

RELATIVE BIOACTIVITY OF HISTAMINE AND SLOW REACTING SUBSTANCE OF ANAPHYLAXIS RELEASED DURING ANAPHYLACTIC REACTION FROM DIFFERENT TISSUES OF GUINEAPIGS AND RATS

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Summary : Relative amount of histamine and SRS-A released during Schultz Dale reaction were indirectly estimated. Percent block produced by specific antagonists, mepyramine and FPL 55712, on antigen induced response in various tissues was compared with controls. The relative bioactivity of histamine and SRS-A released was 93% and 7%, respectively, in rat lungs : 37% and 63% in guinea pig lungs; 82% and 18% in guinea pig intestine, 59% and 41% in rat intestine and 100% and 0% in skin of guinea pig and rats. Expectedly, individual experiments showed gross variations because anaphylaxis is rarely dose related.

Key words : slow reacting substance of anaphylaxis platelet activity factor
chemotectic factor of anaphylaxis eosinophil SRS-A blocker

INTRODUCTION

Mediators of anaphylactic reaction in different species are well documented. In addition to histamine, slow reacting substances of anaphylaxis (SRS-A) platelet activity factor (PAF) eosinophil chemotectic factor of anaphylaxis (ECFA), prostaglandins and bradykinin are also released during antigen antibody reaction *in vivo* (6). Although SRS-A may be physiologically more important mediator of anaphylaxis in man (8) both SRS-A and histamine are released from lung (9,1), intestine (3) and from mast cells (4) during *in vitro* anaphylactic reaction. In the present study relative potency of SRS-A and histamine released during antigen antibody reaction at various sites has been estimated, indirectly, by the use of specific antagonists.

MATERIAL AND METHODS

Sensitization and challenge : Guinea pigs weighing 400-500 g were sensitised with egg albumin (60 mg) in two divided doses, (30 mg, ip and 30 mg, sc). Twenty one days later, the animals were sacrificed. Rats (120-170 g) were sensitised with egg albumin 10 mg/kg (sc) along with Freund's adjuvant 1 ml/kg (sc) and repeated on 3rd day. On twelfth day, the animals were used.

Mediator release : Lungs of animals were finely chopped to about 2 mm diameter and were suspended in 10 ml of Tyrode solution at room temperature (7). 20 mg of egg albumin was added to lung tissue and maintained for 10 min with intermittent shaking. Solution was withdrawn at the end of 10 min and was tested on guinea pig ileum as such and in the presence of mepyramine maleate and FPL 55712 (SRS-A blocker).

Mediator release from skin in sensitised guinea pig and rat was studied using skin chamber technique (2) modified by Kulkarni and Dhar (5). Ventral portion of skin was depilated and two abrasions (20 x 20 mm) were made. The skin chamber was filled with either antigen solution (5 ml) or saline (control), fixed on skin abrasion and allowed to remain for half an hour; then their contents were removed and tested on guinea pig ileum.

Mediator release after mast cell degranulation was studied in albino rats (previously sensitised). Animals were anaesthetised with pentobarbitone (30 mg/kg, ip). 10 ml of Tyrode solution (containing 5 mEq/ml heparin and 20 mg of egg albumin) was injected (ip). Ten minutes after injection peritoneal fluid was aspirated and tested on guinea pig ileum.

Mediators released from intestine was studied in guinea pigs; sensitised animals were sacrificed, small intestine was removed, washed with Tyrode solution, cut into pieces and dipped in 10 ml of Tyrode and challenged with 20 mg of egg albumin. Fluid was collected after 10 min and tested on normal guinea pig ileum (3).

Sensitised rats were challenged with 5 mg of egg albumin (iv) pretreated either with antihistamine (Mepyramine) alone or antihistaminic and SRS-A blocker (FPL 55712) given $\frac{1}{2}$ hr before challenge and death within 24 hr was recorded.

RESULTS

Perfusate collected after antigenic challenge of isolated tissues (lung, intestine), peritoneal fluid, as well as fluid obtained after local challenge on sensitised skin were tested in guinea pig ileum as such (control) and in presence of antihistamine (mepyramine

maleate) either alone or in combination with SRS-A blocker. Percent inhibition of contraction compared to control was taken as a measure of percent involvement of histamine or SRS-A. Estimates of the relative release of mediators from different organs have been summarised in Table I. It has been found that SRS-A and histamine are mainly involved in the lung (guinea pig and rat) intestine (guinea pig) and peritoneal fluid (rat). Mediators involved in skin was only histamine (rat and guinea pig). Other mediators like 5-HT, bradykinin and prostaglandin were undetectable in preliminary experiment.

TABLE I : Histamine and SRS-A released during anaphylaxis estimated in terms of percent contraction of guinea pig ileum with or without specific antagonists.

<i>No. of experiments</i>	<i>Intestine</i>	<i>Lung</i>	<i>Skin</i>
Guinea pig (6)	Histamine 82±10.2	Histamine 37±8.9	Histamine 100%
	SRS-A 18±10.2	SRS-A 63±8.9	SRS-A Nil
Rat (6)	Histamine 59±10.9	Histamine 93±13.5	Histamine 100%
	SRS-A 41±10.9	SRS-A 7±13.5	SRS-A Nil

Mean ± S.D.

TABLE II : Absence of protection by antihistamine and SRS-A blocker in rats against systemic anaphylaxis.

<i>No. of animals challenged</i>	<i>Systemic anaphylaxis in rats mortality in 24 hours</i>		
	<i>Control</i>	<i>Antihistamine treated</i>	<i>Antihistamine+SRS-A blocker treated</i>
*(5X3)	5	4	5

Antihistamine, mepyramine maleate, 35 mg/kg.

SRS-A blocker, FPL 55712, 1 mg/kg.

*5 animals in each group.

However, sensitised rats could not be protected against systemic antigenic challenge, with combined use of histamine and SRS-A antagonists (Table II).

DISCUSSION

Involvement of histamine and SRS-A in antigen-antibody reactions both *in vivo* and *in vitro* are well known. However, there is a considerable species variation. Rat lung releases mostly histamine and guinea pig lung primarily SRS-A. In the intestine, histamine is more important both in the rat and in the guinea pig compared to SRS-A; skin, however, releases only histamine both in the rat and in the guinea pig. There has been evidence that SRS-A may be physiologically more important as mediator of anaphylaxis in man (8) and anaphylactic reaction in guinea pig is akin to anaphylactic shock in man. As such, release of mediators during *in vitro* reaction in the guinea pig at various sites might be a pointer to the treatment of local allergic disorders. However, generalised anaphylactic reaction is different from *in vitro* reaction and involvement of SRS-A and histamine may not be significant compared to many other mediators involved.

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