

Pharmacological Interactions of Isometamidium and Diminazine With Some Mediator Substances

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Abstract:

The effects of isometamidium and diminazene upon responses of serotonin, noradrenaline and histamine were studied in isolated camel aorta to explain the mechanism of toxicity of these trypanocidal drugs in Dromedary Camel.

It was found that 5-HT, noradrenaline, adrenaline and histamine contracted the isolated camel aorta in a dose-dependent fashion. Isometamidium (1.9, 19, 190 μ M) inhibited 5-HT and noradrenaline responses while histamine responses were inhibited at low doses and potentiated at high doses. Diminazene (1.9, 19, 190 μ M) produced potentiation of 5-HT and noradrenaline responses while histamine responses were inhibited at the same doses. Isometamidium and diminazene were used at different molarities because toxicity was observed at different doses.

Key words:

Diminazene, Isometamidium, Camel aorta , Autacoids, Autonomic antagonists.

Introduction:

The use of antitrypanosomal drugs for cure of trypanosomal infection is one of the several techniques used in controlling trypanosomal infection in man and animals.

Many drugs are now used in the treatment of this infection, such as isometamidium and diminazene.

Use of isometamidium in camels at doses of 0.5 -1.0 mg/kg caused some serious adverse effects including salivation, diarrhoea, frequent urination, defecation, trembling, hind leg weakness and recumbency (Schillinger *et al.*,1985; Ali and Hassan ,1986). While diminazene at doses of 10-40 mg/kg caused hyperaesthesia, salivation, intermittent convulsions, frequent urination and sweating(Leach, 1961; Homeida *et al.*, 1981).

Toxicity in camels have been associated to either the release of or an interaction with autacoids such as serotonin and histamine and cholinergic agents by the trypanocidal drugs within the host tissue (Wien, 1993; Hawking, 1963;Goodwin and Borehan, 1966; Steck, 1971).

The aim of this work is to investigate the interaction of two trypanocides with histamine, 5-HT, adrenaline and noradrenaline responses on isolated camel aorta, in order to elucidate the mechanism of toxic actions of these trypanocidals in Dromedary Camel.

Materials and Methods:

1. Susceptibility of camel isolated aorta to selected autonomic and autacoid mediator substances:

Strips of aorta branch were obtained from freshly killed camel at a local slaughtering house. Branches of the aorta were usually transported to the laboratory in cold Krebs' solution. The tissues were then trimmed of excess fat, connective tissue and parenchyma. Rings 3 mm wide were opened to form strips, mounted in 10 ml organ bath containing Krebs' solution, and gassed with pure O₂ at 37° C. Vascular strips were allowed to equilibrate for approximately 45 min. under a 3g resting tension.

Contractions to the autacoid and autonomic drugs were recorded with isotonic transducer (T₃) connected to FC 100 coupler on Oscillograph 400MD/2(George Washington LTD, England). Cumulative doses for adrenaline, noradrenaline, 5HT, histamine, carbachol, isoprenaline, angiotensin and vasopressin were added to the organ bath and their responses were recorded.

2. Interaction of isometamidium and diminazene with selected autonomic and autacoid mediator substances:

The aorta preparation was prepared as described before.

Then the previous cumulative doses of each autacoid were re-established in the presence of each trypanocide. The trypanocide was left in contact with the tissue for 10 minutes, contractions were recorded with isotonic transducer (T₃).

Three doses of each trypanocide (1.9, 19, 190 μM) were used.

Log concentration response curves of agonists were plotted.

3. Sources of Drugs:

All drugs were of BDH analar except otherwise stated. These included:

- Adrenaline hydrogen tartrate.
- Angiotensin.

- Carbachol.
- Histamine acid phosphate.
- Isoprenaline.
- L-Noradrenaline.
- Serotonin creatinine sulphate
- Vasopressin.
- Diminazene aceturate. (Berenil, Hoechst)
- Isometamidium chloride. (Samorin, M&B)

4. Statistical Analysis:

Results were expressed as mean \pm SEM.

Dose-response curves were fitted using Medusa soft ware and effective median dose (ED₅₀) values were directly determined.

Results:

Susceptibility of camel isolated aorta to selected autonomic and autacoid mediator substances:

The camel aortic strips contracted dose-dependently to 5-HT, noradrenaline, adrenaline and histamine. The tissue was most sensitive to 5-HT, noradrenaline, adrenaline and least sensitive to histamine. Typical individual responses are shown in Figure (1). While carbachol (0.5-64 μ g/ml), isoprenaline (0.05 - 64 μ g/ml), angiotensin (0.01 - 0.64 μ g/ml) and vasopressin (1 IU) did not show any contraction at the doses used.

ED₅₀ values for each agonist were obtained using Medusa soft ware. Results are shown in table (1).

Table (1)

ED₅₀ values for each agonist in camel aorta strips

* Each value is the mean of five experiments.

Drug	ED ₅₀
Adrenaline	34.8 \pm 1.8 μ M
Noradrenaline	9.6 \pm 1.5 μ M
Histamine	82.7 \pm 4.2 μ M
5-HT	0.13 \pm 0.01 μ M

Interaction of isometamidium and diminazene with selected autonomic and autacoid mediator substances:

The two trypanocides had no contractile effects on the camel isolated aorta, but had effects on the autacoid and autonomic agents used.

Effects of isometamidium:

Noradrenaline responses were markedly inhibited and the maximum response was reduced with isometamidium concentrations (1.9, 19, 190 μM) (Fig 2).

5-HT responses were also inhibited dose-dependently, (Fig 3). At 2 μM , histamine responses were weakly inhibited, but at 20, 200 μM histamine responses were potentiated in a dose-dependent manner, (Fig 4).

Effects of diminazene:

At doses (1.9, 19, 190 μM), the noradrenaline responses were shifted to the left (Fig. 5).

5-HT responses were also potentiated (Fig 6).

Histamine responses were inhibited at 119 and 190 μM dose -dependently (Fig. 7).

Discussion:

As shown in the results section, 5-HT, noradrenaline, adrenaline and histamine contracted the isolated camel aorta in a dose-dependent fashion. This observation is consistent with previous results obtained on the pulmonary aorta and vein and the bronchial aorta of cattle; (Archaembault,1977; Arowolo and Eyro, 1979, 1984).

The responses of histamine on the isolated aorta were inhibited by diminazene acetate (1.9, 19 and 190 μM), and isometamidium (2 μM). The results may reflect that at these concentrations, the two trypanocides show antihistaminic properties, and the inhibitory effect on histamine might be due to the blockade of the receptors of histamine by the trypanocides.

Isometamidium (20 μM and 200 μM) enhanced the histamine responses on the isolated aorta. These concentrations of isometamidium are likely to be achieved *in vivo* in Dromedary Camels. Administration of isometamidium chloride (0.5 & 1mg/kg) resulted in plasma concentrations of 44.3 \pm 10.2 and 49.9 \pm 10.2 μM respectively, half an hour after treatment(Ali and Hassan, 1986).

However, at these concentrations, isometamidium might be acting on the mast cell located within the vascular smooth muscle to release more histamine or blocking diamine oxidase (histaminase).

As shown in the results section, isometamidium (1.9, 19, 190 μ M) inhibited 5-HT and noradrenaline responses while diminazene produced potentiation. Drugs that augment or potentiate adrenergic activities act by blocking the uptake of the neurotransmitter and/or inhibiting the monoamine oxidase (MAO) which inactivates noradrenaline. It seemed that diminazene was acting in a similar manner to potentiate noradrenaline response on the isolated camel aorta.

In the camel aorta, inhibitory effects of trypanocides on 5-HT responses can be explained by their effects on the receptor. While the augmentation of the effect may be explained by inhibition of uptake and/or inhibition of the enzymes monoamine oxidase or hydroxyindole-O-methyltransferase.

In conclusion, the reported serious adverse effects of isometamidium and diminazene when these drugs were used in Dromedary Camels, could largely be ascribed to the trypanocides interactions with all the drugs employed.

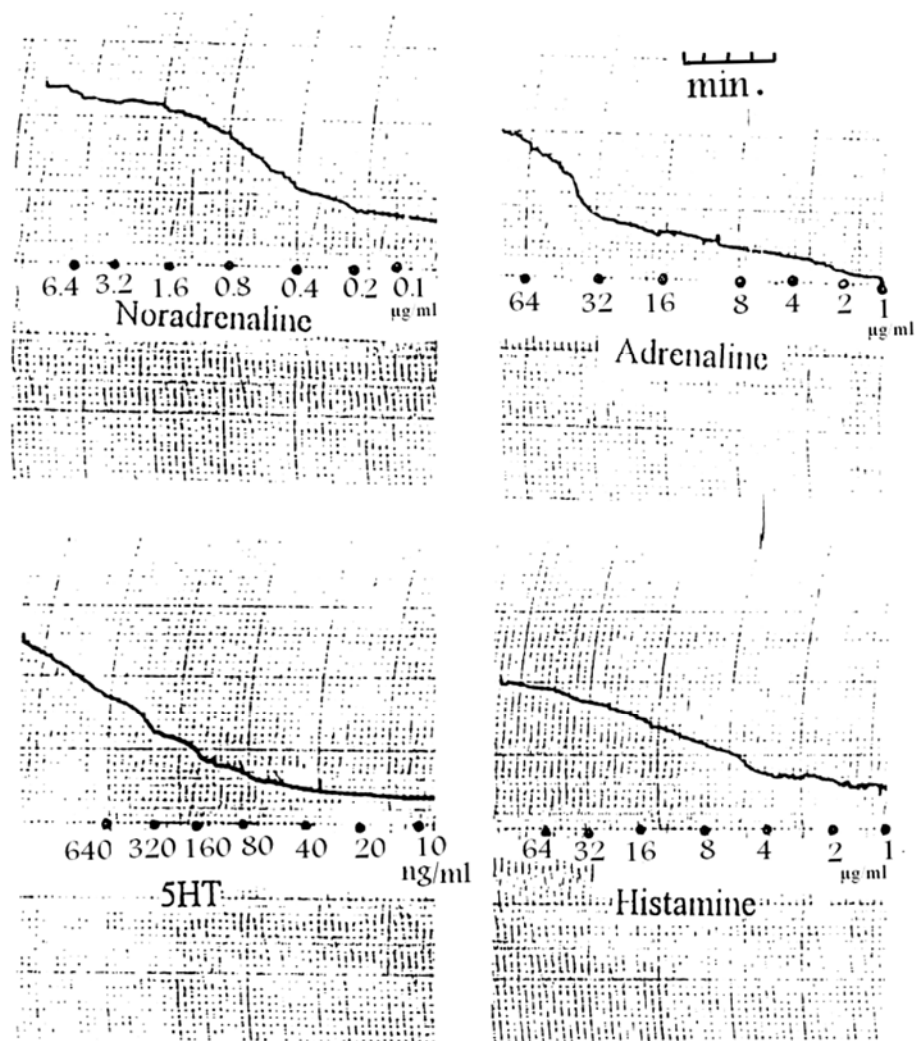


Fig. 1 : Typical in vitro responses of isolated camel artery in Krebs solution aerated with pure O_2 at 37°C . Resting tension 3g. The vessel contracted to cumulative doses of noradrenaline (0.1-6.4 $\mu\text{g/ml}$), adrenaline (1-64 $\mu\text{g/ml}$), 5HT (10-640 ng/ml), and histamine (1-64 $\mu\text{g/ml}$)

* Indicate the time points when additional compound was added to the incubation solution

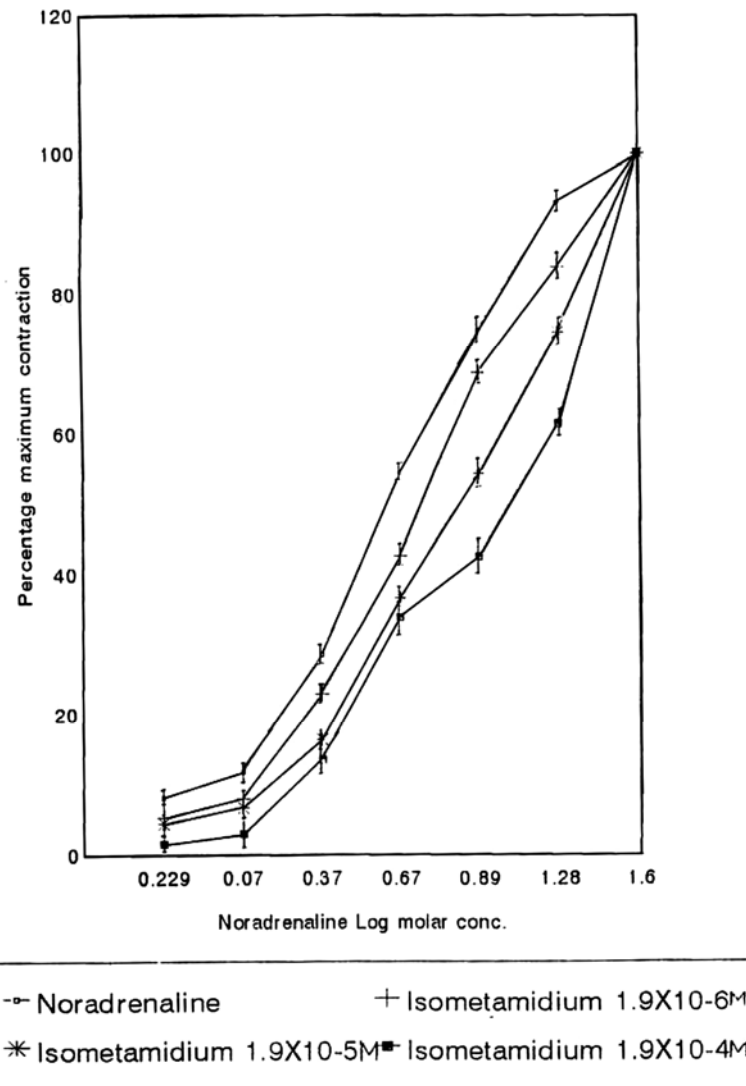
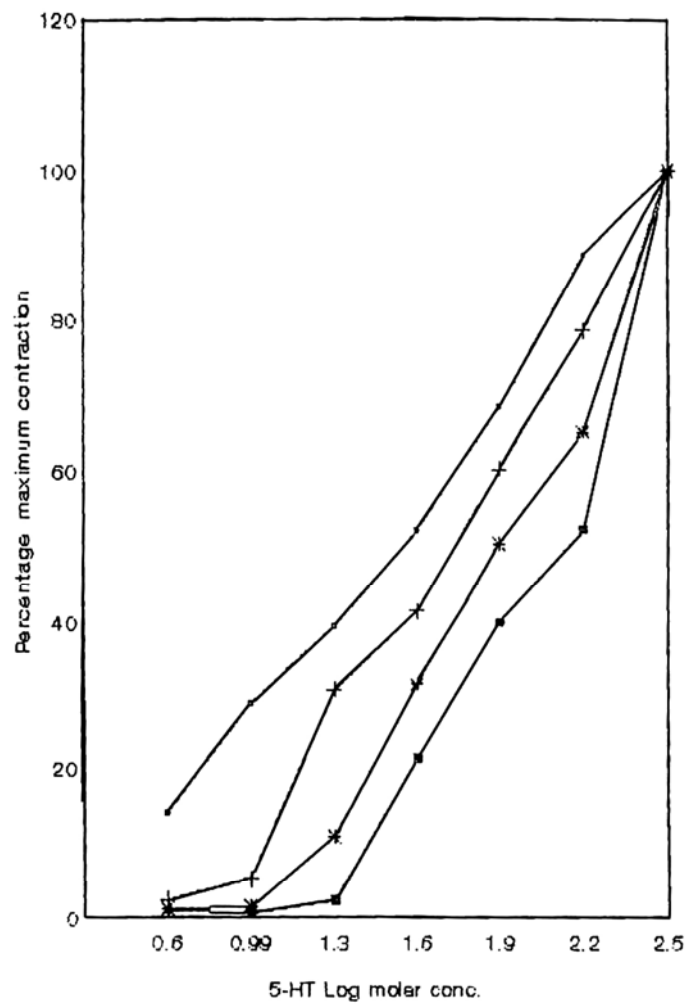


Fig. (2) : Effects of isometamidium (Samorian) on the cumulative dose-response curves for noradrenaline in isolated camel aorta

* Each point represents the mean of five animals



○ 5-HT

+ Isometamidium 1.9X10-6

* Isometamidium 1.9X 10-5

■ Isometamidium 1.9X10-4

Fig. (3) : Effects of isometamidium (samorin) on the cumulative dose-response curves for 5HT in isolated camel aorta

* Each point represents the mean of five animals

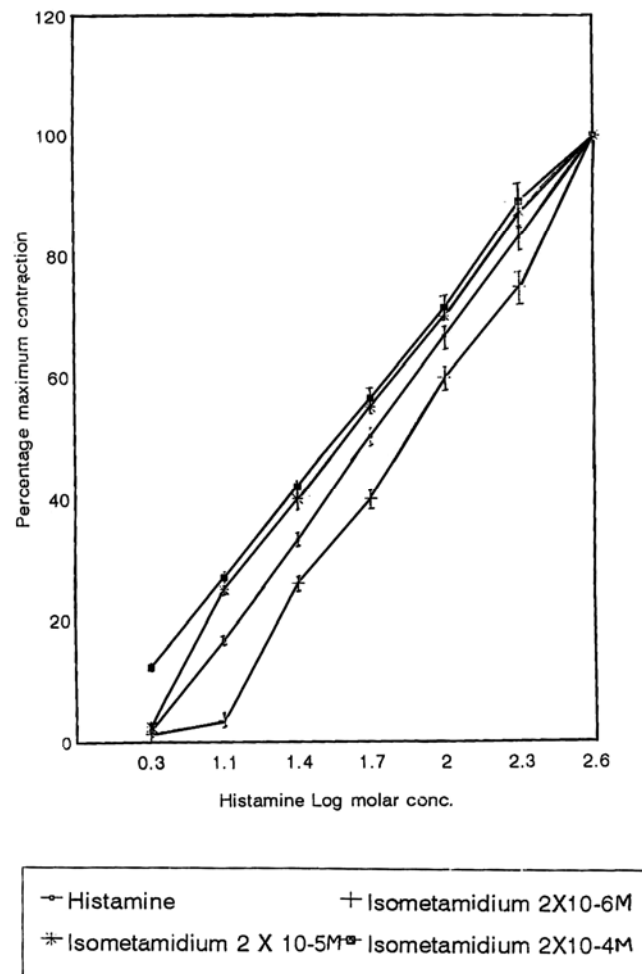


Fig. (4) : Effects of isometamidium (Samorin) on the cumulative dose-response curves for histamine in isolated camel aorta

* Each point represents the mean of five animals

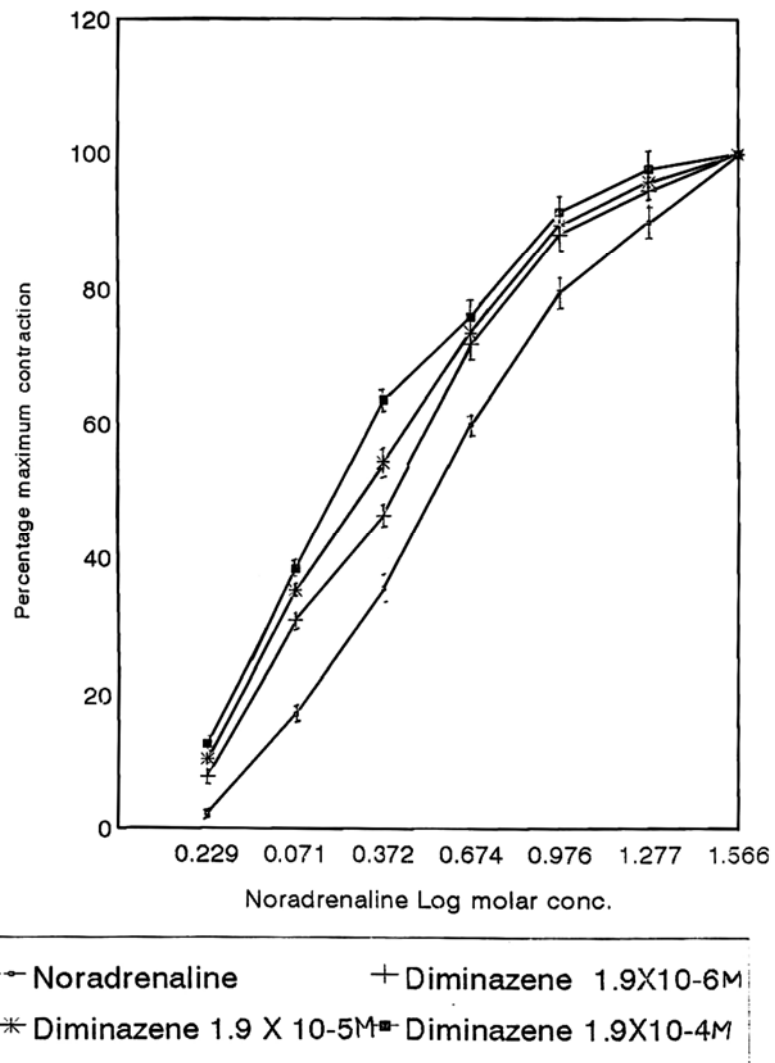


Fig. (5) : Effects of diminazene aceturate (Berenil) on the cumulative dose-response curves for noradrenaline in isolated camel aorta.

* Each point represents the mean of five animals

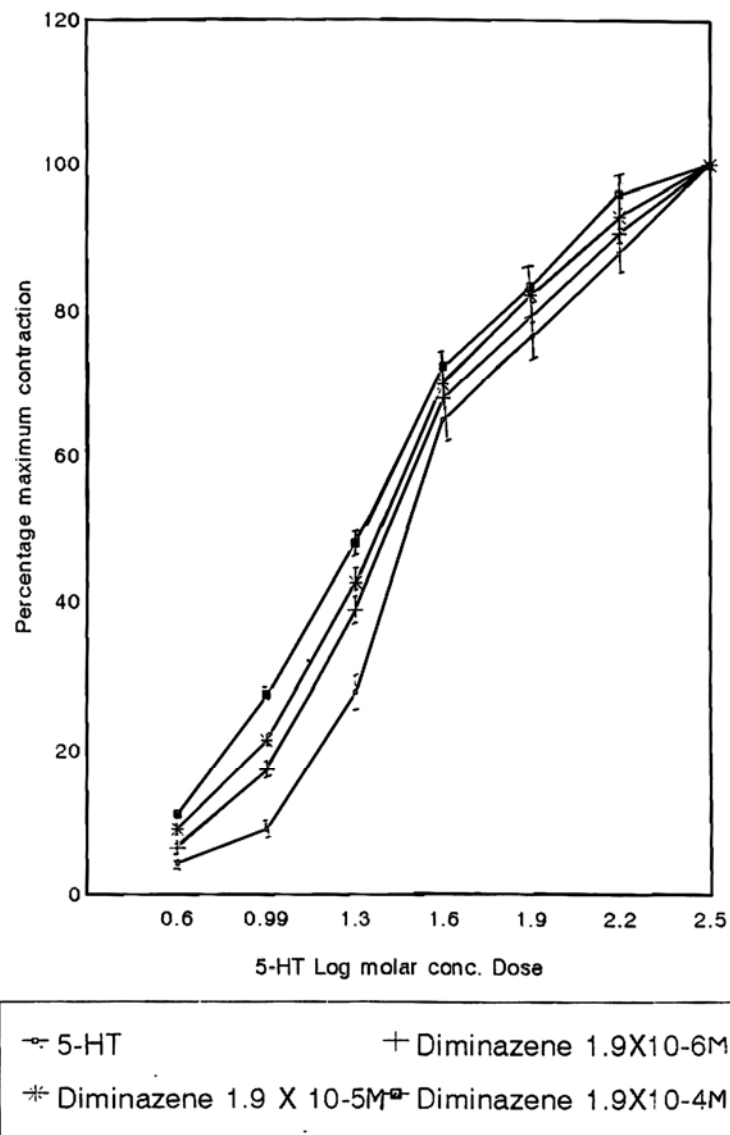


Fig. (6) : Effects of diminazene aceturate (Berenil) on the cumulative dose-response curves for 5HT in isolated camel aorta

* Each point represents the mean of five animals

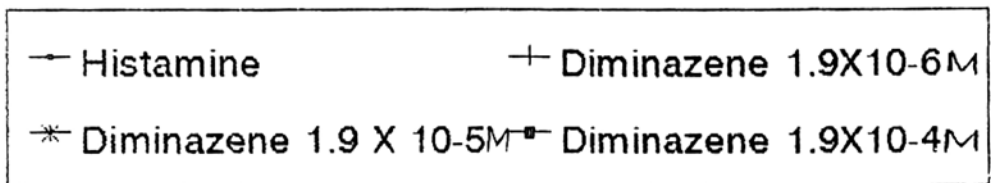
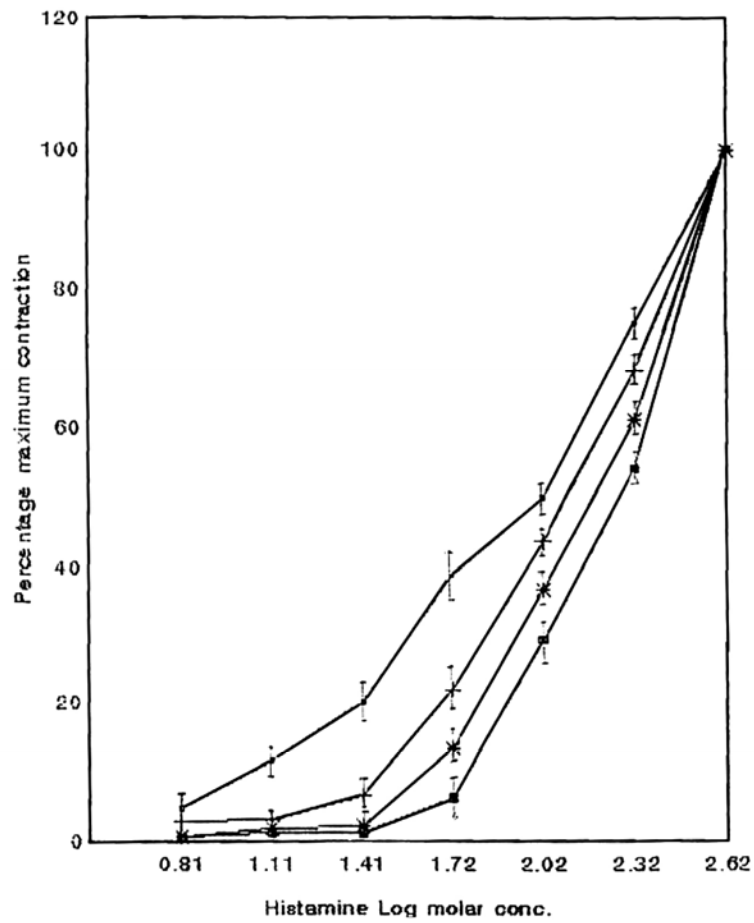


Fig. (7) : Effects of diminazene aceturate (Berenil) on the cumulative dose-response curves for histamine ifn isolated camel aorta

* Each point represents the mean of five animals

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التداخلات الدوائية لعقاري الأيسوميتاميديم و الدايمينازين مع بعض الوسائط الموضعية

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الملخص:

أجرى هذا البحث لدراسة تأثيرات لمضادين من مضادات المثقيبات(التربانسوما) وهما الأيسوميتاميديم والدايمينازين على التقلصات التي أحدثها كل من السيروتونين والنورأدرينالين والهستامين في الشريان الأبهري المعزول من الجمال. وذلك لتفسير بعض أعراض السمية التي تظهر عند استعمال هذين العقارين في الجمال.

أدت إضافة كل من السيروتونين و النورأدرينالين والهستامين إلى الشريان الأبهري لأحداث تقلصات تتناسب طردياً مع جرعات المحفز.

أدت معالجة الأبهري بعقار الأيسوميتاميديم بجرعات (١, ٩, ١٩, ١٩٠ مايكرومولار) إلى تشييط التقلصات التي أحدثها كل من السيروتونين والنورأدرينالين و الجرعات الصغيرة من الهستامين ولكن زادت من التقلصات التي أحدثتها الجرعة الكبيرة من الهستامين.

أدت معالجة الأبهري بعقار دايمينازين بنفس الجرعات أعلاه إلى زيادة التقلصات التي أحدثتها كل من مادتي السيروتونين والنورأدرينالين بينما ثبقت تلك التي أحدثها الهستامين.