compared to group I. In treated group (group III) moxonidine produced insignificant ($P > 0.05$) increase in HDL-C concentration to 45.13 ± 1.82 mg/dl compared to group II (Table 4, Fig. 17).

Moreover, in group I the serum LDL-cholesterol concentration was 21.18 ± 1.28 mg /dl increased significantly ($P < 0.001$) to 112.08 ± 5.44 mg/dl in group II and decreased significantly ($P<0.01$) to 66.35 ± 4.71 mg /dl in moxonidine treated group (Group III) compared to group II (Table 4, Fig. 18).

Table (4): Effect of moxonidine treatment on lipid profile of hypertensive hypercholesterolemic rats.

Groups / Parameters	Group I	Group II	Group III
Total serum cholesterol(mg/dl)	92.5 ± 5.59	241.5 ± 5.29 $P_1 < 0.001$*	145.5 ± 3.62 $P_2 < 0.001$*
Triglycerides (mg/dl)	113.33 ± 4.72	139.67 ± 4.07 $P_1 < 0.01$*	125.83 ±4.16 $P_2 < 0.05$*
HDL-cholesterol (mg/dl)	41.18 ± 1.38	43.14 ± 1.42 $P_1 > 0.05$	45.13 ± 1.82 $P_2 > 0.05$
LDL-cholesterol (mg/dl)	21.18 ± 1.28	112.08 ± 5.44 $P_1 < 0.001$*	66.35 ± 4.71 $P_2 < 0.01$*

Data represented as mean ± SEM of six experiments

Group I : Control group

Group II : DOCA-salt hypertensive hypercholesterolemic group.

Group III : DOCA-salt hypertensive hypercholestrolemic moxoni-dine (0.5 mg/kg I.P. for 2 weeks) treated group.

P_1: Comparing results of group II with that of group I

P_2: Comparing results of group III with that of group II

* Significant at $P < 0.05$

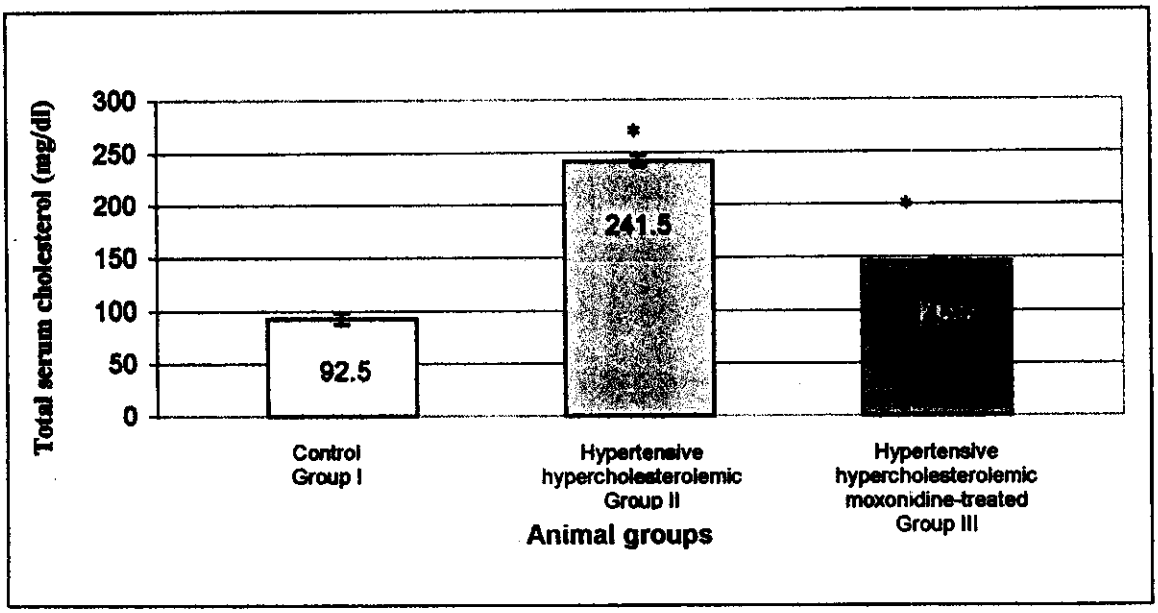


Fig. (15): Histogram showing total serum cholesterol in various groups

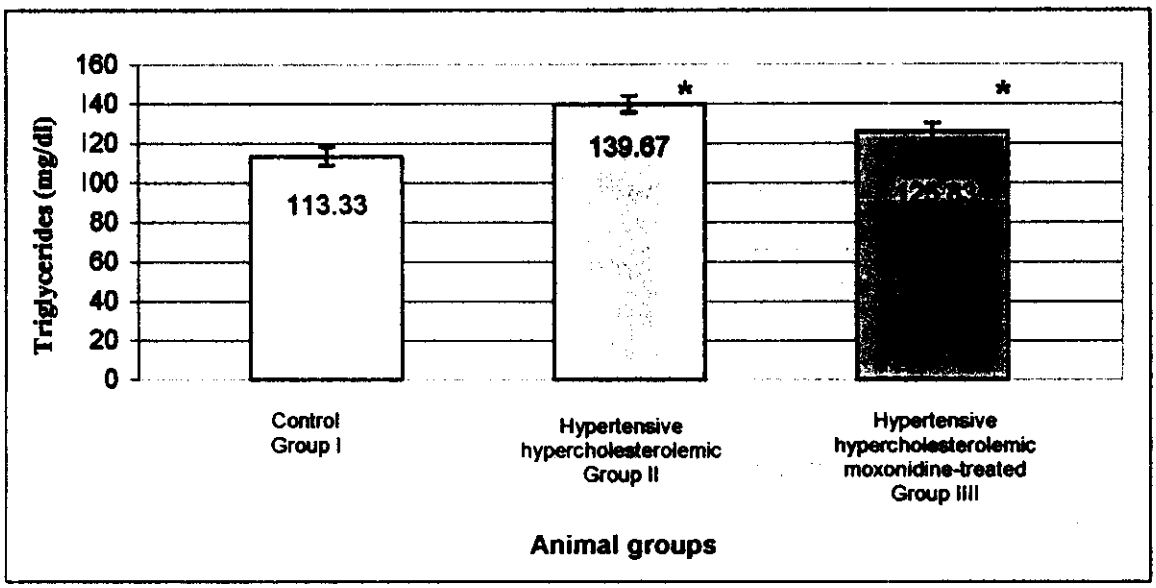


Fig. (16): Histogram showing triglycerides in various groups.

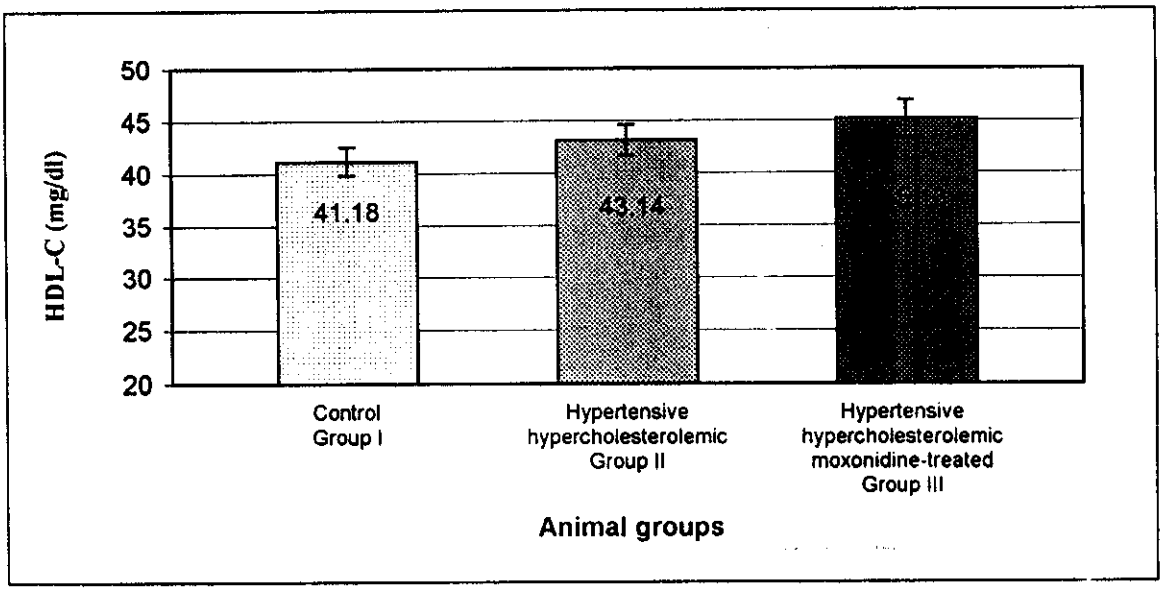


Fig. (17): Histogram showing HDL-Cholesterol in various groups.

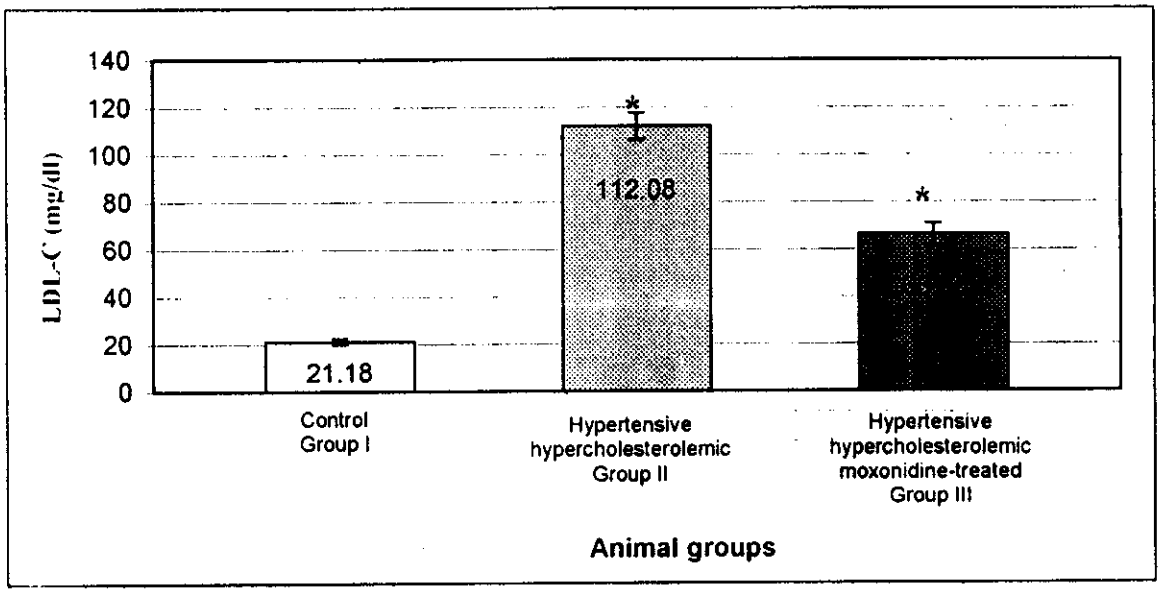


Fig. (18): Histogram showing LDL-Cholesterol in various groups

- ***Effect of moxonidine treatment on serum sodium and potassium levels of hypertensive hypercholesterolemic rats***

The serum sodium concentration in control group (group I) was 135.83 ± 5.17 mEq/l increased insignificantly ($P > 0.05$) to 141.33 ± 1.84 mEq/l in hypertensive hypercholesterolemic rats (group II). In moxonidine-treated group (group III) serum sodium concentration was significantly ($P < 0.05$) decreased to 128.83 ± 5.33 mEq/l compared to group II (Table 5, Fig. 19).

The serum potassium concentrations in various groups (I, II and III) were 3.79 ± 0.38, 3.91 ± 0.105 and 3.83 ± 0.194 m mol/l respectively. There was no statistical significant change ($P > 0.05$) among the three groups (Table 5, Fig. 20).

- ***Effect of moxonidine treatment on serum creatinine of hypertensive hypercholesterolemic rats.***

Serum creatinine level was 0.89 ± 0.05 mg / dl in group I increased insignificantly ($P > 0.05$) to 1.08 ± 0.165 mg/ dl in group II. Treatment with moxonidine (0.5 mg/kg, I.P for 2 weeks) insignificantly decreased the serum creatinine level to 0.97 ± 0.07 mg/dl in group III compared to group II (Table 5, Fig. 21).

Table (5): Effect of moxonidine treatment on serum sodium and potassium and creatinine of hypertensive hyper-cholesterolemic rats.

Groups / Parameters	Group I	Group II	Group III
Serum sodium mEq/l	135.83 ± 5.17	141.33 ± 1.84 $P_1 > 0.05$	128.83 ± 5.33 $P_2 < 0.05$*
Serum potassium mmol/l	3.79 ± 0.38	3.91 ± 0.11 $P_1 > 0.05$	3.83 ± 0.19 $P_2 > 0.05$
Serum creatinine (mg/dl)	0.89 ± 0.05	1.08 ± 0.17 $P_1 > 0.05$	0.97 ± 0.07 $P_2 > 0.05$

Data represented as mean ± SEM of six experiments

Group I : Control group

Group II : DOCA-salt hypertensive hypercholesterolemic group.

Group III : DOCA-salt hypertensive hypercholestrolemic moxonidine (0.5 mg/kg, I.P. for 2 weeks) treated group.

P_1: Comparing results of group II with that of group I

P_2: Comparing results of group III with that of group II

* Significant at $P < 0.05$

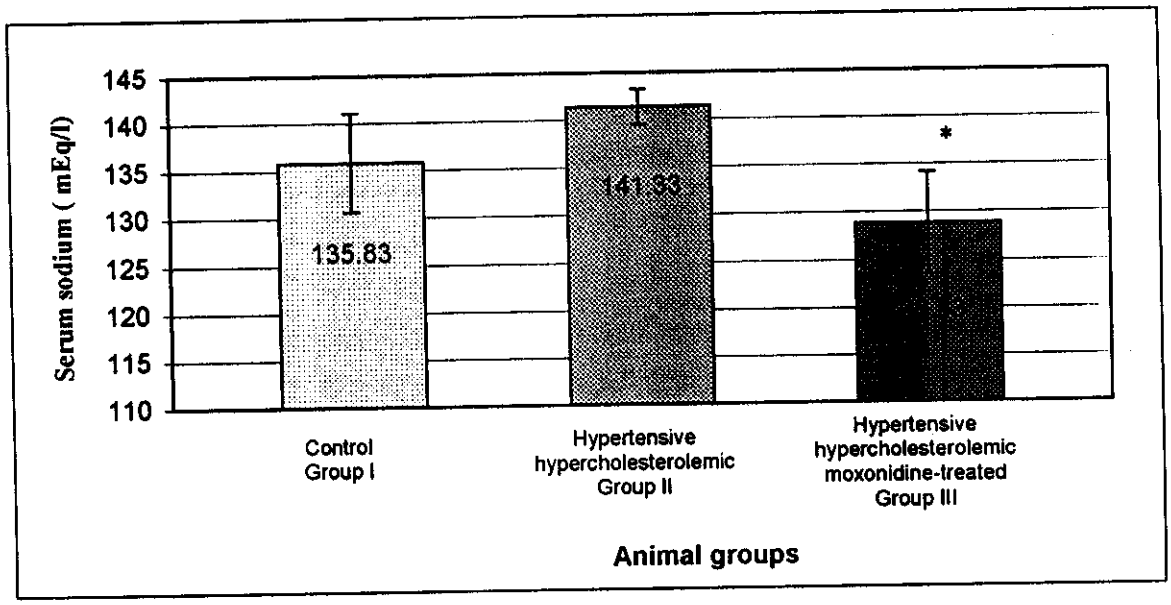


Fig. (19): Histogram showing serum sodium in various groups.

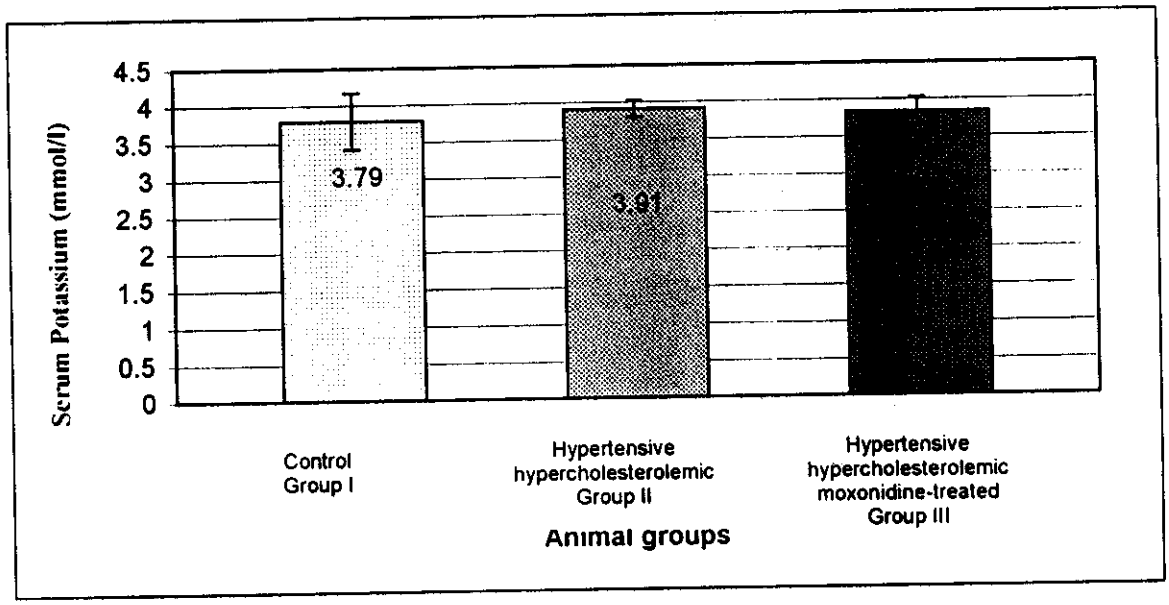


Fig. (20): Histogram showing serum potassium in various groups.

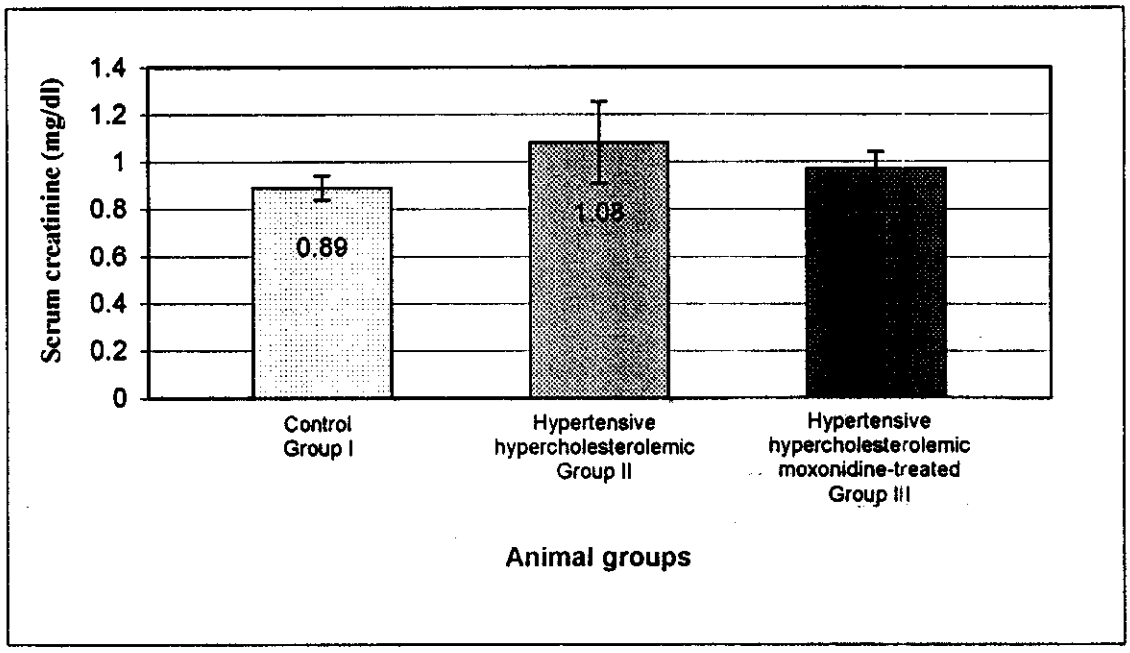


Fig. (21): Histogram showing serum creatinine in various groups.

III. Histopatholgical results:

- **Effect of moxonidine treatment on histopathological changes in liver, kidney and aorta of DOCA-salt hypertensive hypercholesterolemic rats**

1. The liver

Histological examination of the liver of the control group (group I) showed that the liver formed of classical hepatic lobules, each lobule was formed of a central vien radiating from it liver cell plates. Between these plates slit-like spaces were seen representing the blood sinusoids. The hepatocytes appeared as polygonal cells with granular and eosinophilic cytoplasm and central, large and rounded nuclei with one or more nucleoli. The connective tissue of the liver were demonstrated as a thin layer of collagenous fibers in the wall of the central vein and appeared surrounding the classic hepatic lobules (Fig. 22).

Histological examination of the liver of the DOCA-salt hypertensive hypercholesterolemic rats (group II) showed that hepatocytes contained cytoplasmic vacuoles which were sharply defined and of variable size with their nuclei indented, most probably denoting fatty change (Fig. 23). Centrilobular hepatocytes showed mild affection in the form of swollen cells with pale cytoplasm (cloudy swelling) and vaculated cells with normal nuclei (hydropic degeneration) (Fig. 24). Furthermore, central vein congestion and dilatation of sinusoids were observed (Fig. 25). In moxonidine-treated rats (group III) (0.5 mg/kg, I.P for 2 weeks), it was observed that the hepatocytes showed decrease in cytoplasmic vacuolation, appearance of more regular hepatic cords and decreased congestion and dilatation of central vein (Fig. 26).

2. The kidney

Histological examination of the kidney of group I showed that the kidney was formed of an outer cortex and inner medulla and was surrounded by a connective tissue capsule. The cortex of the kidney consisted of malpighian renal corpuscles formed of a glomerulus and Bowman's capsule, proximal convoluted tubules (P.C.T.) lined with pyramidal cells, loop of Henle lined by simple squamous epithelium and cuboidal cells, and distal convoluted tubules (D.C.T) lined with cubical cells. The medulla was formed of loops of Henle and collecting tubules lined by cuboidal cells, the larger ones were by columnar cells (Fig. 27).

In group II, histological examination of the kidney revealed endothelial cell proliferation (arteriosclerosis) (Fig. 28), narrowing of the Bowman's capsule due to mesangeal cell proliferation, thicking of the glomerular capillary basement membrane (glomerulosclerosis) and intracellular and extracellular hyalinosis (Fig. 29).

Moxonidine treatment (0.5 mg/kg, I.P) for 2 weeks of group III produced mild improvement of all glomerular lesions appeared as increased lumen of the glomerular capillary tuft and increased capsular space (Fig. 30).

3. The aorta

Histological examination of a cut section of the aorta of group I showed that the wall of the aorta consists of a tunica intima, tunica media, and tunica adventitia. The tunica intima consists of an endothelial coat of flattened squamous cells resting on a complete basal lamina and is supported by a subendothelial loose connective tissue. The tunica media consists largely of elastic, concentric laminae and variable amounts of

smooth mucle cells. The tunica adventitia contains bundles of collagen fibers and a few elastic fibers both of which have a loose, helical arrangement (Fig.31).

In group II, histological examination of the aorta showed ulcerated endothelial cells of the intima with collection of fat globules and foamy cells and formation of fatty streaks. The media and adventitia showed fibrosis and inflammatory cell infiltration (Fig. 32).

In group III, moxonidine treatment decreased the size of fatty streaks. Regeneration of the endothelial cells of the intima was observed (Fig. 33).

Fig. (22): A photomicrograph of a section in the liver of a control rat (group I) showing hepatic plates radiating from the central veins and separated by blood sinusoids (H x & E x 200)

Fig. (23): A photomicrograph of a section in the liver of a hypertensive hypercholesterolemic rat (group II). The cytoplasm of hepatocytes showing intracellular sharply defined fat vacuoles (fatty change). The nuclei are pushed to one side. (H x & E x 400)

Fig. (24): A photomicrograph of a section in the liver of a rat from group II. The hepatocytes showing vacuolated cytoplasm with normal nuclei (hydropic degeneration). (H x & E x 400)

Fig. (25): A photomicrograph of a section in the liver of a rat from group II showing congested central vein. (H x & E x 400)

Fig. (26): A photomicrograph of a section in the liver of a rat treated with moxonidine (group III) showing improved fatty and hydropic degeneration of hepatocytes and decrease in central vein congestion. (H x & E x 400)

Fig. (27): A photomicrograph of a section in the kidney of a control rat (group I) showing renal corpuscles formed of glomeruli, surrounded by proximal convoluted tubules, distal convoluted tubules, and collecting tubules.

(H x & E x 200)

Fig. (28): A photomicrograph of a section in the kidney of a hyertensive hypercholesterolemic rat (groupII) showing endothelial cell proliferation of the intima and thickening of the media (Arteriolosclerosis)

(H x & E x 400)

Fig. (29): A photomicrograph of a section in the kidney of rat from group II showing thickening of the glomenular capillary basement membrane (glomerulosclerosis). The surrounding tubules showing both inracellular and extracellular hyalinosis leading to obliterated lumen.

(H x & E x 400)

Fig. (30): A photomicrograph of a section in the kidney of rat treated with moxonidine (group III) showing mild degenerative changes affecting renal tubules and glomerular capillary tuft with increased lumen and capsular space.

(H x & E x 400)

Fig. (31): A photomicrograph of a cut section in the aorta of a control rat (group I) showing intact flat endothelial cell lining, elastic media and well fitted adventitia. (H x & E x 200)

Fig. (32): A photomicrograph of a cut section in the aorta of a hypertensive hypercholesterolemic rat (group II) showing ulcerated endothelial cells of the intima, collection of foamy histocytes and fat globules with formation of fatty streaks. The adventitia shows fibrosis and inflammatory cell infiltration. (H x & E x 200)

Fig. (33): A photomicrograph of a cut section in the aorta of a rat treated with moxonidine (group III) showing regenerated endothelial cells of he intima, mild medial fibrosis and reduced size of the fatty streaks. (H x & E x 200)

(B) Acute studies

1. Effect of acute intravenous injection of moxonidine on renal hemodynamic parameters in normotensive rats

Acute intravenous administration of bolus doses of moxonidine (0.1, 0.3 and 1.0 mg/kg) increased the renal blood flow velocity from a basal value of 4.22 ± 0.32 cm/s to 4.3 ± 0.28, 4.87 ± 0.39 and 6.63 ± 0.53 cm/s respectively. This increase was statistically insignificant ($P > 0.05$) at the doses of 0.1 and 0.3 mg/kg but was significant ($P < 0.05$) at the dose of 1mg /kg (Table 6, Figs. 36, 38).

Furthermore, moxonidine decreased the renal artery resistance from a basal value of 0.97 ± 0.02 to 0.89 ± 0.07, 0.83 ± 0.06 and 0.91 ± 0.02 for the doses of 0.1, 0.3 and 1 mg/kg respectively. This decrease in the resistance parameter was statistically insignificant at the dose of 0.1 mg / kg but was significant ($P < 0.05$) at the doses of 0.3 and 1 mg/kg (Table 6, Figs. 37, 38).

The basal value of pulsatility index was 2.83 ± 0.73 insignificantly ($P > 0.05$) affected by I.V. injection of different doses of moxonidine 0.1, 0.3 and 1 mg/kg to be 3.44 ± 0.58, 3.62 ± 0.63 and 2.69 ± 0.66 respectively (Table 6, Figs. 37, 38).

Moreover, the maximum systole in the renal artery increased insignificantly ($P > 0.05$) from a mean of 28.57 ± 2.58 cm/s to 32.82 ± 1.67, 37.62 ± 6.18 and 33.97 ± 2.66 cm/s for the doses 0.1, 0.3 and 1 mg/kg of moxonidine respectively. The minimum systole increased insignificantly from 12.2 ± 2.09 cm/s to 16.17 ± 2.85, 16.25 ± 1.65 and 15.05 ± 1.14 cm/s (Table 6, Figs. 34, 38).

There was insignificant ($P > 0.05$) increase of maximum diastolic blood pressure in renal artery from 4.13 ± 0.75 cm/s 4.08 ± 0.21, 4.16 ± 0.3 and 4.18 ± 0.24 and there was insignificant changes in the minimum diastolic pressure from 2.57 ± 0.95 cm/s to 2.2 ± 0.82, 2.4 ± 0.97 and 2.6 ± 0.62 cm/s for the doses of moxonidine 0.1, 0.3 and 1 mg/kg respectively (Table 6, Figs. 35, 38).

The systole / diastole ratio was 7.43 ± 1.95 insignificantly ($P > 0.05$) affected by different doses of moxonidne 0.1, 0.3 and 1 mg/kg to be 7.85 ± 2.6, 5.76 ± 0.96 and 7.32 ± 0.72 respectively (Table 6, Figs. 36, 38).

Table (6): Effect of acute administration of moxonidine on renal hemodynamic parameters in normotensive rats.

Parameters / Groups	MAX S cm/s	MN S cm/s	MAX D cm/s	MN D cm/s	MN V cm/s	S/D	RP	PI
Control group	28.57 ± 2.58	12.2 ± 2.09	4.13 ± 0.75	2.57 ± 0.95	4.22 ± 0.32	7.43 ± 1.95	0.97 ± 0.02	2.83 ± 0.73
Moxonidine 0.1 mg/kg	32.82 ± 1.67	16.17 ± 2.85	4.08 ± 0.21	2.2 ± 0.82	4.3 ± 0.28	7.85 ± 2.6	0.89 ± 0.07	3.44 ± 0.58
P	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05
Moxonidine 0.3 mg/kg	37.62 ± 6.18	16.25 ± 1.65	4.16 ± 0.3	2.4 ± 0.97	4.87 ± 0.39	5.76 ± 0.96	0.83 ± 0.06	3.62 ± 0.63
P	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	<0.05*	>0.05
Moxonidine 1 mg/kg	33.97 ± 2.66	15.05 ± 1.14	4.18 ± 0.24	2.6 ± 0.62	6.63 ± 0.53	7.32 ± 0.72	0.91 ± 0.02	2.69 ± 0.66
P	>0.05	>0.05	> 0.05	>0.05	<0.05*	>0.05	<0.05*	>0.05

Data represented as mean ± SEM of six experiments

* Significant at $P < 0.05$ compared to control group.
MAX S: Maximum systole, **MAX D:** Maximum diastole, **MN S:** Minimum systole, **MN D:** Minimum diastole, **MN V:** Mean blood flow velocity, **S/D:** Systole/diastole ratio, **RP:** Resistance parameter in the renal artery, **PI:** Pulsatility index.

2. Effect of acute intravenous injection of moxonidine on hepatic hemodynamic parameters in normotensive rats

The basal hepatic blood flow velocity was 5.38 ± 0.35 cm/s. Acute intravenous administration of bolus doses of moxonidine 0.1, 0.3 and 1 mg/kg increased the hepatic blood flow velocity to be 5.52 ± 0.33, 6.12 ± 0.48 and 9.8 ± 1.99 cm/s respectively. This increase was statistically insignificant ($P > 0.05$) at the doses of 0.1 and 0.3 mg/kg but was significant ($P < 0.05$) at the dose of 1 mg/kg (Table 7, Figs. 41, 43).

Furthermore, the basal value of the resistance in hepatic artery was 0.89 ± 0.05. Acute intravenous injection of moxonidine in a dose of 0.1 mg/kg did not produce any change in the hepatic artery resistance parameter to be 0.89 ± 0.03.

However, moxonidine in doses of 0.3 and 1 mg/kg decreased the resistance parameter to 0.86 ± 0.03 and 0.72 ± 0.08 respectively. This decrease was significant ($P < 0.05$) at the higher dose (Table 7, Figs. 42, 43).

Acute intravenous administration of moxonidine 0.1, 0.3 and 1 mg/kg did not induce any significant change ($P > 0.05$) in pulsatility index from a mean of 2.77 ± 0.52 to 2.42 ± 0.43, 1.97 ± 0.23 and 2.14 ± 0.26 respectively (Table 7, Figs. 42, 43).

Moreover, moxonidine in different doses of 0.1, 0.3 and 1 mg/kg, I.V. did not show any changes ($P > 0.05$) in other parameters of hepatic artery. The maximum systolic blood pressure increased from 57.77 ± 13.38 to 48.8 ± 8.19, 44.83 ± 4.52 and 63.38 ± 6.79 cm/s, the minimum

systole changed from 20.87 ± 3.76 to 19.82 ± 2.64, 21.87 ± 2.4 and 30.42 ± 3.23 (Table 7, Figs. 39, 43).

The maximum diastole decreased from 13.42 ± 5.06 to 5.9 ± 1.19, 5.37 ± 1.04 and 5.85 ± 1.39 cm/s and the minimum diastole changed from 2.85 ± 0.65 to 2.98 ± 0.94, 2.03 ± 0.76 and 2.57 ± 0.94 cm/s (Table 7, Fig. 40, 43) and the systole / diastole ratio increased from 4.58 ± 1.08 to 4.32 ± 2.62, 7.45 ± 1.62 and 12.09 ± 4.17 respectively (Table 7, Figs. 41, 43).

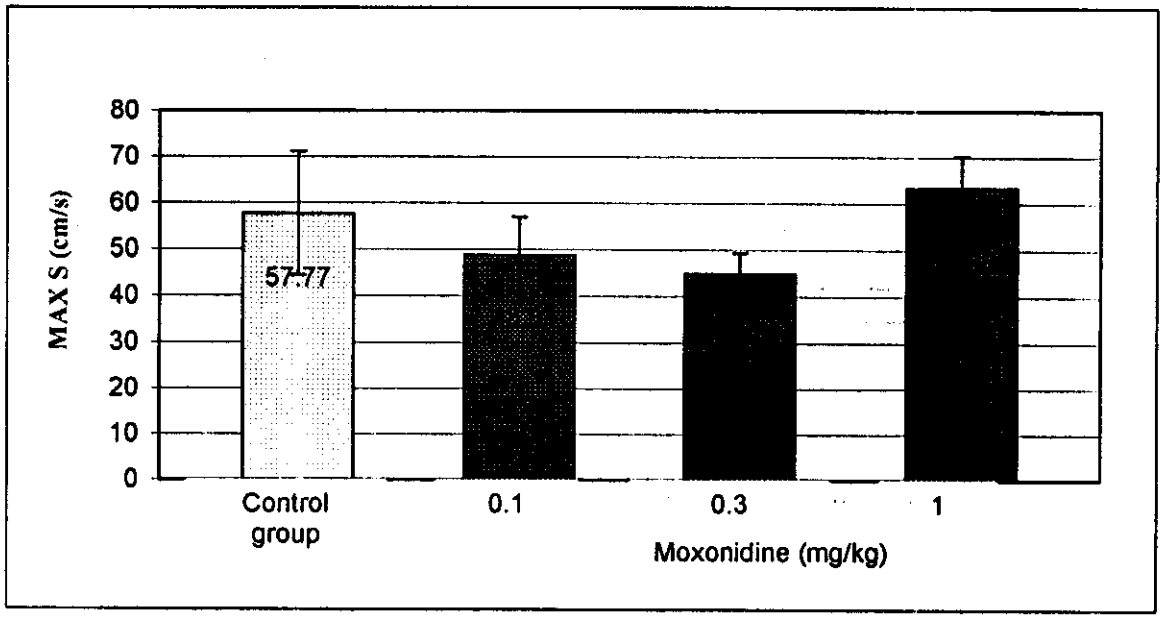
Table (7): Effect of acute administration of moxonidine on hepatic hemodynamic parameters in normotensive rats.

Parameters / Groups	MAX S cm/s	MN S cm/s	MAX D cm/s	MN D cm/s	MN V Cm/s	S/D	RP	PI
Control group	57.77 ± 13.38	20.87 ± 3.76	13.42 ± 5.06	2.85 ± 0.65	5.38 ± 0.35	4.58 ± 1.08	0.89 ± 0.05	2.77 ± 0.52
Moxonidine 0.1 mg/kg	48.8 ± 8.19	19.82 ± 2.64	5.9 ± 1.19	2.98 ± 0.94	5.52 ± 0.33	4.32 ± 2.62	0.89 ± 0.03	2.42 ± 0.43
P	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05
Moxonidine 0.3 mg/kg	44.83 ± 4.52	21.87 ± 2.4	5.37 ± 1.04	2.03 ± 0.76	6.12 ± 0.48	7.45 ± 1.62	0.86 ± 0.03	1.97 ± 0.23
P	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05	>0.05
Moxonidine 1 mg/kg	63.38 ± 6.79	30.42 ± 3.23	5.85 ± 1.39	2.57 ± 0.94	9.8 ± 1.99	12.09 ± 4.17	0.72 ± 0.08	2.14 ± 0.26
P	>0.05	> 0.05	>0.05	>0.05	<0.05*	>0.05	<0.05*	>0.05

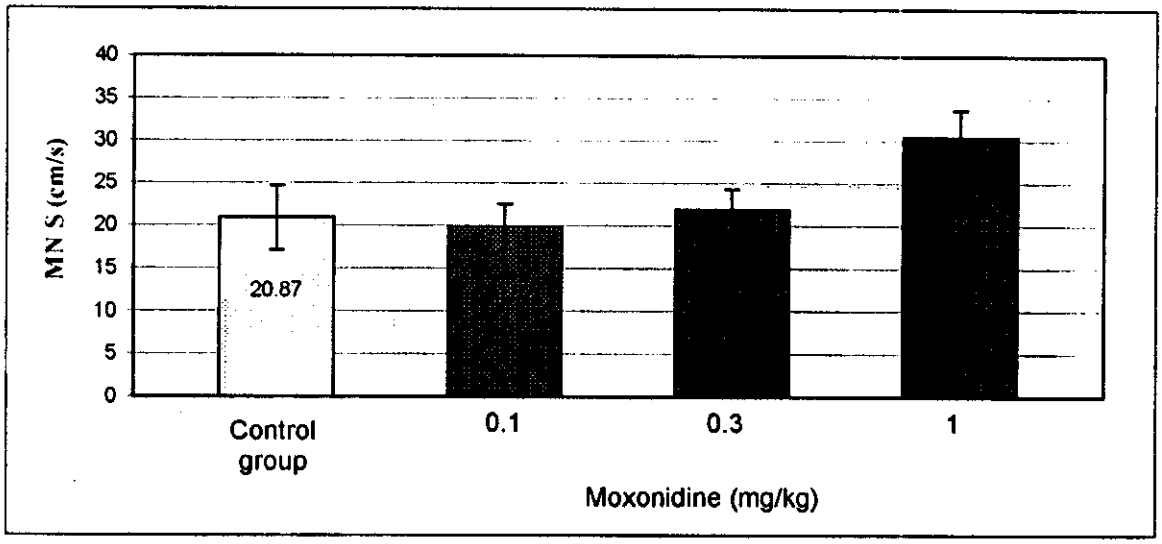
Data represented as mean ± SEM of six experiments

* Significant at $P < 0.05$ compared to control group.

MAX S: Maximum systole, **MAX D:** Maximum diastole, **MN S:** Minimum systole, **MN D:** Minimum diastole, **MN V:** Mean blood flow velocity, **S/D:** Systole/diastole ratio, **RP:** Resistance parameter in the renal artery, **PI:** Pulsatility index.

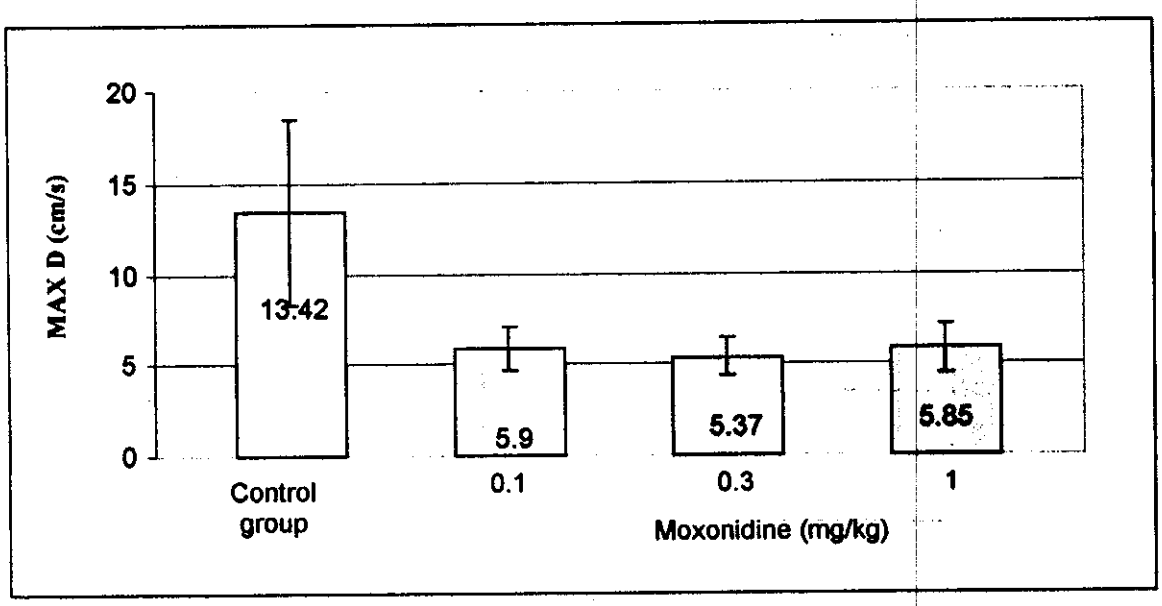


(Maximum Systole)

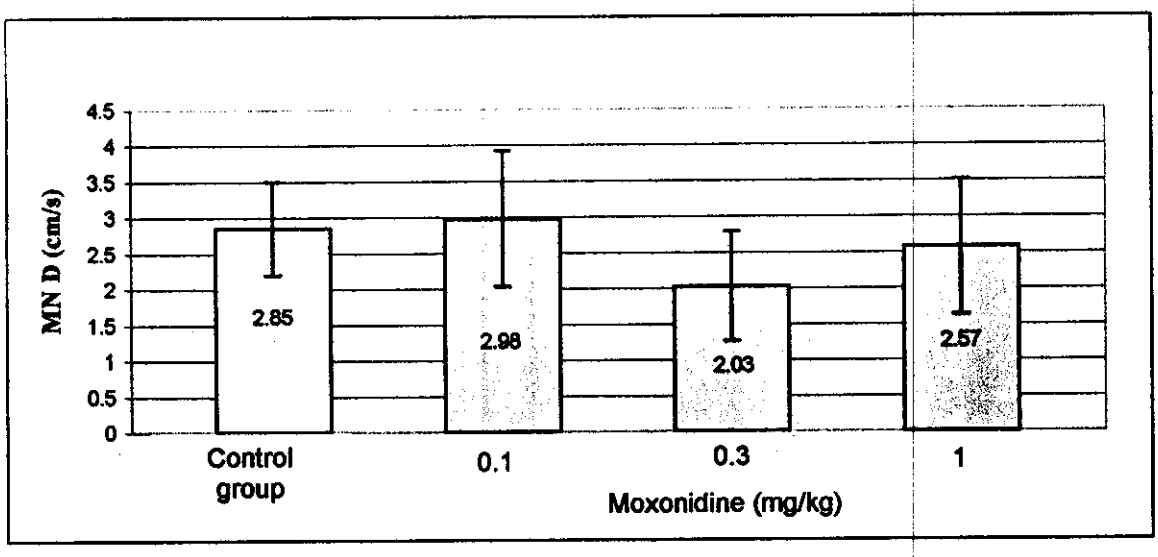


(Minimum Systole)

Fig.(39): Histogram showing the effect of acute administration of moxonidine on maximum and minimum systolic values of hepatic artery hemodynamics.

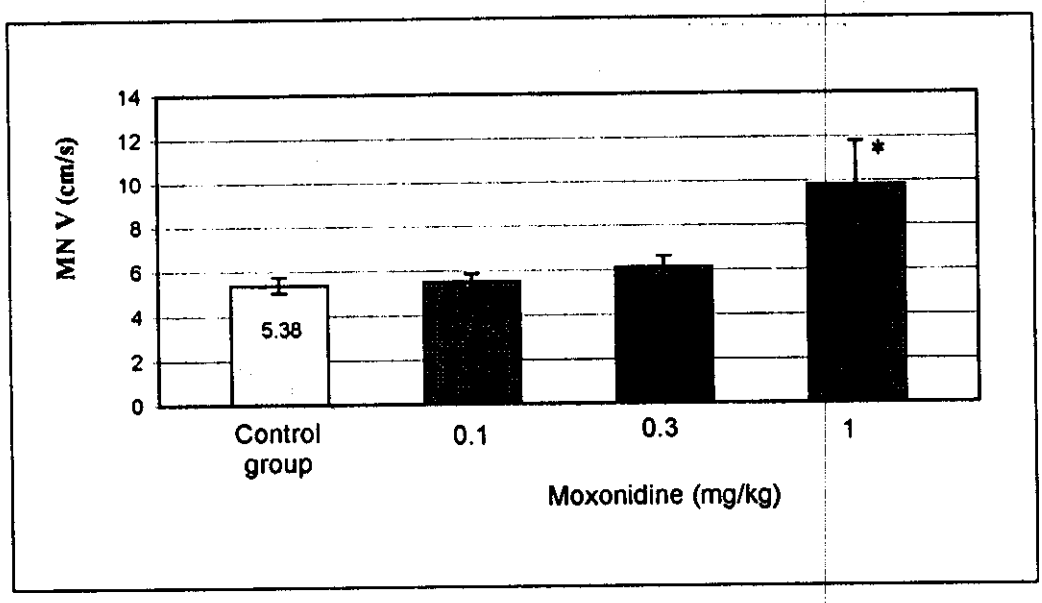


(Maximum Diastole)

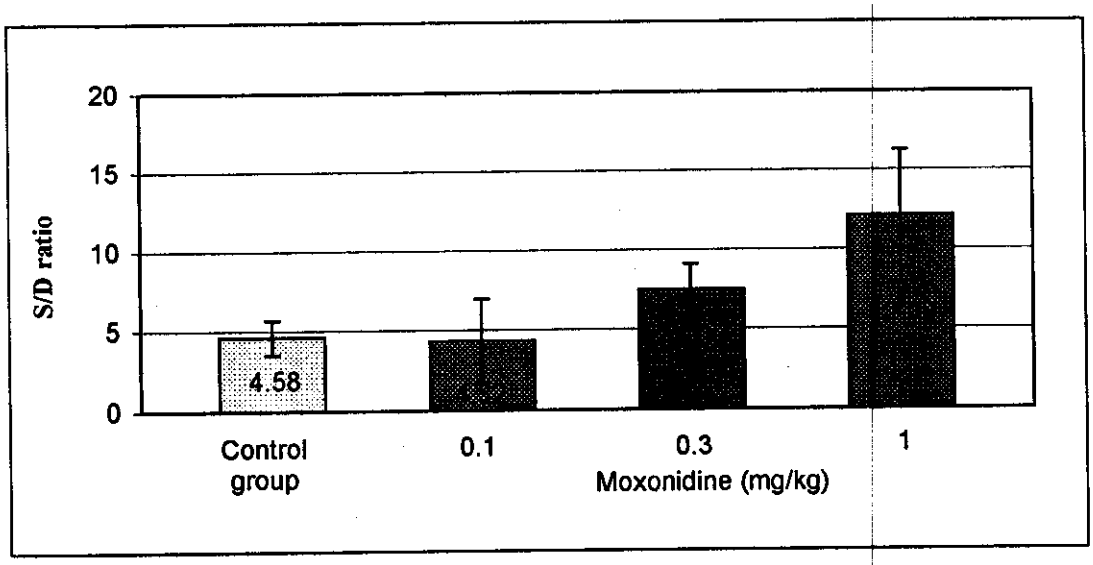


(Minimum Diastole)

Fig (40): Histogram showing the effect of acute administration of moxonidine on maximum and minimum diastolic values of hepatic artery hemodynamics.

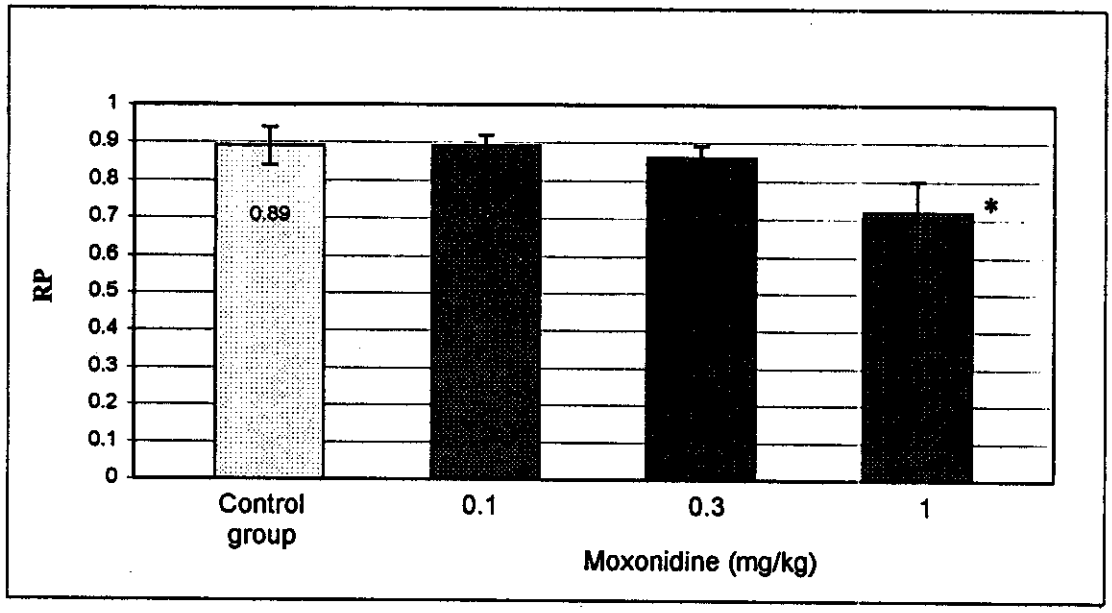


(Mean flow velocity)

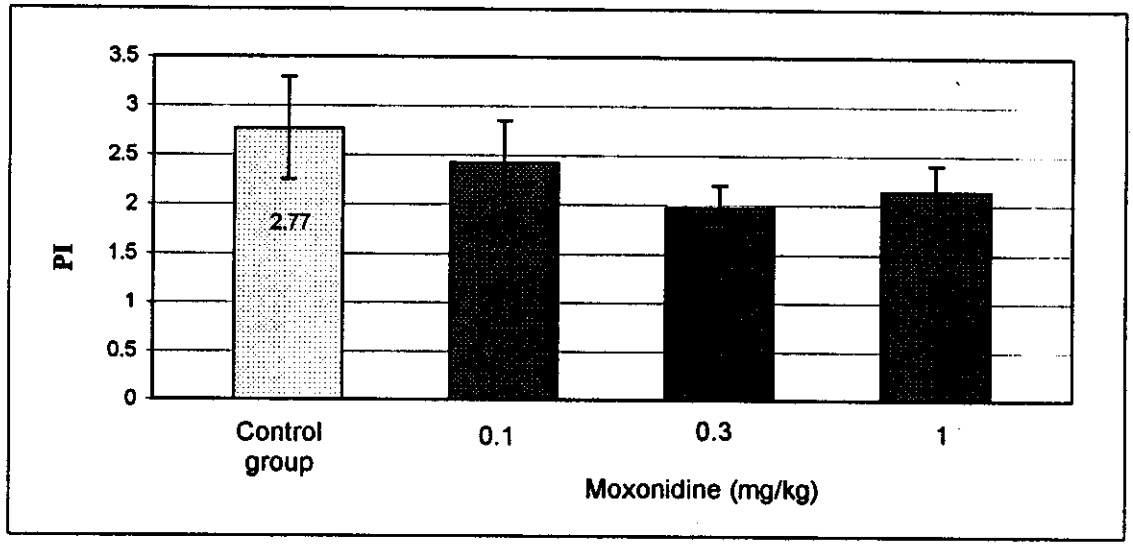


(Systole / Diastole ratio)

Fig (41): Histogram showing the effect of acute administration of moxonidine on hepatic hemodynamic parameters



(Resistance Parameter)



(Pulsatility Index)

Fig (42): Histogram showing the effect of acute administration of moxonidine on hepatic hemodynamic parameters.

3. The prophylactic effect of moxonidine on drug-induced ventricular arrhythmias in anaesthetized rats

3.1 Effect of moxonidine on heart rate in urethane-anaesthetized rats

The heart rate before drug administration was 316.7 ± 20.07, 312.5 ± 12.49 and 304.17 ± 16.09 beat / min in 3 different groups of rats.

Intravenous injection of moxonidine in doses of 0.1, 0.3 and 1 mg/kg produced dose-dependent decrease in heart rate, when recorded after 15 min., to 295.8 ± 18.73, 260.67 ± 13.67 and 232 ± 8.05 beat / min respectively. The decrease in heart rate was found to be statistically insignificant ($P > 0.05$) with the dose of 0.1 mg/kg but was significant ($P < 0.01$) with the doses of 0.3 and 0.1 mg/kg (Table 8, Figs. 44, 45).

Table (8): Effect of acute intravenous injection of different doses of moxonidine on heart rate of urethane-anaesthetized rats.

Dose of moxonidine (mg/kg)	Before moxonidine	After moxonidine	P
	Beat / minute		
0.1	316.7 ± 20.07	295.8 ± 18.73	> 0.05
0.3	312.5 ± 12.49	260.67 ± 13.67	< 0.01*
1	304.17 ± 16.09	232 ± 8.05	< 0.01*

Data represented as mean ± SEM of six experiments.
* Significant at $P < 0.05$.

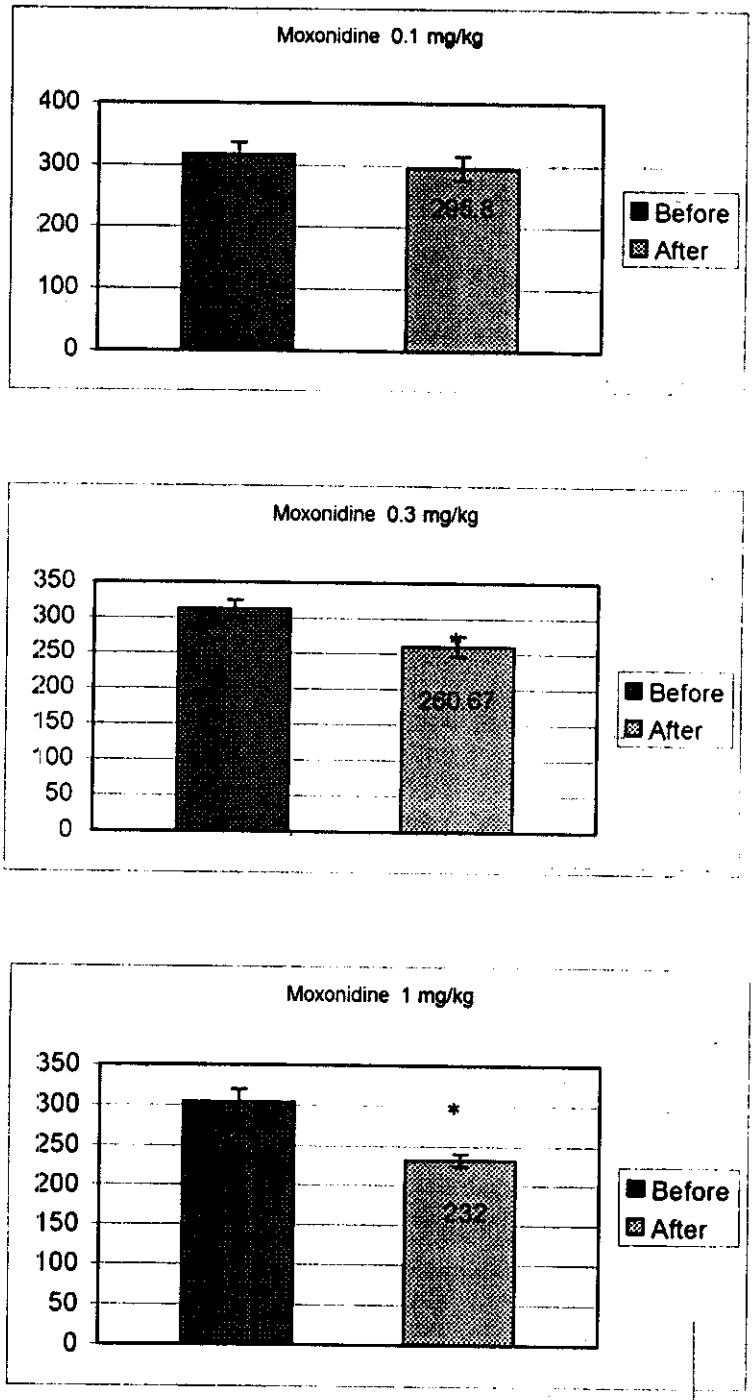
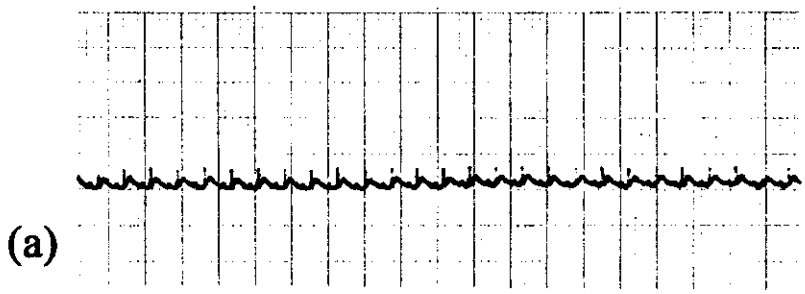
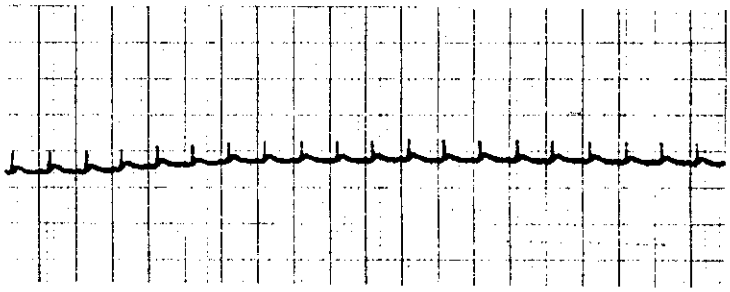


Fig. (44): Histogram showing the effect of acute intravenous injection of moxonidine on heart rate of anaesthetized rats



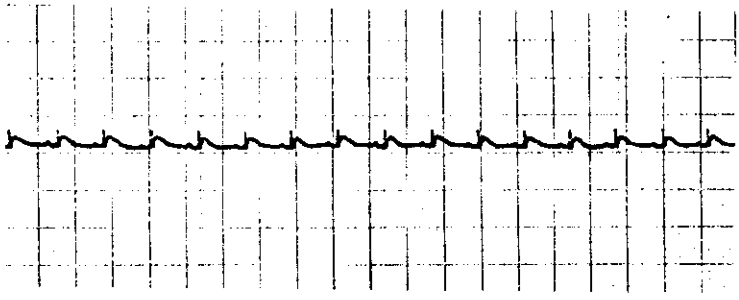
(a)

Control



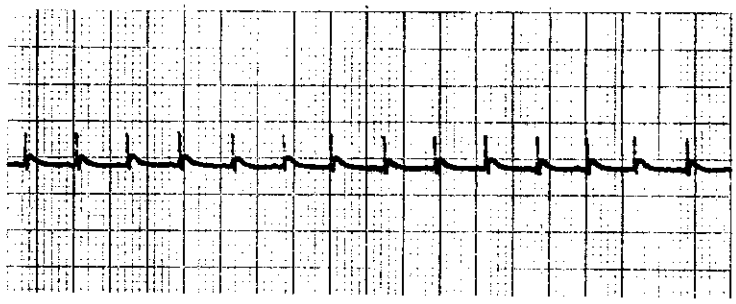
(b)

Moxonidine (0.1 mg /kg).



(c)

Moxonidine (0.3 mg /kg)



(d)

Moxonidine (1 mg /kg)

Fig.(45) ECG traces demonstrating the effect of intravenous injection of moxonidine on heart rate of urethane – anesthetized rats:

(a) Control record with heart rate 375 beats/min.
(b) Moxonidine (0.1 mg/kg) reduced heart rate to 300 beats/ min.
(c) Moxonidine (0.3 mg/kg) reduced heart rate to 214 beats/ min.
(d) Moxonidine (1.0 mg/kg) reduced heart rate to 188 beats/ min.

(0.3 mg/kg) 15 minutes prior to adrenaline injection increased the MAD of adrenaline significantly ($P < 0.01$) to 2.75 ± 0.31 μg/Kg (Table 9, Figs. 46, 51). The onset of adrenaline-induced arrhythmia was significantly ($P<0.01$) delayed to be 17.17 ± 1.42 seconds (Table 10, Fig. 47) and the duration of arrhythmia was significantly ($P < 0.001$) decreased to 22.67 ± 1.36 seconds (Table 11, Fig. 48). The average number of ectopic beats was also decreased to 17.67 ± 2.49 ectopic beat/ minute and this decrease was statistically significant ($P < 0.01$) (Table 12, Fig. 49).

In the third group of rats (group 3), administration of adrenaline intravenously in a mean dose of 1.83 ± 0.31 μg/kg (Table 9, Fig. 46) produced multiple successive ventricular ectopic beats started 10.16 ± 0.95 seconds after adrenaline injection (Table 10, Fig. 47). The average number of ectopic beats was 29.67 ± 3.29 ectopic beat / minute (Table 12, Fig. 49) and the total duration of arrhythmia was 32.5 ± 2.39 seconds (Table 11, Fig. 48).

Moxonidine in a dose (1 mg/kg) injected intravenously 15 minutes prior to intravenous adrenaline injection was found to increase the MAD of adrenaline significantly ($P < 0.01$) to 12.5 ± 2.58 μg/kg (Table 9, Figs. 46, 52). The onset of arrhythmia was significantly ($P < 0.01$) delayed to be 18.17 ± 1.3 seconds (Table 10, Fig. 47). The average number of ectopic beats was significantly ($P< 0.01$) decreased to 2.5 ± 0.56 ectopic beat / minute (Table 12, Fig. 49). Furthermore, the total duration of arrhythmia was significantly ($P < 0.01$) decreased to 5.17 ± 1.7 seconds (Table 11, Fig. 48).

Table (9): Effect of different doses of moxonidine on the minimal arrhythmogenic dose (MAD) of adrenaline in urethane-anaesthetized rats.

Dose of moxonidine (mg/kg)	Minimal arrhythmogenic dose of adrenaline (MAD) µg/kg		P
	Before administration	After administration	
0.1	2 ± 0.32	2.16 ± 0.25	> 0.05
0.3	1.75 ± 0.25	2.75 ± 0.31	< 0.01*
1	1.83 ± 0.31	12.5 ± 2.58	< 0.01*

Table (10): Effect of different doses of moxonidine on the time of onset of adrenaline-induced ventricular arrhythmia in urethane-anaesthetized rats.

Dose of moxonidine (mg/kg)	Onset of arrhythmia (S)		P
	Before administration	After administration	
0.1	12.3 ± 1.28	15.3 ± 1.76	< 0.01*
0.3	11.83 ± 1.27	17.17 ± 1.42	< 0.01*
1	10.16 ± 0.95	18.17 ± 1.3	< 0.01*

Data represented as mean ± SEM of six experiments

* Significant at $P < 0.05$

Table (11): Effect of different doses of moxonidine on the total duration of adrenaline-induced ventricular arrhythmia in urethane-anaesthetized rats.

Dose of moxonidine (mg/kg)	Total duration of arrhythmia (s)		P
	Before administration	After administration	
0.1	30.67 ± 0.84	27.67 ± 1.3	< 0.05*
0.3	32.83 ± 1.7	22.67 ± 1.36	< 0.01*
1	32.5 ± 2.39	5.17 ± 1.7	< 0.01*

Table (12): Effect of different doses of moxonidine on the average number of adrenaline-induced ventricular ectopic beats in urethane-anaesthetized rats.

Dose of moxonidine (mg/kg)	The average number of ventricular ectopic beats (ectopic beat /min.)		P
	Before administration	After administration	
0.1	27.17 ± 2.81	22.83 ± 2.86	< 0.01*
0.3	32.33 ± 2.99	17.67 ± 2.49	< 0.01*
1	29.67 ± 3.29	2.5 ± 0.56	< 0.01*

Data represented as mean ± SEM of six experiments

* Significant at $P < 0.05$

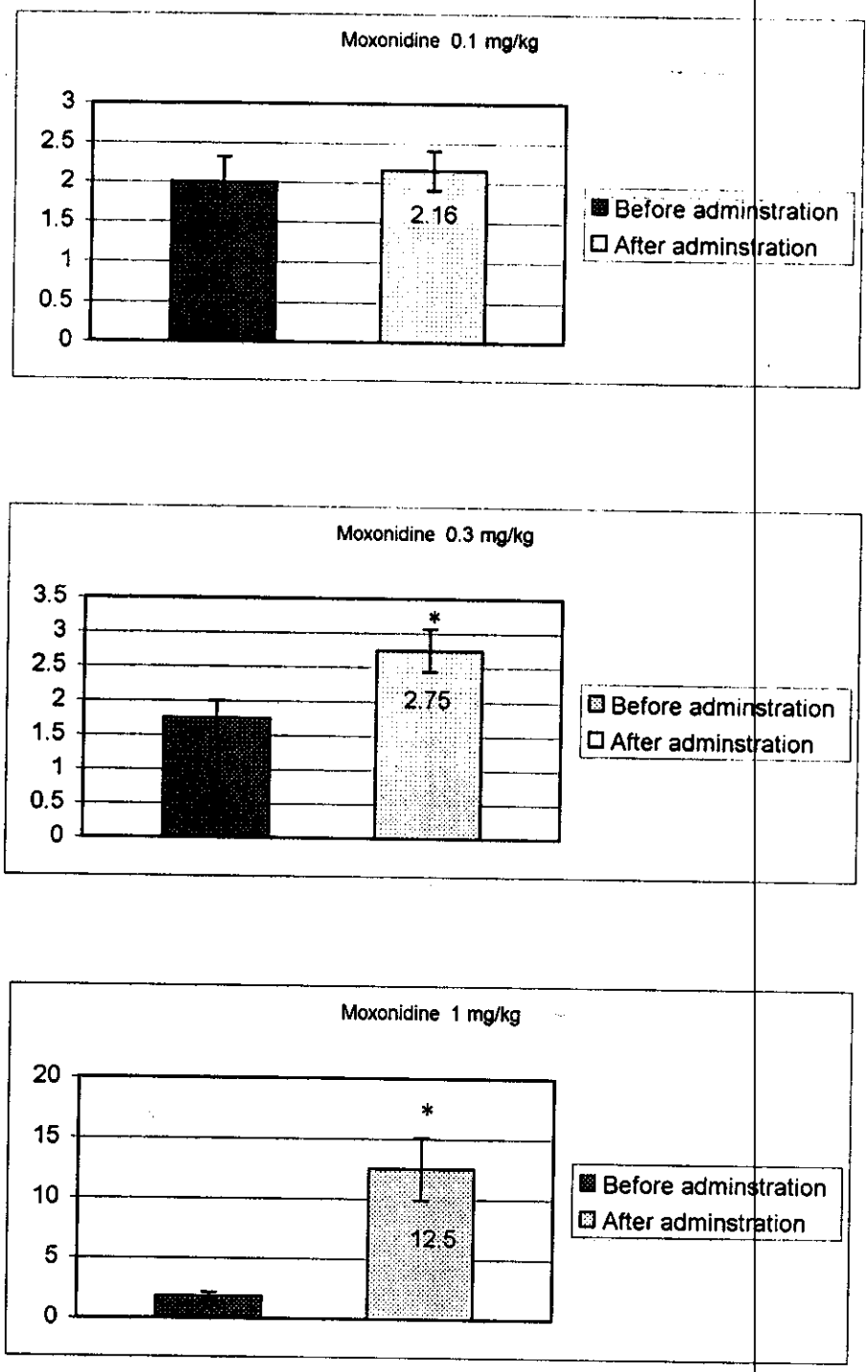


Fig. (46): Histogram showing the effect of moxonidine on arrhythmogenic dose of adrenaline in anaesthetized rats.

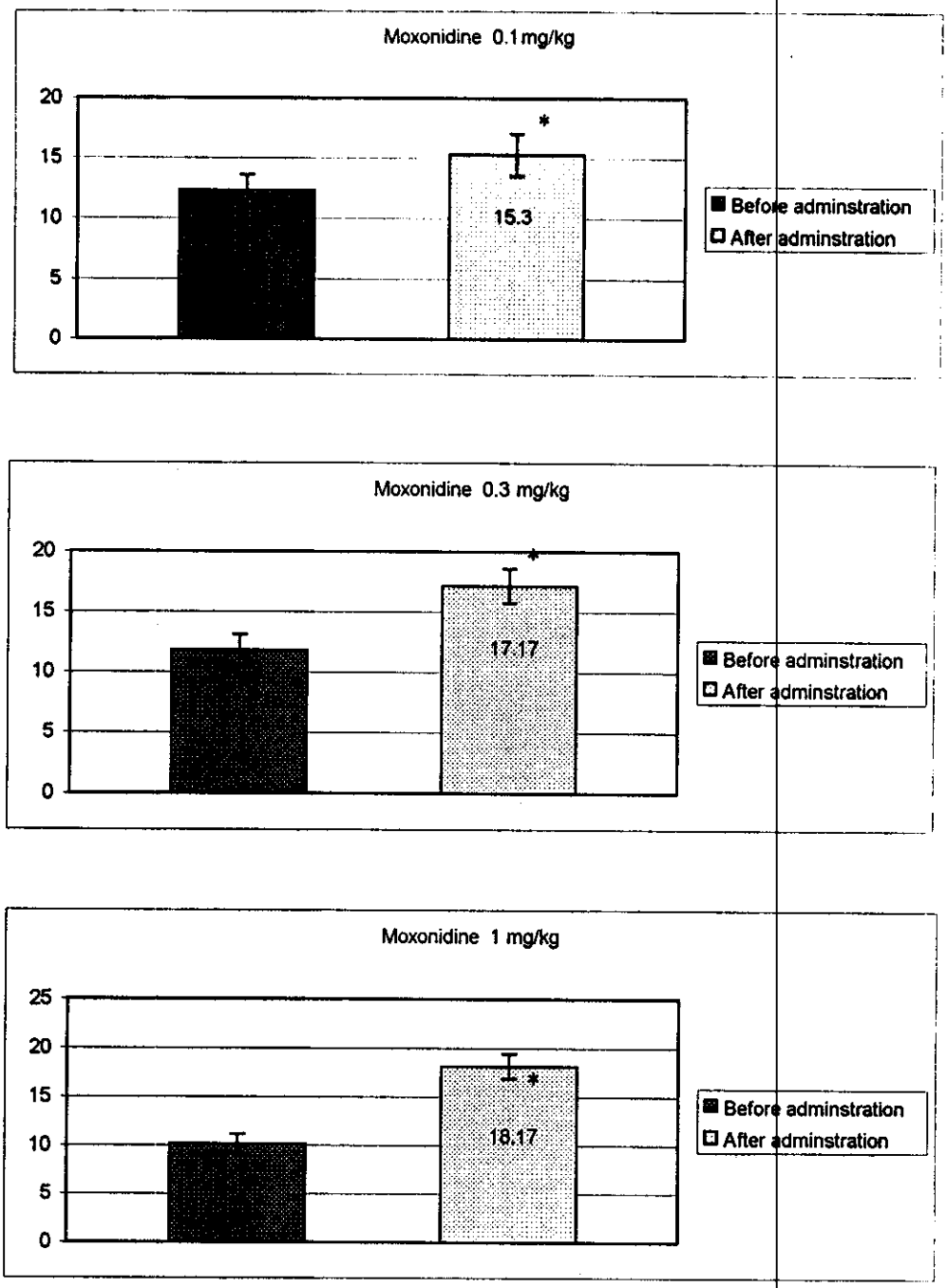


Fig. (47): Histogram showing the effect of different doses of moxonidine on the time of onset of adrenaline-induced ventricular arrhythmia in anaesthetized rats.

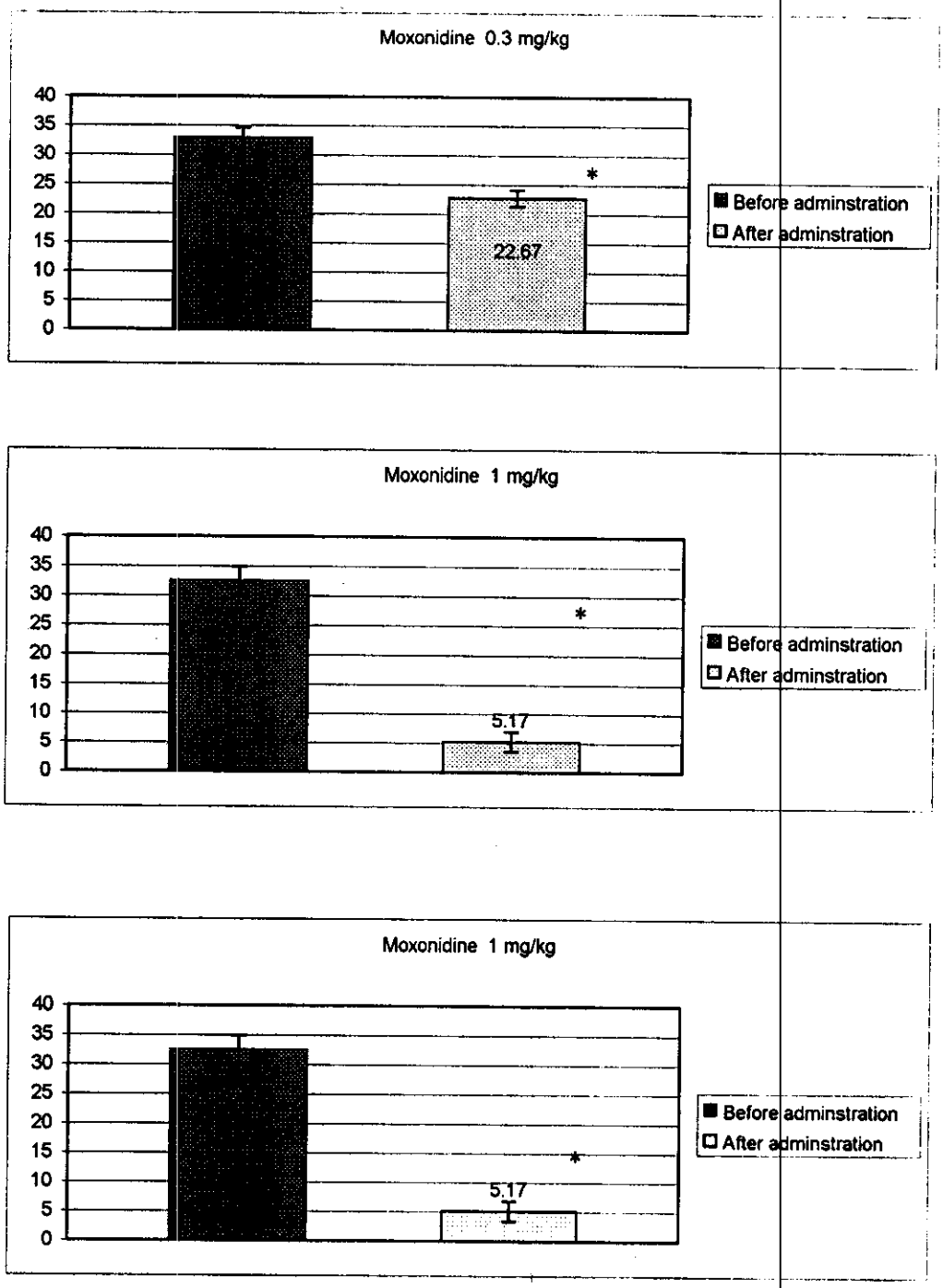


Fig. (48): Histogram showing the effect of different doses of moxonidine on the total duration of adrenaline induced ventricular arrhythmia in anaesthetized rats.

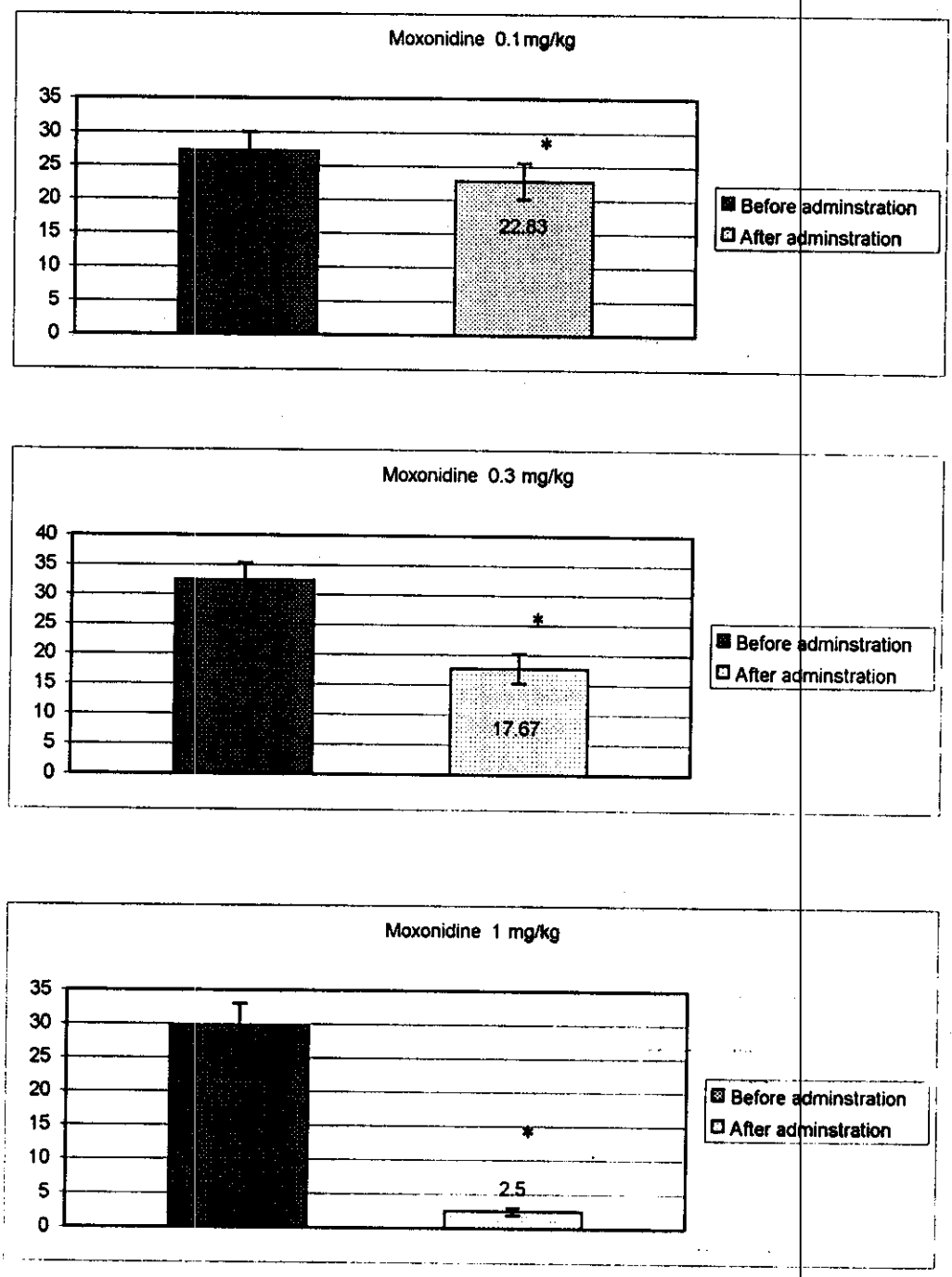


Fig. (49): Histogram showing the effect of different doses of moxonidine on the average number of adrenaline induced ventricular ectopic beats in anaesthetized rats.

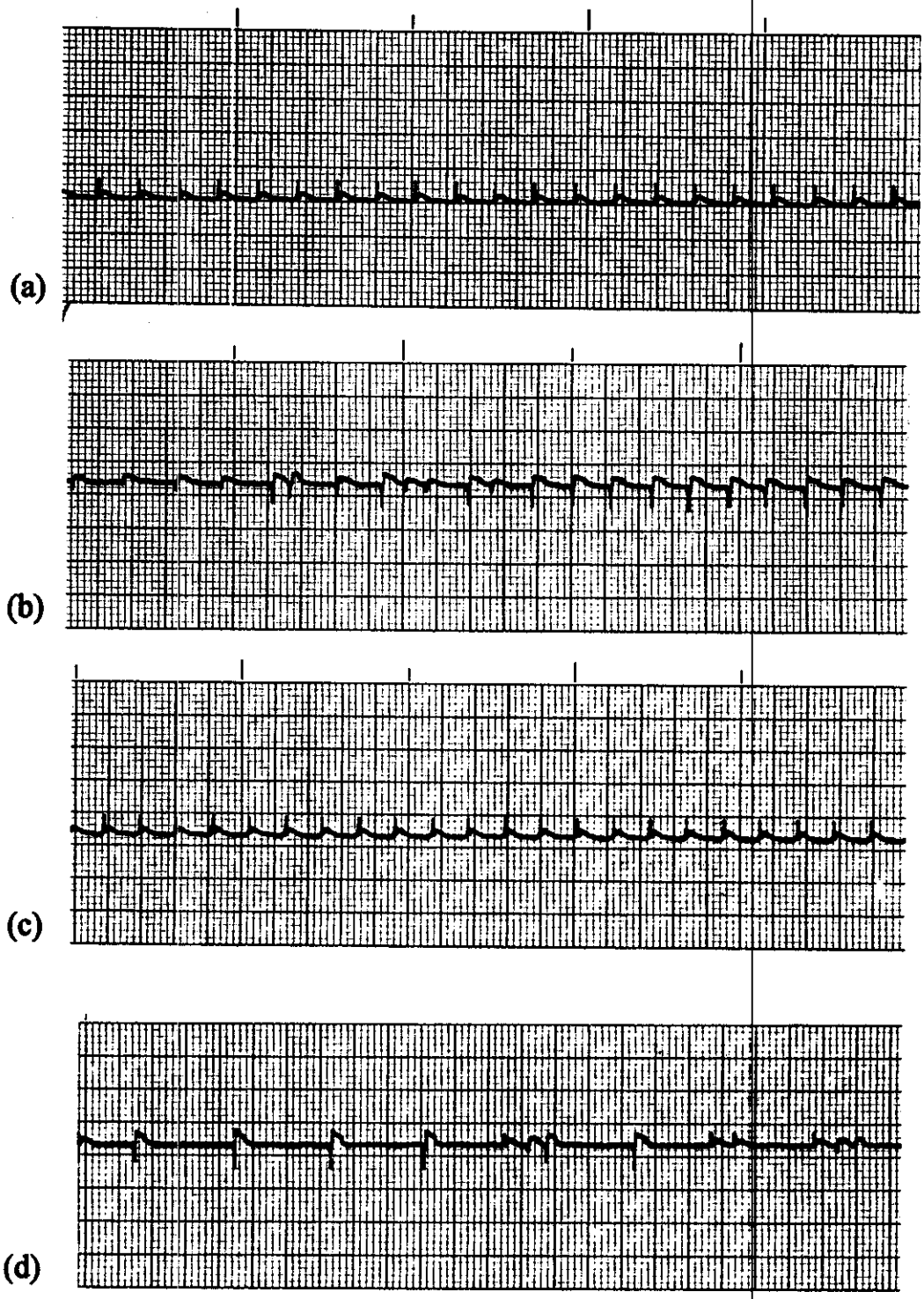


Fig. (50) ECG record showing the effect of moxonidine (0.1 mg / kg I.V.) on the adrenaline – induced ventricular arrhythmia in urethane – anesthetized rats:

(a) Control tracing with heart rate 300 beats /min.
(b) Adrenaline (2ug/kg I.V.) produced successive ventricular extrasystoles
(c) Moxonidine (0.1 mg/kg) injected 15 minutes prior to adrenaline reduced heart rate to 250 beats/ min.
(d) Adrenaline (2ug/kg I.V.) produced successive ventricular extrasystoles.

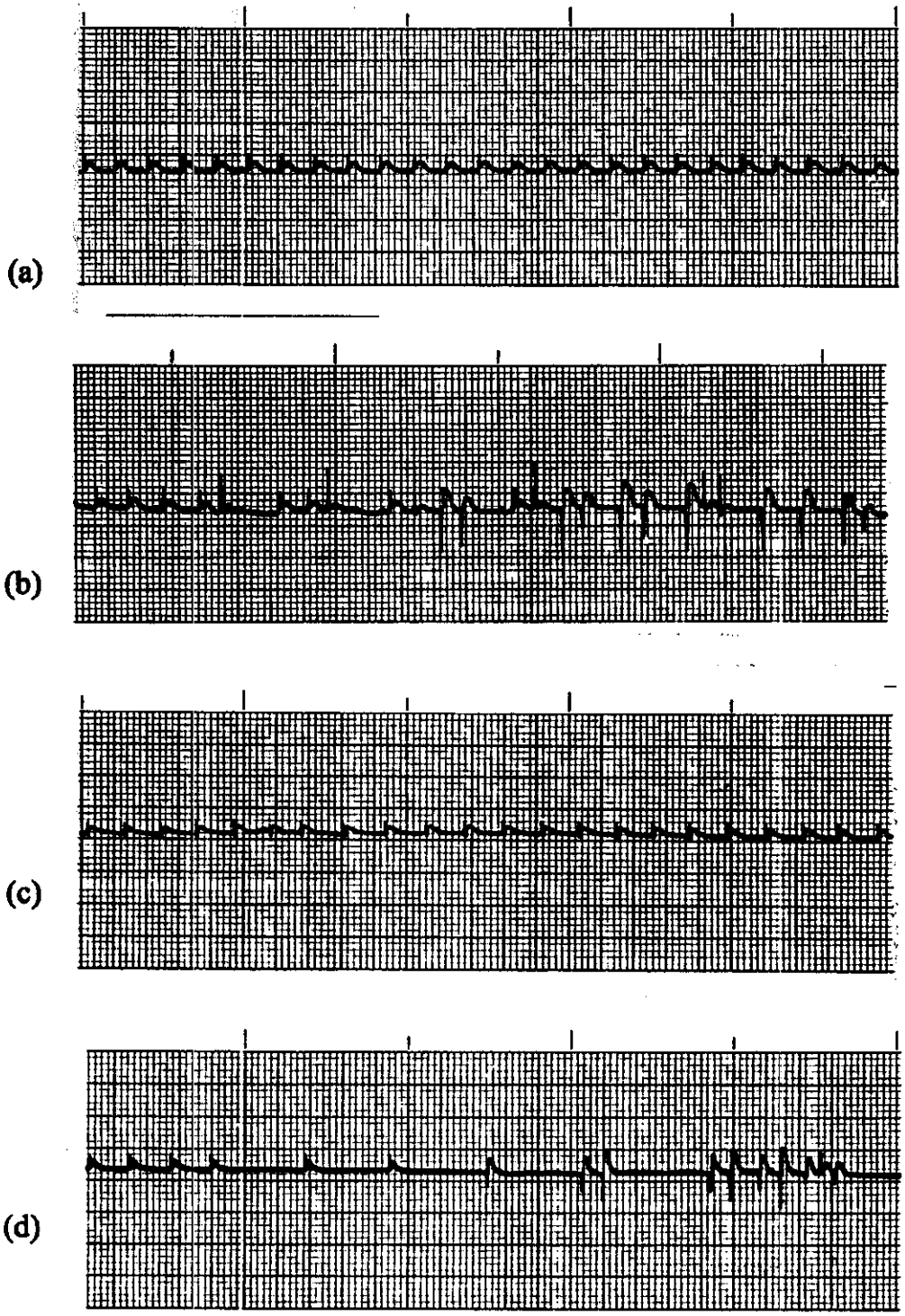


Fig. (51) ECG traces showing the effect of moxonidine (0.3 mg / kg I.V.) on the adrenaline – induced ventricular arrhythmia in urethane – anesthetized rats:

(a) Control record Heart rate 300 beats /min.
(b) Adrenaline (2ug/kg I.V.) produced successive ventricular extrasystoles
(c) Moxonidine (0.3 mg/kg) administered 15 minutes before adrenaline injection reduced heart rate to 214 beats/ min.
(d) Adrenaline (2.75 ug/kg I.V.) produced multiple ventricular extrasystoles.

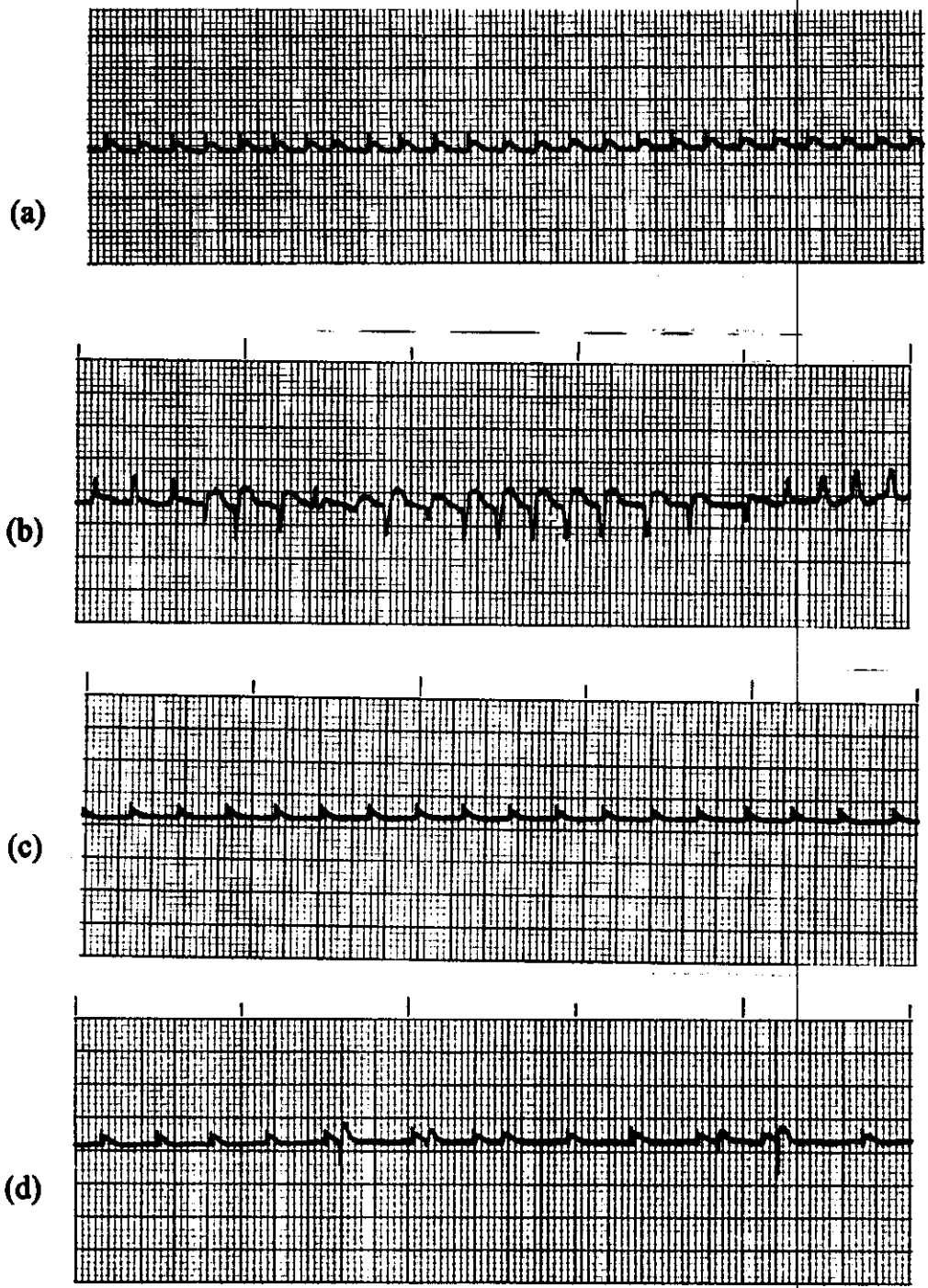


Fig. (52) ECG traces showing the effect of moxonidine (1.0 mg / kg I.V.) on the adrenaline – induced ventricular arrhythmia in urethane – anesthetized rats.

(a) Control record Heart rate 250 beats /min.

(b) Adrenaline (2ug/kg I.V.) produced successive ventricular extrasystoles

(c) Moxonidine (1.0 mg/kg) injected 15 minutes prior to adrenaline administration reduced heart rate to 188 beats/ min.

(d) Adrenaline (12 ug/kg I.V.) failed to produce successive ventricular extrasystoles, only 4 were detected.

3.3 Effect of moxonidine on ouabain-induced ventricular arrhythmia in anaesthetized rats

In control group of rats which were injected with ouabain only, the mean arrhythmogenic dose of ouabain (MAD) was 5.41 ± 1 μg /kg. I.V. (Table 13, Figs. 53, 55) This type of ouabain arrhythmia was in the form of appearance of successive ventricular ectopic beats. The mean dose of ouabain that produced venricular fibrillation and lethality (MFD) was 12.5 ± 1.12 μg/kg (Table 14, Figs. 54, 55).

In moxonidine treated groups, intravenous injection of moxonidine in a dose of 0.1 mg/kg to the first group 15 minute before the start of the repeated ouabain doses produced insignificant ($P > 0.05$) increase in the mean arrhythmogenic dose of ouabain to 7.91 ± 1.19 μg/kg. At the same time, the mean fibrillating dose (MFD) of ouabain was insignificantly ($P > 0.05$) increased to 14.1 ± 1.4 μg/kg compared to control group (Table 13,14, Figs. 53, 54, 56).

On the other hand, pretreatment with moxonidine in a dose of 0.3 mg/kg to the second group 15 minutes prior to ouabain injection, increased the mean arrhythmogenic dose of ouabian significantly ($P < 0.05$) to 12.08 ± 1.4 μg/kg. Also, the mean fibrillating dose of ouabian was increased significantly ($P < 0.05$) to 16.67 ± 1.79 μg/kg compared to control group (Table 13, 14, Figs. 53, 54, 57).

In the third group of rats, moxonidine in a dose of 1 mg/kg injected intravenously 15 minutes before ouabain injection was found to increase the mean arrhythmogenic dose of ouabain (MAD) significantly ($P < 0.01$) to 24.16 ± 3 μg/kg and also increase the mean fibrillating dose of ouabain to 32.5 ± 2.04 μg/kg and this increase was statistically significant ($P < 0.01$) compared to the control group (Table 13, 14, Figs. 53, 54, 58).

Table (13): Effect of moxonidine on arrhythmogenic dose of ouabain in urethane-anaesthetized rats.

Dose of moxonidine (mg/kg)	Arrhythmogenic dose of ouabain (μg/kg)		P
	Control group	Moxonidine-treated group	
0.1	5.41 ± 1	7.91 ± 1.19	> 0.05
0.3		12.08 ± 1.4	< 0.05*
1		24.16 ± 3	< 0.01*

Table (14): Effect of moxonidine on fibrillating dose of ouabain in urethane-anaesthetized rats.

Dose of moxonidine (mg/kg)	Fibrillating dose of ouabain (μg/kg)		P
	Control group	Moxonidine-treated group	
0.1	12.5 ± 1.12	14.1 ± 1.4	> 0.05
0.3		16.67 ± 1.79	< 0.05*
1		32.5 ± 2.04	< 0.01*

Data represented as mean ± SEM of six experiments

* Significant at $P < 0.05$

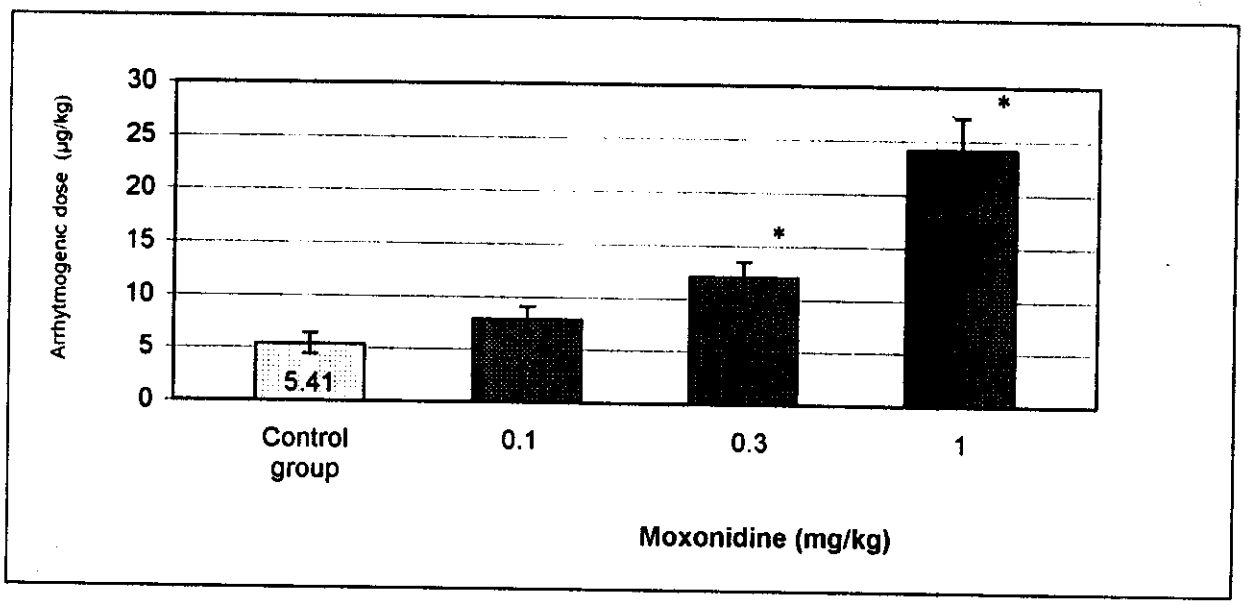


Fig. (53): Histogram showing the effect of moxonidine on arrhythmogenic dose of ouabain in anaesthetized rats.

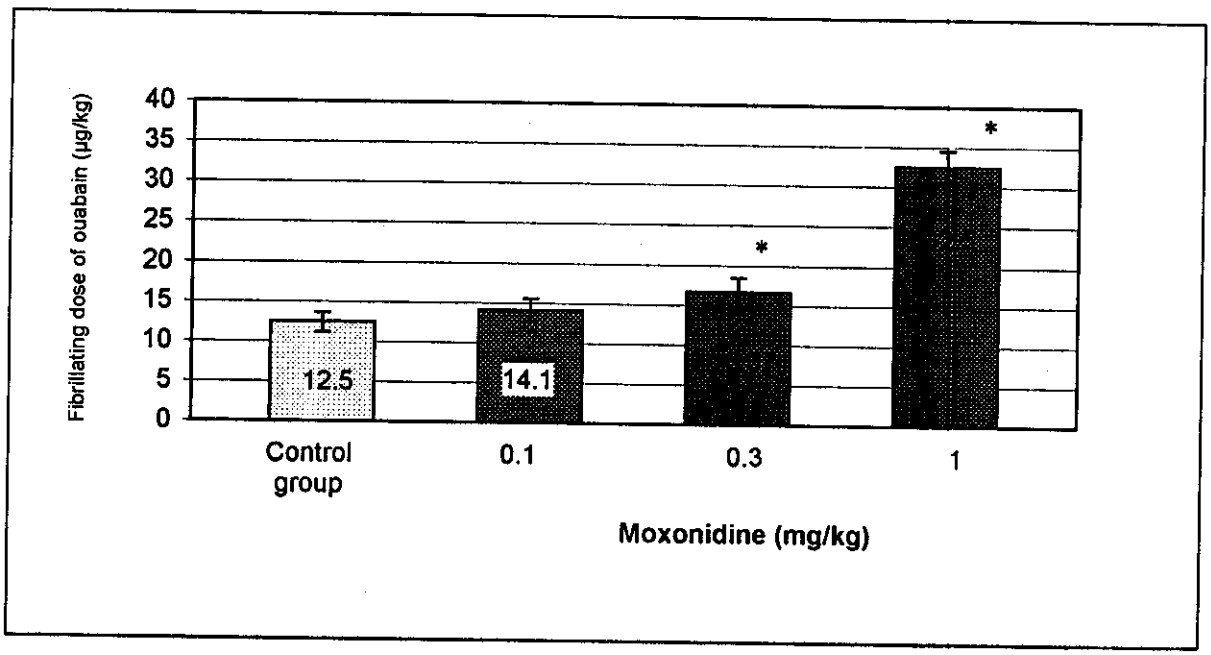


Fig. (54): Histogram showing the effect of moxonidine on fibrillating dose of ouabain in anaesthetized rats.

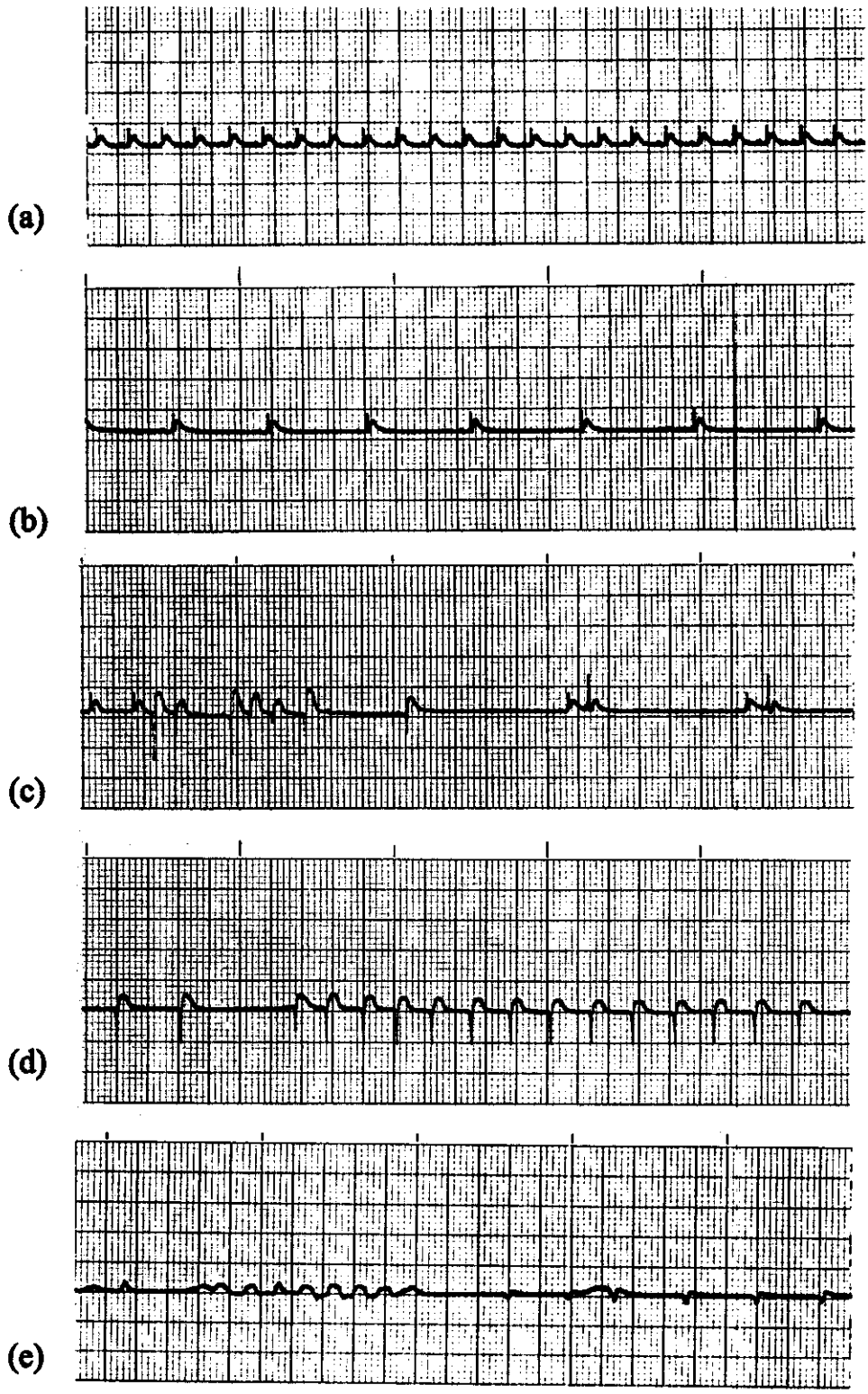


Fig. (55) A trace showing the effect of repeated intravenous doses of ouabain on the ECG of urethane – anesthetized rats (Control group):

(a) Normal tracing , heart rate 300 beats/min.
(b) Ouabain (0.5 ug/ kg) reduced heart rate to 83 beats/min.
(c) Ouabain (5 ug/ kg) produced multiple ventricular extrasystoles.
(d) Ouabain (7 ug/ kg) produced successive ventricular extrasystoles.
(e) Ouabain (10 ug/ kg) produced ventricular fibrillation and death.

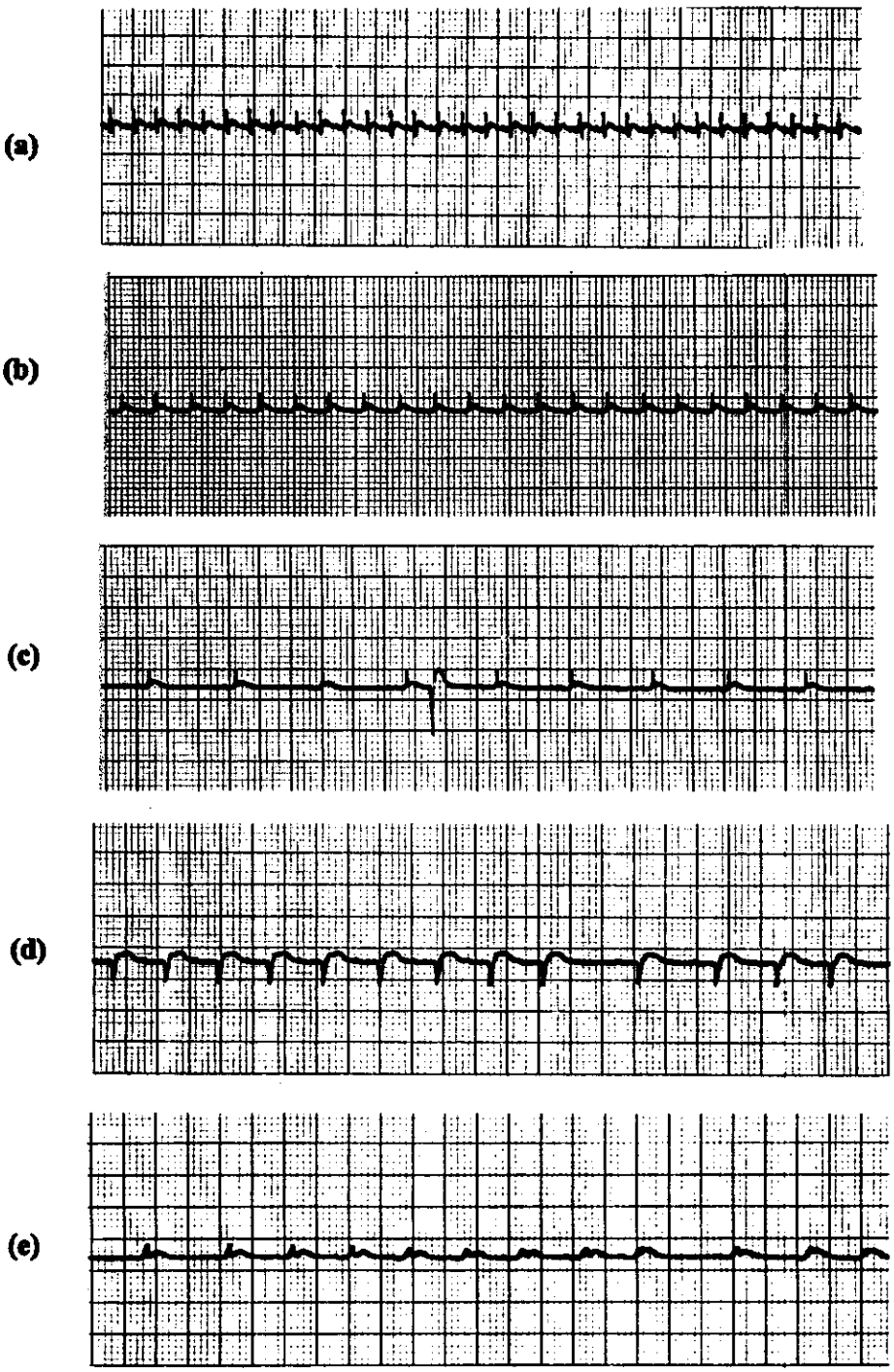


Fig. (56) ECG traces showing the prophylactic effect of moxonidine (0.1 mg / kg) on ouabain – induced arrhythmia in urethane – anesthetized rats:

a. **Control tracing with heart rate 375 beats /min.**
b. **Moxonidine (0.1 mg/kg I.V.) was injected 15 minutes prior to Ouabian injection reduced heart rate to 250 beats/ min.**
c. **Ouabain (5 ug/ kg I.V.) produce one ventricular extrasystole with heart rate 115 beats /min.**
d. **Ouabain (7 ug/ kg I.V.) produced multiple ventricular successive extrasystoles.**
e. **Ouabain (10 ug/ kg I.V.) produced ventricular fibrillation and death.**

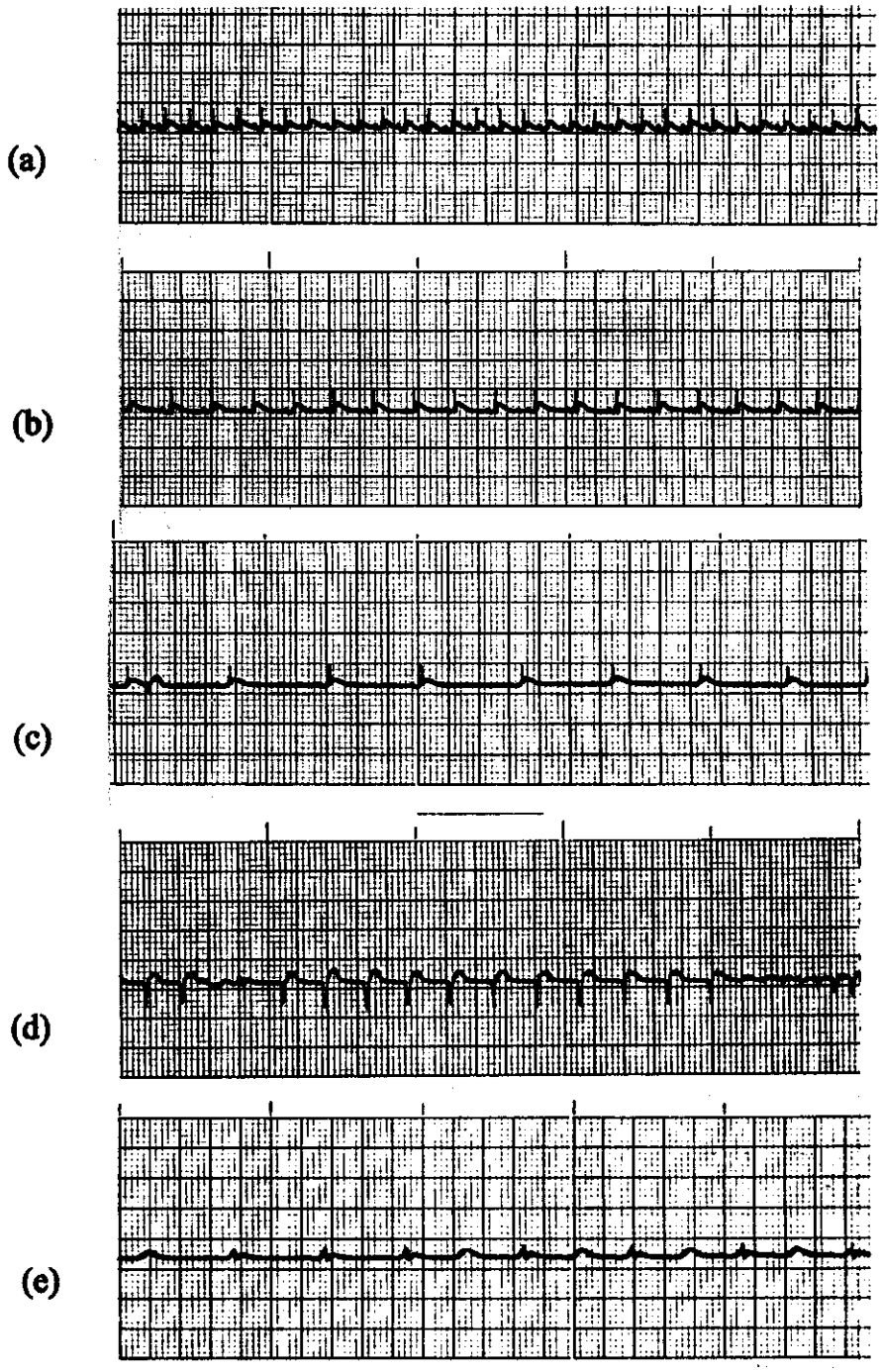


Fig. (57) ECG traces showing the prophylactic effect of moxonidine (0.3 mg / kg) on ouabain – induced arrhythmia in urethane – anesthetized rats:

(a) Control record with heart rate 375 beats /min.
(b) Moxonidine (0.3 mg/kg I.V.) was injected 15 minutes prior to Ouabain injection reduced heart rate to 214 beats/ min.
(c) Ouabain (5 ug/ kg I.V.) failed to produce ventricular extrasystole, reduced heart rate to 107 beats/ min.
(d)Ouabain (10 ug/ kg I.V.) produced multiple successive ventricular successive extrasystoles.
(e) Ouabain (15 ug/ kg I.V.) produced ventricular fibrillation and death.

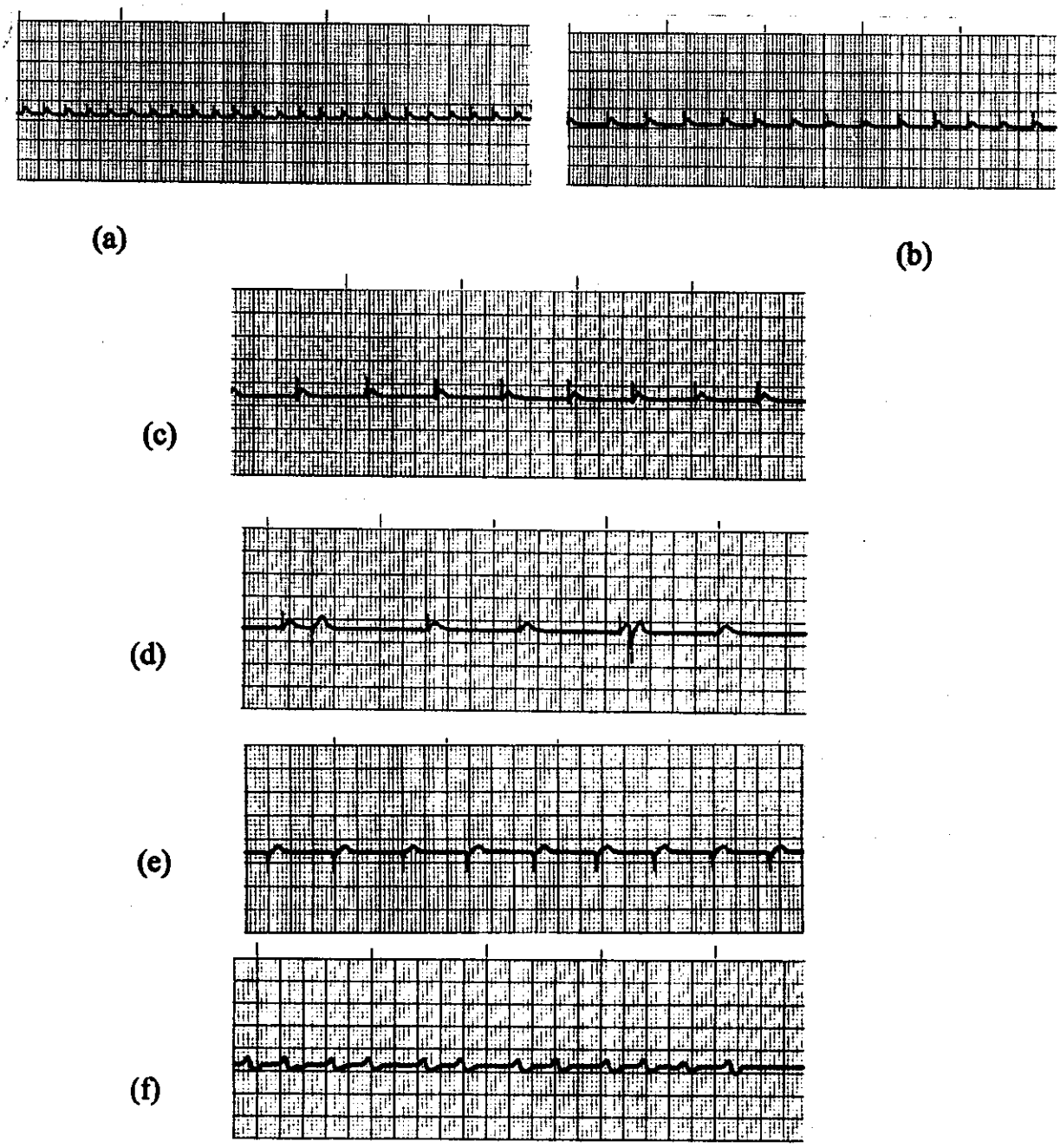


Fig. (58) ECG traces showing the prophylactic effect of moxonidine (1.0 mg / kg) on Ouabain – induced arrhythmia in urethane – anesthetized rats:

(a) Control record with heart rate 300 beats /min.
(b) Moxonidine (1.0 mg/kg I.V.) was injected 15 minutes prior to Ouabain injection reduced heart rate to 167 beats/ min.
(c) Ouabain (5 ug/ kg I.V.) failed to produce ventricular extrasystole, heart rate was decreased to 100 beats/ min.
(d) Ouabain (10 ug/ kg I.V.) produced only 2 ventricular extrasystoles.
(e) Ouabain (20 ug/kg I.V.) produced multiple ventricular successive extrasystoles.
(f) Ouabain (30 ug/ kg I.V.) produced ventricular fibrillation and death

4. The acute analgesic effect of moxonidine in rats

The mean basal forces (g) which were necessary to make the rats respond to nociceptive stimuli were 335.83 ± 16.03, 351.83 ± 15.08 and 346.17 ± 14.15 g for the various doses of moxonidine 0.1, 0.3 and 1 mg / kg, I.P. respectively in 3 different group of rats.

After 15 minute of moxonidine administration the mean forces became 337.0 ± 12.64, 420.33 ± 16.2 and 828.67 ± 68.7 g for the doses of 0.1, 0.3 and 1mg / kg, I.P. of moxonidine respectively.

After 30 minute the mean forces increased to be 371.5 ± 6.89, 558.83 ± 26.49 and 1070.83 ± 26.39 g for the doses of 0.1, 0.3 and 1 mg/kg, I.P. of moxonidine respectively.

After 45 minute the mean forces also increased to be 427.67 ± 17.71, 835.17 ± 70.24 and 1181.83 ± 31.5 g for the doses of 0.1, 0.3 and 1mg/kg, I.P of moxonidine respectively.

After 60 minute the mean forces still high to be 463.33 ± 19.52, 1182.33 ± 74.03 and 1313.5 ± 23.27 g for the dose of 0.1, 0.3 and 1 mg /kg, I.P of moxonidine respectively.

Thus, moxonidine produced an increase in the mean forces in gradual manner over the time period (60 min.). This increase was statistically significant for the dose of 0.1 mg/kg, I.P. ($P < 0.01$) and the doses of 0.3 and 1mg/kg, I.P ($P < 0.001$) in references to duration (Table 15, Fig. 59).

Table (15): The acute antinociceptive effect of different doses of moxonidine in rats.

Time (min) / Dose of Moxonidine (mg/kg)	0	15	30	45	60	P
	The mean force (g) ± SEM					
0.1	335.83 ± 16.03	3[illegible]7.0 ± 12.64	371.5 ± 6.89	427.67 ± 17.71	463.33 ± 19.52	<0.01*
0.3	351.83 ± 15.08	420.33 ± 16.2	558.83 ± 26.49	835.17 ± 70.24	1182.33 ± 74.03	<0.001*
1.0	346.17 ± 14.15	828.67 ± 68.7	1070.83 ± 26.39	1181.83 ± 31.5	1313.5 ± 23.27	<0.001*

Data represented as mean ± SEM of six experiments

* Significant at $P < 0.05$

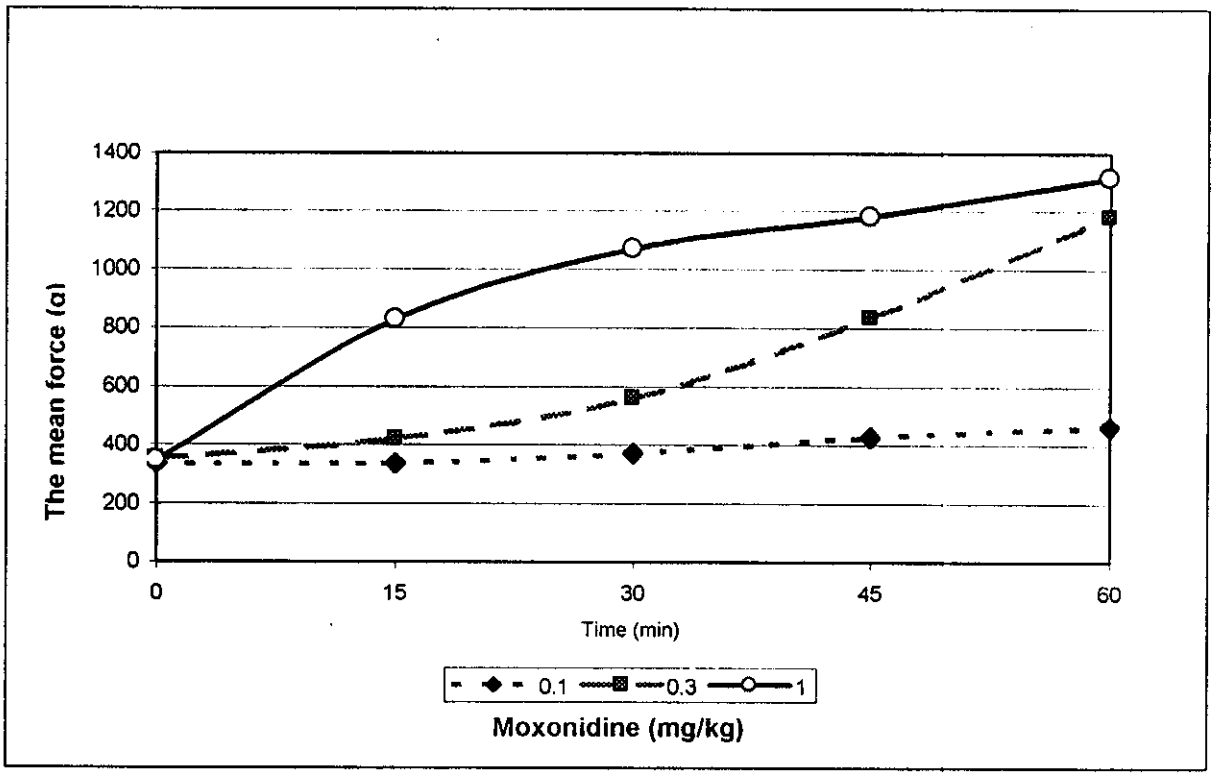


Fig. (59): Histogram showing the acute antinociceptive effect of different doses of moxonidine in rats.

Effect of yohimbine (α_2-adrenoceptor antagonist) (1mg/kg,I.P) pretreatment on the analgesic effect of moxonidine in rats

The mean basal forces (g) which were necessary to make the rats respond to nociceptive stimuli were 335.83 ± 16.03, 351.83 ± 15.08 and 346.17 ± 14.15 g for the various doses of moxonidine 0.1, 0.3 and 1 mg/kg. I.P. respectively in 3 different group of rats.

After 15 minute of moxonidine administration, in rats pretreated with yohimbine (1mg/kg I.P) 15 minutes prior to moxonidine injection, the mean forces became 328.5 ± 18.27, 354.33 ± 12.58 and 504 ± 6.39 g for the doses of 0.1, 0.3 and 1mg/kg, I.P. of moxonidine respectively.

After 30 minute the mean forces became 333.0 ± 17.76, 396.0 ± 6.09 and 539.17 ± 10.49 g for the doses of 0.1, 0.3 and 1mg/kg I.P. of moxonidine respectively.

After 45 minute the mean forces became 336.33 ± 16.46, 541.33 ± 6.12 and 739.83 ± 12.46 g for the doses of 0.1, 0.3 and 1mg/kg, I.P. of moxonidine respectively.

After 60 minute the mean forces increased to be 344.17 ± 18.14, 706.33 ± 7.24 and 887.67 ± 7.42 g for the doses of 0.1, 0.3 and 1mg /kg, I.P. of moxonidine respectively.

Comparing the mean forces (g) after different doses of moxonidine (0.1, 0.3 and 1mg/kg, I.P.) with those after yohimbine (1mg/kg, I.P.) pretreatment, it was found that yohimbine significantly ($P < 0.05$) and completely antagonized the analgesic effect of moxonidine in a dose of 0.1 mg/kg, and also significantly ($P < 0.01$) decreased the analgesic effect of the doses of 0.3 and 1mg/kg (Table 16, Fig. 60).

Table (16): Effect of yohimbine pretreatment on the analgesic effect of different doses of moxonidine in rats.

Dose of moxonidine (mg/kg)	Time (min.)	Moxonidine	Moxonidine after yohimbine	P
		The mean force (g) ± SEM		
0.1	0	335.83 ± 16.03		-
	15	337.0 ± 12.64	328.5 ± 18.27	> 0.05
	30	371.5 ± 6.89	333.0 ± 17.76	< 0.05*
	45	427.67 ± 17.71	336.33 ± 16.46	< 0.05*
	60	463.33 ± 19.52	344.17 ± 18.14	< 0.01*
0.3	0	351.83 ± 15.08		-
	15	420.33 ± 16.2	354.33 ± 12.58	< 0.01*
	30	558.83 ± 26.49	396.0 ± 6.09	< 0.01*
	45	835.17 ± 70.24	541.33 ± 6.12	< 0.01*
	60	1182.33 ± 74.03	706.33 ± 7.24	< 0.01*
1	0	346.17 ± 14.15		-
	15	828.67 ± 68.7	504.0 ± 6.39	< 0.01*
	30	1070.83 ± 26.39	539.17 ± 10.49	< 0.001*
	45	1181.83 ± 31.5	739.83 ± 12.46	< 0.001*
	60	1313.5 ± 23.27	887.67 ± 7.42	< 0.001*

Data represented as mean ± SEM of six experiments

* Significant at $P < 0.05$

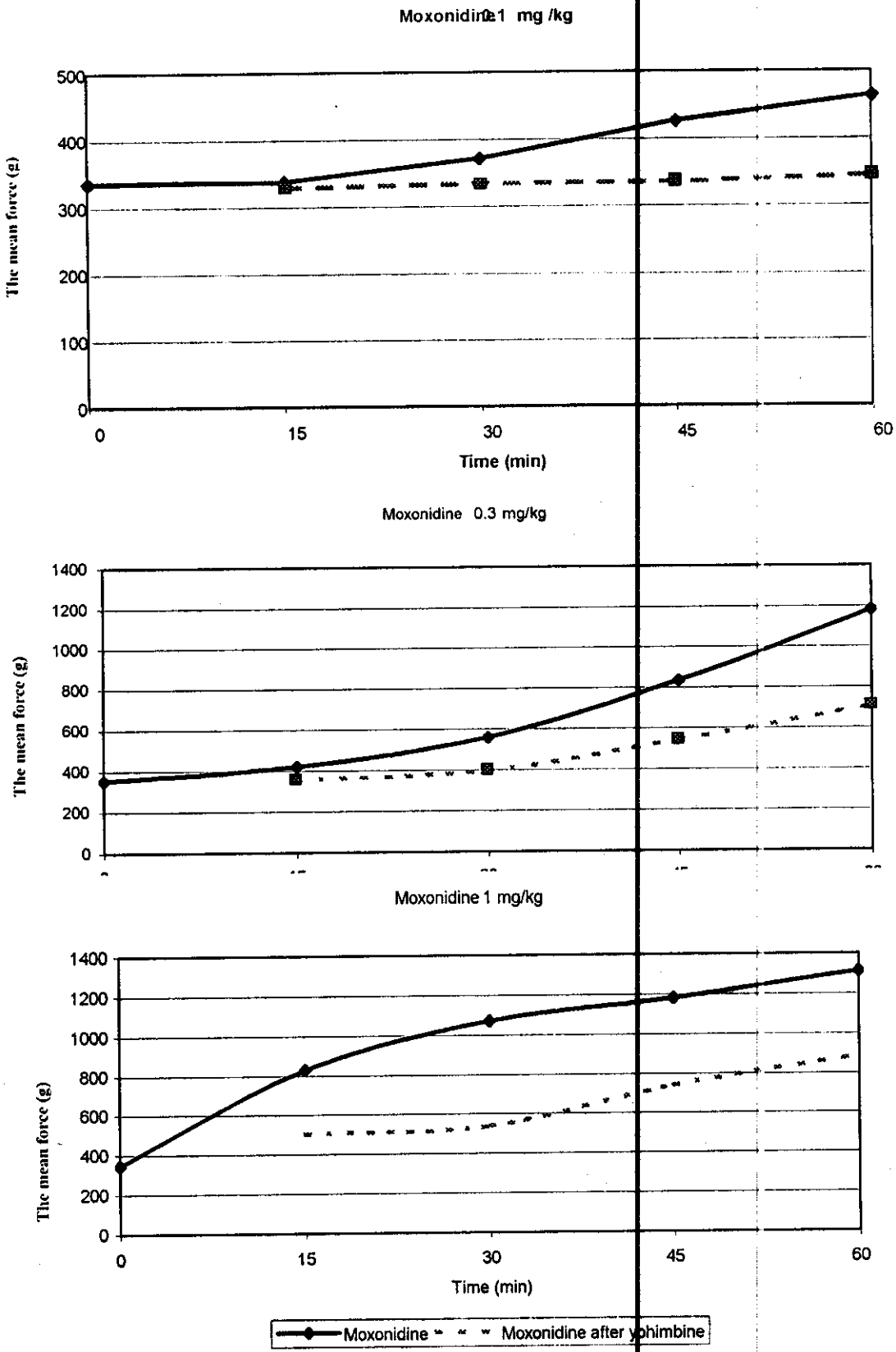


Fig. (60): Histogram showing the effect of yohimbine pretreatment on the antinociceptive effect of different doses of moxonidine.

II. In vitro Experiments

1. Effect of moxonidine on isolated perfused rabbit's heart

It was noticed that moxonidine in increasing doses (1, 3, 10, 30 and 100 μg/ml) produced inhibition of the force of contraction of the isolated perfused rabbits heart in a dose-dependent manner (Figs. 61, 62).

This inhibition of the force of spontaneous contraction of the isolated perfused rabbit heart was statistically insignificant with the doses of 1 and 3 μg/ml with percentage of reduction 0.53 and 11.97 % respectively. Meanwhile, it was significant ($P < 0.01$) with the doses of 10 and 30 μg/ml with percentage of reduction 33.49 and 57.75 % respectively, and significant ($P < 0.001$) with the dose of 100μg/ml with percentage or reduction 76.58 % compared to basal value (Table 17).

Site of action of moxonidine on isolated perfused rabbit heart

It was found that moxonidine added in a dose of 30 μg/ml produced inhibitory effect on the spontaneous rhythmic contractility of the isolated rabbit heart. This inhibitory effect was completely abolished after the addition of atropine in a dose of 10 μg/ml and also after the addition of nicotine in a large dose of 100 μg/ml (Fig. 63).

Table (17): Effect of moxonidine on the amplitude of spontaneous contraction of isolated perfused rabbit heart.

Dose of moxonidine (μg/ml)	Level of contraction before /cm	Level of contraction after /cm		% of Reduction	P
1	5.68 ± 0.14	5.65 ± 0.12		0.53	> 0.05
3		5.47 ± 0.14		11.97	> 0.05
10		3.78 ± 0.09		33.45	< 0.01*
30		2.4 ± 0.21		57.75	< 0.01*
100		1.33 ± 0.03		76.58	< 0.001*

Data represented as mean ± SEM of six experiments

* Significant at P < 0.05

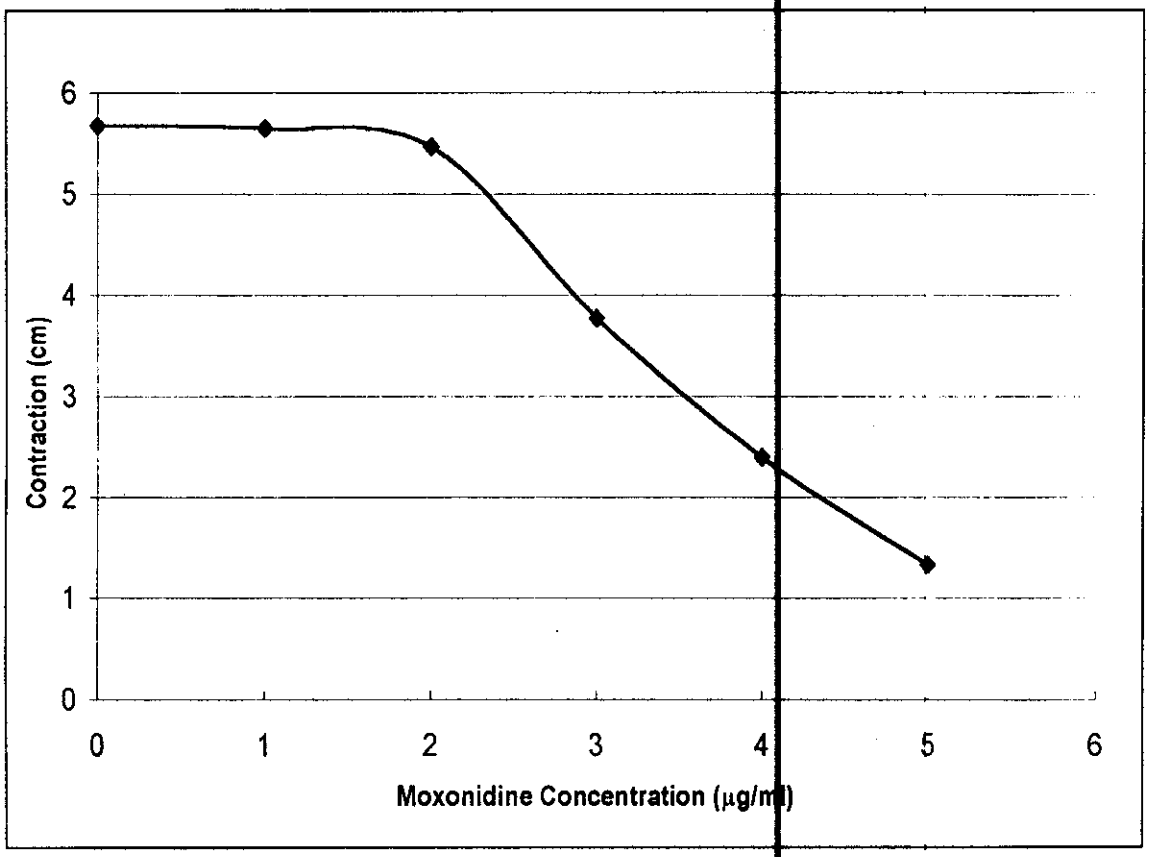


Fig. (61): Histogram showing the effect of different doses of moxonidine on the amplitude of spontaneous contraction of isolated rabbit's heart.

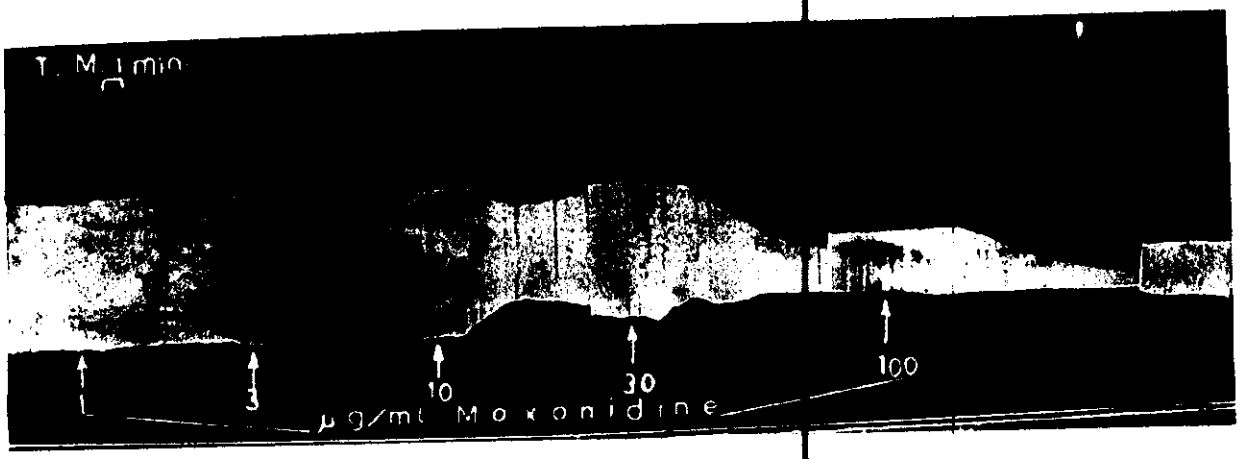


Fig. (62): A record demonstrating the effect of gradually increasing concentration of moxonidine on the isolated perfused rabbit's heart.

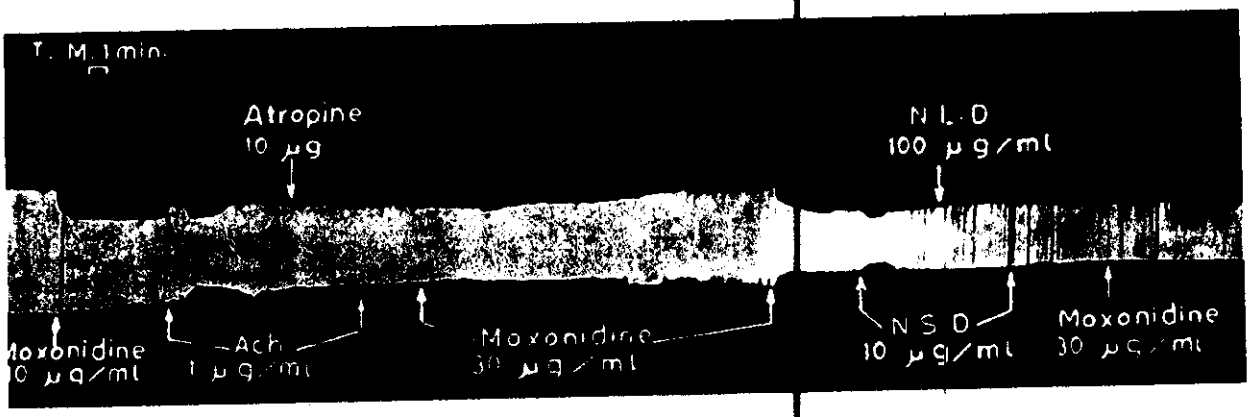


Fig. (63): A record showing the site of action of moxonidine on the isolated perfused rabbit's heart.

2. Effect of moxonidine on isolated rabbit's aortic spiral strip

It was observed that addition of moxonidine in different dose levels (0.1, 0.3, 1, 3, 10, 30 and 100 μg/ml) produced dose related contraction of the rabbit's aortic spiral strip (Table 18, Fig. 65).

It was found that preincubation of yohimbine (1μg/ml) for 5 minutes prior to addition of different doses of moxonidine (0.1, 0.3, 1, 3, 10, 30 and 100 μg/ml) produced significant ($P < 0.05$) reduction of moxonidine-induced contraction of the isolated rabbit's aortic strip by 85.5 %, 68.2 %, 50%, 54.9 %, 57.28 %, 51.6% and 51.6 % respectively (Table 18, Fig. 66).

Thus, yohimbine produced right shift of the dose response curve of moxonidine (Fig. 64).

Meanwhile, preincubation of prazosin (1.0 μg/ml) for 5 minutes prior to addition of different doses of moxonidine produced insignificant ($P > 0.05$) reduction in moxonidine induced contraction of isolated aortic strip. The percentage of reduction were 32.7 %, 32.4%, 16.8%, 22.1%, 14.3%, 8.4% and 10.5 % for the doses of 0.1, 0.3, 1, 3, 10, 30 and 100 μg/ml respectively (Table 18, Figs. 64, 67). Thus, prazosin produced slight right shift of the dose response curve of moxonidine (Fig. 64).

Table (18): Effect of moxonidine on the isolated rabbit's aortic strip before and after yohimbine and prazosin.

Moxonidine µg/ml	Original response (cm)	Response after yohimbine (1µg)			Response after prazosin (1µg/ml)		
		(cm)	% of reduction	P	(cm)	% of reduction	P
0.1	0.55±0.03	0.08±0.05	85.50	<0.05	0.37±0.04	32.7	>0.05
0.3	1.48±0.45	0.47±0.03	68.20	<0.05	1.0±0.06	32.4	>0.05
1	2.2±0.46	1.1±0.17	50.00	<0.05	1.83±0.32	16.8	>0.05
3	3.17±0.49	1.43±0.03	54.90	<0.05	2.47±0.23	22.1	>0.05
10	4.05±0.31	1.73±0.09	57.28	<0.05	3.47±0.07	14.3	>0.05
30	4.07±0.16	1.97±0.12	51.60	<0.05	3.73±0.12	8.4	>0.05
100	4.55±0.13	2.2±0.15	51.60	<0.05	4.0±0.09	10.5	>0.05

Data represented as mean ± SEM of six experiments

* Significant at $P < 0.05$

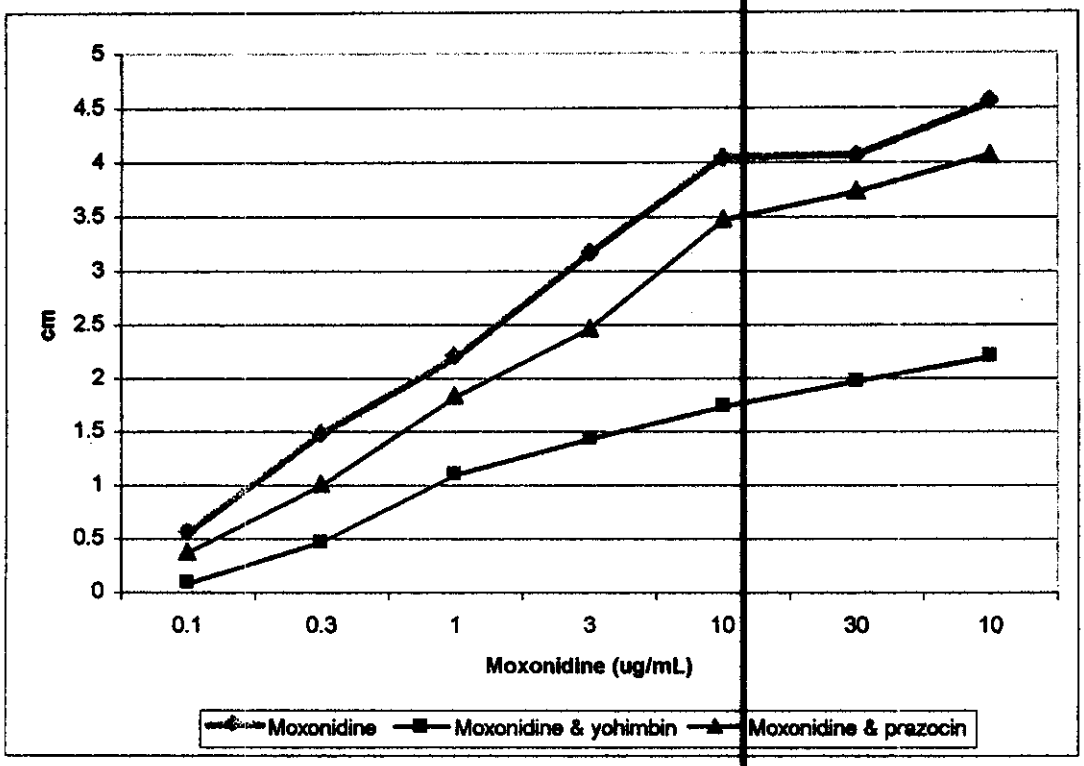


Fig. (64): Histogram showing the effect of yohimbine and prazosin on moxonidine induced contraction of isolated rabbit's aortic strip.

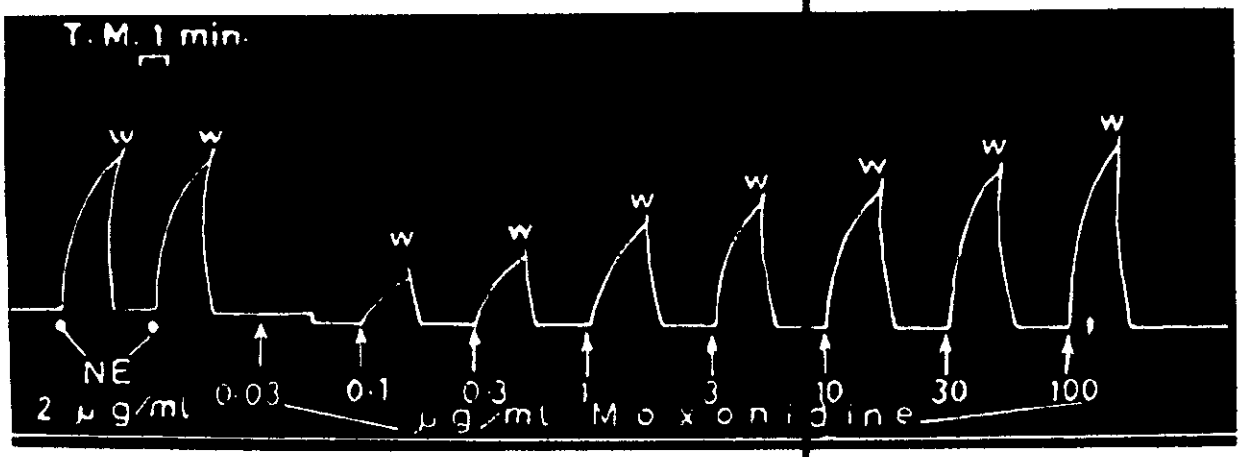


Fig. (65) : A record showing the effect of gradually increasing concentration of moxonidine on isolated rabbit's aortic strip

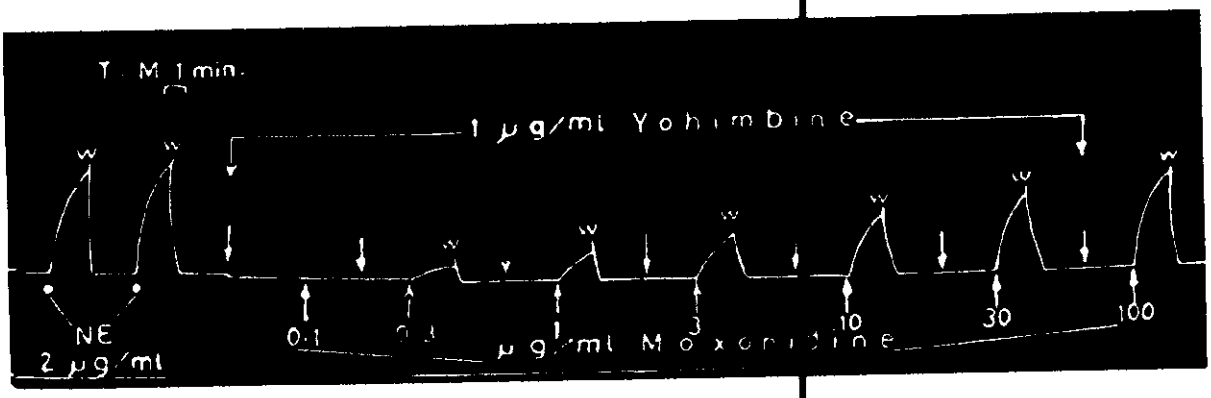


Fig. (66) : A record showing the effect of yohimbine on moxonidine - induced contraction of the isolated rabbit' s aortic strip.

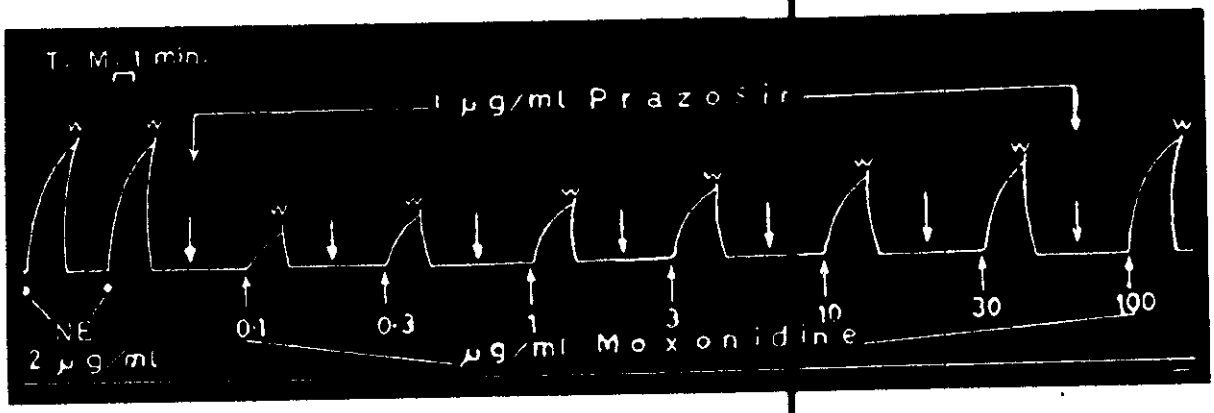


Fig. (67) : A record showing the effect of prazosin on moxonidine - induced contraction of the isolated rabbit' s aortic strip.