COMPARISON OF THE ACTION OF BRADYKININ, KALLIDIN, METHIONYL-LYSYL-BRADYKININ, AND ELEDOISIN ON CUTANEOUS VASCULAR PERMEABILITY

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The cutaneous vessels of rabbits and rats are equally sensitive to the action of bradykinin and methionyl-lysyl-bradykinin, and one-third less sensitive to kallidin than to these agents. The sensitivity of the rabbit's cutaneous vessels to eledoisin is 25,000 times less, while that of rats is four times higher, than to bradykinin. After intradermal injection of bradykinin, kallidin, or methionyl-lysyl-bradykinin, the duration of their action as permeability factors is short.

Considerable importance has recently been attached to the kinins as mediators of inflammation. In extremely small doses the kinins have been shown to increase vascular permeability, to produce pain and emigration of leukocytes, and to stimulate phagocytosis [2].

Until recently, however, the relative activity of the various kinins has received insufficient study, although in vivo and, in particular, under pathological conditions, the various plasma kinins (bradykinin, kallidin, methionyl-lysyl-bradykinin) may be formed and accumulate. It was therefore decided to study a number of kinins, including eledoisin.

Eledoisin, a kinin from the salivary glands of the octopus, is not found in the human body. However, it is close in its amino-acid composition and properties to the substance P found in the central nervous system and small intestine of mammals (the structure of substance P has not yet been finally settled). In addition, the study of eledoisin is interesting from the pharmacological point of view, for it possesses a powerful hypotensive action which is more prolonged than that of the plasma kinins [7].

EXPERIMENTAL METHOD

Experiments were carried out on 20 chinchilla rabbits (2.9-3.6 kg) and 30 Wistar rats (180-230 g). The hair was clipped with scissors 3-4 days before the experiment began. The effect of kinins on the cutaneous vascular permeability was studied by Ramsdell's principle [8] in the modification of Oivin and Shchegel' [4]. The dye Evans blue (T-1824) was injected intravenously as a 1% solution in a dose of 20 mg/kg body weight. Various doses of kinins, made up in 0.1 ml physiological saline, were injected intradermally 5 min after injection of the dye. Physiological saline was injected into control areas of skin. The criterion of a disturbance of permeability of the cutaneous vessels was the appearance of a blue-stained papule, not less than 7 mm in diameter, in the course of 10 min. The duration of action of the kinins on vascular permeability was determined by the method of Oivin et al. [3].

The results were analyzed by the probit method of Litchfield and Wilcoxon as modified by Roth [1].

The following synthetic preparations of the kinins were used in the investigation: bradykinin (Sandoz, Switzerland), methionyl-lysyl-bradykinin (Schering, A. G., West Germany), kallidin (Sandoz, Switzerland), and eledoisin (Schering, A. G., West Germany).

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EXPERIMENTAL RESULTS

The experiments showed that small doses (0.001-0.01 ng) of plasma kinins, if injected intradermally, disturbed the cutaneous vascular permeability in rabbits. The papules thus formed were diffusely stained blue with the Evans dye, and the outlines of the stains were blurred. An increase in the dose of kinins to 10-100 ng caused appreciable dilation of the vessels as reflected in the appearance of slight erythema. These doses of plasma kinins evoked a deep reddening of the papule, the boundaries of which were clearly defined. The criterion for comparison of the substances tested was the dose causing a disturbance of vascular permeability in 50% of animals (ED_{50}). Bradykinin and methionyl-lysyl-bradykinin were found to be similar in their activity. ED_{50} of bradykinin for the rabbit was 0.025 (0.010-0.060),* and (at of methionyl-lysyl-bradykinin 0.021 (0.09-0.48) ng. Intradermal injection of 10 ng bradykinin or methionyl-lysyl-bradykinin in every case disturbed the cutaneous vascular permeability of the rabbit. ED_{50} of kallidin was 0.018-0.74 ng, and of eledoisin 630 (390-1010) ng, i.e., the sensitivity of the cutaneous vessels of the rabbit to eledoisin was 25,000 times less than to bradykinin.

Compared with the rabbit's cutaneous vessels, those of the rat were less sensitive to the action of the kinins. ED_{50} of bradykinin for rats was 17 (10-30) ng, of methionyl-lysyl-bradykinin 15 (9-25) ng, and of kallidin 31 (19-49) ng. Intradermal injection of 100 ng bradykinin, kallidin, or methionyl-lysyl-bradykinin in all cases disturbed the cutaneous vascular permeability in rats. ED_{50} of eledoisin in rats was 4 (2-8) ng.

Despite the high activity of the plasma kinins, the disturbances of vascular permeability produced by them were of short duration. In these experiments 500 mg bradykinin, kallidin, or methionyl-lysyl-bradykinin, when injected intradermally into a rabbit, produced a disturbance of vascular permeability lasting on the average 12.8-14 min. Eledoisin, in a dose of 50,000 ng (approximately one-tenth as active as 500 ng bradykinin) produced a distrubance of vascular permeability in the rabbit's skin which lasted rather longer – about 16 min.

The "threshold dose" is usually used as the main criterion when comparing mediators of permeability. The concept of threshold dose in pharmacology means the dose of a preparation or magnitude of a stimulus evoking an effect in 30% of cases [1]. In physiology, the term threshold dose is usually taken to mean the magnitude of a stimulus evoking an effect in the majority of cases (in 70, 80, or 90%). It therefore seems more reasonable to assess species sensitivity to a stimulus by the dose of a preparation evoking an effect in 50% of animals. Besides this more precise definition, the criterion suggested also appears more adequate because it is statistically the most stable criterion and can be used to compare individual indices with each other. When this criterion was used, the cutaneous blood vessels of the rabbit were shown to be highly sensitive to the action of all three plasma kinins. These results showing the equal sensitivity of the rabbit's blood vessels to bradykinin and to methionyl-lysyl-bradykinin are in agreement with results obtained by Elliot and Lewis [6], who found that the threshold dose of both kinins was 5 mg. The lower activity of kallidin than of bradykinin (one-third) was demonstrated in the experiments of Carr and Wilhelm [5]. Similar results were obtained in the present experiments; the sensitivity of the skin vessels to bradykinin was 1.5 times higher, and to methionyl-lysyl-bradykinin 1.7 times higher than to kallidin. The same pattern of behavior was found in experiments on rats. The results of these experiments, indicating higher sensitivity of the rat's blood vessels than those of the rabbit to eledoisin, are also in agreement with data published elsewhere [5]. The kininase activity of the rabbits skin is high. Even when such a large dose as 500 ng bradykinin, kallidin, or methionyl-lysyl-bradykinin was injected, judging from the duration of the disturbances of vascular permeability (and assuming that the reactivity of the cutaneous vessels at the site of injection of bradykinin remains unchanged [2]), the inactivation time did not exceed 14 min. It is interesting to note that methionyl-lysyl-bradykinin, which differs from kallidin and bradykinin in its higher resistance to plasma kininase [6], when injected intradermally acted only a little longer than bradykinin. This indicates differences in the nature of the enzymes inactivating methionyl-lysyl-bradykinin in the blood and tissues.

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