

SHORT COMMUNICATION

**EFFECT OF MEPYRAMINE ON THE HISTAMINASE RELEASE INDUCED BY
HEPARIN INJECTION AND ANAPHYLAXIS IN GUINEAPIGS**

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Summary : Injection of heparin as well as antigenic challenge in sensitized guineapigs are known to produce a release of histaminase into the plasma. In the present study, tissue histaminase estimation was done by Spencer's method (17) and plasma estimation by Kapellar Adler's (7) method. Mepyramine pretreatment considerably decreased the histaminase release by both heparin and anaphylaxis. Mepyramine did not antagonise the anticoagulant action of heparin *in vitro*.

Key words: histaminase mepyramine heparin anaphylaxis

INTRODUCTION

Release of histaminase from various organs of rabbit, rat and guinea-pig into the blood, during acute anaphylactic shock has been reported by Rose and Leger (12), Code *et al.* (2), and Logan (9). Incubation of blood from sensitized animals with corresponding antigen, does not increase plasma histaminase, indicating its release from tissues. Dave and Sachdev (3) demonstrated depletion of histaminase in the guinea-pig liver during protracted anaphylaxis.

Neither acute nor chronic administration of histamine in animals or human beings produces a similar release of histaminase (8, 15, 16), excluding histamine as an autoregulator of its own metabolism. Heparin however, was shown to be a potent releaser of histaminase from the guinea-pig liver (15).

Kanakambal *et al.* (6) reported that in cases of acute allergic rhinitis, antihistamines, reduced the raised blood histaminase levels. Whether antihistamines block the release of histaminase, induced by anaphylaxis or heparin injections, has therefore been investigated in guinea-pig.

MATERIALS AND METHODS

Guineapigs of both sexes weighing 250-1000 g were used.

(A) *Estimation of tissue histaminolytic activity after protracted shock :*

Spencer's method (17) was used for tissue histaminase activity. The details of the method, animal sensitisation and antigenic challenge have been described in detail in an earlier communication (3). The tissue histaminolytic activity is expressed as Dt 50 which stands for the time in min, taken by the 2 g equivalent of supernatant of liver homogenate to destroy 50% of added histamine.

Five groups of animals were used : 1) controls, 2) treated with 1 mg/kg im of mepyramine, 3) treated with mepyramine 1 mg/kg + im and antigenic challenge, 4) treated with mepyramine, 5 mg/kg im and 5) treated with mepyramine 5 mg/kg im + im antigenic challenge.

Antigenic challenge was given im to sensitized animals, one hr after mepyramine injection and the animals were sacrificed one hr after either mepyramine alone or mepyramine plus antigen injection.

(B) *Estimation of plasma histaminase activity after acute anaphylactic shock :*

Guineapigs were anaesthetized with 30 mg/kg pentobarbitone sodium ip and the carotid artery and the jugular veins were cannulated with fine polythene tubes.

A control blood sample of 5 ml was drawn from the carotid artery into a calibrated centrifuge tube containing 10 IU heparin per ml of blood. After 10 min of the control sample either heparin 500 IU/kg or 0.1 ml of freshly prepared 5% of egg albumin solution was injected into the jugular vein. Five min after the heparin injection and three min after antigenic challenge another blood sample was collected as before. The samples were immediately centrifuged, plasma collected and histaminase estimation was done within 1 hr of collection. Care was taken to avoid contamination with heparin of instruments and polythene tubes used in these experiments. Plasma histaminase was estimated by Kapellar Adler (7) method and was expressed as permanganate units (PU) per ml of plasma.

Animals were divided into different groups : 1) control anaesthetized animals challenged with iv antigen, 2) anaesthetised animals pretreated with mepyramine 5 mg/kg im and then challenged with antigen iv, 3) control anaesthetised animals treated with

heparin 500 IU/kg iv and 4) anaesthetised animals pretreated with mepyramine 5 mg/kg im, followed by 500 IU/kg of heparin.

(C) *Anti-heparin activity of mepyramine maleate :*

To test whether mepyramine has any anti-heparin activity, like that of protamine, experiments were conducted, following the method described by Sabir and Bhide (13). The following concentrations of mepyramine and heparin were used.

Test tube No. 1: 0.1 ml of blood of human volunteer-control for the normal coagulation time.

Test tube No. 2: 0.1 ml of 500 IU/ml heparin was added to the blood.

Test tubes No. 3 to 7: 0.1 ml of 500 IU/ml heparin in each + 0.1 ml of mepyramine to give ultimate concentration of 1, 10, 100, 1000 μ g/ml of blood.

Drugs used were : Mepyramine maleate (Anthisan, May and Baker), heparin (Biological Evans Limited), histamine (B.D.H.), pentobarbitone sodium (Abbotts Pharmaceuticals) and egg albumin (B.D.H. Analar). All the other chemicals used for preparing physiological solutions were of B.D.H. "analar" grade.

RESULTS AND DISCUSSION

Guinea-pig liver histaminase activity :

Dt 50 was estimated as the time in min taken by 2 g equivalent of supernatant of liver homogenate to destroy 50% of the added histamine. Prolongation of the time was taken to mean decreased histaminolytic activity. The mean Dt 50 estimate of 6 control animals was 42 ± 8.8 min. With 1 mg/kg and 5 mg/kg mepyramine treatment the Dt 50 values were 74 ± 14.2 and 58 ± 3.7 respectively, indicating that mepyramine either inhibited histaminase or its release from the liver. However, the difference between the control group and mepyramine treated group was not significant. Protracted im antigenic challenge given to the mepyramine treated (1 and 5 mg/kg im) animals failed to produce any further reduction in the histaminolytic activity of the liver; the Dt 50 values being 65 ± 5.7 and 67 ± 10.6 (Table I). Since antigenic challenge has been shown by Dave and Sachdev (3) to produce a significant depletion of histaminolytic activity in the guinea-pig, it can be argued that mepyramine blocked the release of histaminase during protracted anaphylactic shock.

Plasma histaminase activity estimated as permanganate units (PU) :

To prove that mepyramine acts through blockade of the release of histaminase from the tissues into the blood, the blood histaminase was estimated in mepyramine treated

controls, antigen challenged controls and antigen challenged mepyramine pretreated animals.

TABLE I: Effect of mepyramine on the histaminolytic activity of guinea-pig liver homogenate, and the depletion of histaminase with antigenic challenge in sensitized animals. Dt 50 is the time during which 50% of the added histamine is destroyed by the homogenate.

Group	n	Mean Dt 50 \pm S.E. in minutes	P
Control	6	42.0 \pm 8.8	
Mepyramine 1 mg/kg, im	4	74.2 \pm 14.2	>0.05 (1 vs 2)
Mepyramine 1 mg/kg, im+antigen, im	4	65.5 \pm 5.7	>0.05 (2 vs 3)
Mepyramine 5 mg/kg, im	3	58.3 \pm 3.7	>0.05 (1 vs 4)
Mepyramine 5 mg/kg, im+antigen im	3	67.0 \pm 10.6	>0.05 (4 vs 5)

The mean plasma histaminase level in 11 untreated guinea-pigs (controls) was 2.82 ± 0.8 (PU). Intravenous injection of antigen in sensitized animals, raised the plasma histaminase to 15 ± 2.291 (PU), the increase being highly significant (Table II). In mepyramine treated (5 mg/kg im) control group the plasma histaminase level was 4.54 ± 1.196 (PU). With antigenic challenge in another mepyramine-treated group the mean value of plasma histaminase was 14 ± 4.54 PU. The difference between these two groups was statistically insignificant (Table II). This indicated that mepyramine blocks the release of histaminase from guinea pig tissue during acute antigenically-induced anaphylactic shock.

TABLE II: Effects of mepyramine on release of histaminase into the blood of guinea-pigs by acute antigenic challenge and heparin.

Group	n	Mean PU \pm S.E.	P
Control	11	2.82 \pm 0.809	
Mepyramine 5 mg/kg, im	11	4.54 \pm 1.196	>0.05 (1 vs 2)
Antigen alone, iv.	5	15.00 \pm 2.291	<0.01 (1 vs 3)*
5 mg/kg mepyramine im+antigen iv	5	14.00 \pm 4.454	>0.05 (2 vs 4)
Heparin 500 IU/kg alone iv	6	66.00 \pm 8.250	<0.001 (1 vs 5)*
Mepyramine 5 mg/kg im+heparin iv	6	46.16 \pm 11.7	>0.05 (5 vs 6)

*Statistically significant.

To locate the site of release of histaminase during anaphylactic shock and to find out whether effect of a known selective histaminase releaser, from guinea-pig liver (4), i.e. heparin, is also blocked by mepyramine, another set of experiments were carried out.

Injection of 500 IU of heparin produced a highly significant release of histaminase into the plasma (Table II). Prior treatment with 5 mg/kg of mepyramine reduced this heparin-induced release (though statistically not significant). This was again suggestive of a partially blocked heparin induced histaminase release by mepyramine. Mepyramine did not neutralise the anticoagulant activity of heparin *in vitro*.

The exact mechanism of blocking action of mepyramine is difficult to explain since histamine does not take part in the release of histaminase (8, 15, 16) and the releasing activity of heparin was only blocked equivocally. The partial blockade produced by mepyramine may therefore be considered a nonspecific, membrane stabilising action on the liver cells (10, 11) involved in the release of histaminase. Kanakambal *et al.* (6), have reported that administration of antihistamines reduced the raised histaminase levels in patients suffering from allergic rhinitis. Sainath and Sachdev (14), found that antihistamines decreased blood histaminase levels in human pregnancy.

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