Influence of Antifibrinolytic Substances on Allergic Reactions. Experiments with ε-Aminocaproic Acid and Amino-Methyl-Cyclohexane-Carbonic Acid

Anaphylactic reactions are caused by the pharmacological effects of several 'mediator-substances'. The chain of reactions which is initiated by the contact of antigen and antibody and which ends in the liberation and activation of these mediator-substances (e.g. histamine, 5-hydroxytryptamine, kinins) is not yet clearly understood. The 'enzymatic theory of allergy' states that enzyme activations play an important role in the development of anaphylactic reactions[1]. One of the most important arguments in favour of this theory is the increase in fibrinolytic activity of serum and tissues following antigen administration in man and laboratory animals[1-3]. The question whether activation of fibrinolysis should be considered an important step in the pathogenesis of anaphylactic reaction, or whether it lacks any causal importance, has not yet been answered satisfactorily. Therefore, further investigations seemed warranted, using the most potent antifibrinolytic drugs. In addition to clinical observations, the intensity of anaphylactic shock should be measured by recording biochemical changes with simultaneous determination of antifibrinolytic effects.

Experiments and results. (1) Anaphylactic shock in guinea-pigs. 20 guinea-pigs (300–400 g) sensitized against horse serum were submitted to anaphylactic shock[4]. Pretreatment with ε-aminocaproic acid (EACA, 0.5 g/kg) or amino-methyl-cyclohexane-carbonic acid (AMCA, 0.2 g/kg) 60 min prior to antigen administration did not exert any significant influence on anaphylactic shock.

(2) Anaphylactic shock in rats. 50 Wistar rats (250 to 300 g) were sensitized against human albumin and submitted to anaphylactic shock[5]. Pretreatment with antifibrinolytic agents (EACA 0.5 g/kg; AMCA 0.2 g/kg) did not suppress the clinical appearance of anaphylactic shock following adequate challenge.

(3) The influence of antifibrinolytic substances on anaphylactic sensitization. Daily administration of EACA (0.5 g/kg in 10 rats) or AMCA (0.2 g/kg in another 20 rats) for 10 days following sensitization procedures did not inhibit the development of anaphylactic reactivity.

(4) Allergic reactions of delayed type. In 10 guinea-pigs (350–400 g), allergy of delayed type was induced (dinitrochlorobenzene eczema according to[6]). Pretreatment with AMCA (0.2 g/kg) 60 min prior to allergic challenge (0.09% dinitrochlorobenzene solution in acetone) did not suppress allergic eczematous reaction. In 5 guinea-pigs, an influence of AMCA on development of sensitization was ruled out by daily administration of AMCA for 10 days following primary contact.

(5) Influence of AMCA on renal enzyme excretion following anaphylactic shock. As previously reported[7], an increase in both 'leucine aminopeptidase' (LAP) and alkaline phosphatase (AP) activities occurs in rat urine following anaphylactic shock. LAP-activity increases six-fold, AP-activity three-fold. In 20 rats, pretreatment with AMCA (0.1 g/kg, 60 min prior to antigen administration) did not change the increase in urinary AP-activity. LAP-activity, however, rose two-fold, only. Renal enzyme excretion following histamine shock[8] was neither changed by pretreatment with AMCA (0.2 g/kg in 20 rats) nor by pretreatment with EACA (0.5 g/kg in 20 rats).

Discussion. In guinea-pigs and rats, no influence of EACA or AMCA on clinical pattern of anaphylactic shock was found. Similar results were reported in mice[9]. Allergic reaction of delayed type (dinitrochlorobenzene-eczema) was not inhibited by pretreatment with AMCA, either. It must be concluded that inhibition of fibrinolysis does not suppress allergic reactions in guinea-pigs, rats or mice.

Investigation of renal enzyme excretion provides a reliable model for determination of severity of vascular shock. In addition, such investigations supply information on the state of fibrinolytic system[5,7], as activity of renal peptidases (e.g. LAP) excreted in urine depends upon renal plasmin[10]. Anaphylactic shock provokes hypoxic tubular damage leading to increased enzymatic activities (AP, LAP) in urine. In addition, antigen-antibody reactions activate a cytokinase converting plasminogen to plasmin. Increased renal plasmin levels provoke a further increase of urinary LAP-activity following anaphylactic shock[5,7]. These facts explain the relatively more pronounced rise in urinary LAP in comparison to AP.

The antifibrinolytic substances under investigation exert an antiplasmin as well as an antiactivator effect. The antiplasmin effect suppresses the activation of renal peptidase by plasmin, as was shown in control series[11,12]. The inhibitory action of EACA[11] and AMCA on the increase of urinary LAP-activity following anaphylactic shock leads to the conclusion that antiactivator effect of these 2 antifibrinolytic substances is not only directed against streptokinase and urokinase but also against the cytokinase activated in anaphylaxis.

In contrast to AMCA, EACA exerts an inhibitory action on enzyme systems activated in anaphylactic shock and thus provokes a slight diminution of shock intensity[11]. Control experiments ruled out any direct inhibition of enzymatic activities determined in urine by the action of EACA and AMCA[11,12].

It must be concluded that activation of fibrinolysis does not play an important role in the pathogenesis of allergic reactions in laboratory animals.

Zusammenfassung. In experimentellen Untersuchungen an Meerschweinchen und Ratten wurde der Einfluss zweier Fibrinolysehemmstoffe, ε-Aminocapronsäure (EACA) und Aminomethylcyclohexancarbonsäure (AMCA) auf allergische Reaktionen untersucht. Weder beim anaphylaktischen Schock beider untersuchten Spezies noch beim allergischen Ekzem des Meerschweinchens liess sich eine signifikante Hemmwirkung der beiden untersuchten Antifibrinolytika nachweisen.

W. RAAB

Universitätsinstitut für medizinische Chemie, A-1090 Wien (Austria), 4 October 1967.

[1] W. RAAB and E. KAISER, Klin. Wschr. *43*, 234 (1965).
[2] F. C. LOWELL, W. FRANKLIN, I. W. SCHILLER and E. M. FOLLENSBY, J. Allergy *27*, 369 (1956).
[3] W. M. MEYERS and K. L. BURDON, Archs Biochem. *62*, 6 (1956).
[4] E. KAISER, W. RAAB and W. LEPIER, Arch. klin. exp. Dermat. *220*, 321 (1964).
[5] W. RAAB, Wien. klin. Wschr. *78*, 364 (1966).
[6] J. R. FREY and P. WENK, Dermatologica *112*, 265 (1956).
[7] W. RAAB, Nature *212*, 953 (1966).
[8] W. RAAB, Z. ImmunForsch. exp. Ther. *131*, 192 (1966).
[9] B. MELANDER, G. GLINIECKI, B. GRANSTRAND and G. HANSHOFF, Acta pharmac. tox. *22*, 340 (1965).
[10] I. INNERFIELD, R. HARVEY, F. LUONGO and E. BLINCOE, Proc. Soc. exp. Biol. Med. *116*, 573 (1964).
[11] W. RAAB, Z. ImmunForsch. exp. Ther. *131*, 294 (1966).
[12] W. RAAB, Proc. Eur. Acad. Allergol, Praha 113 (1967).