Tabelle II. Dünnschichtchromatographisches Verhalten der in *Phaseolus coccineus* L. nachgewiesenen neuen Gibberelline «Phaseolus α-ε»

| Substanz | Entwicklungsgemisch [a] | $R_{St.}$ (bez. auf Rf $A_3 = 1{,}00$) |
|---|---|---|
| α | I | 0,82 |
| β | II | 0,30 |
| γ | II | 0,24 [b] |
| δ | II | 0,11 |
| ε | I, II | 0,00 |
| | III | 0,38 |

[a] I: Chloroform/Essigester/Eisessig (60:40:5), $R_{St.}$ $A_3 = 0{,}50$; II: Chloroform/Essigester/Eisessig (70:30:5), $R_{St.}$ $A_3 = 0{,}50$; III: *n*-Propanol/5 *n* $NH_3$ (5:1), $R_{St.}$ $A_3 = 0{,}70$. Mit I 4 h, mit III 12 h aufsteigende und mit II 15 h aufsteigend durchlaufende Entwicklung bei 20°C an Kieselgel-G (Merck); vgl.[1]. Ausser den angegebenen wurden weitere Entwicklungsgemische zur Charakterisierung der Substanzen herangezogen.

[b] Beim $H_2SO_4$-Nachweis gelbe Fluoreszenz.

eine im Vergleich zu «ε» weniger polare Substanz mit Essigester extrahieren, die bei $H_2SO_4$-Detektion ähnlich wie die herkömmlichen Gibberelline fluoresziert. Bei Dünnschichtchromatographie mit Gemisch I bzw. III (vgl. Tabelle II) wurde $R_{St}$ 0,70 bzw. 0,85 festgestellt. Dieses Spaltprodukt zeigt ein gleiches dünnschichtelektrophoretisches Verhalten[5] wie die bekannten Gibberelline (anodische Wanderung). Im Hydrolysat von «Phaseolus ε» (wässerige Phase nach Essigester-Ausschüttelung) konnten dünnschichtchromatographisch zusätzlich folgende Substanzen nachgewiesen werden: (1) mindestens 2 mit Anilinhydrogenphthalat positiv reagierende Verbindungen, von denen eine in mehreren Entwicklungsgemischen[6,7] mit Glucose übereinstimmt und die andere stets einen kleineren Rf-Wert aufwies; (2) mindestens 3 ninhydrinpositive Substanzen[6], deren Identifizierung noch aussteht.

Zusammenfassend sei festgestellt, dass in *Phaseolus coccineus* L. neben Gibberellin $A_1$, $A_5$, $A_6$ und $A_8$[3] zusätzlich $A_3$ sowie 5 neue gibberellinwirksame Substanzen, «Phaseolus α–ε», aufgefunden wurden. Nach bisher vorliegenden Ergebnissen handelt es sich zumindest bei «ε» um ein an Kohlenhydrate und ninhydrinpositive Komponenten gebundenes Gibberellin. Ein Vorkommen von Gibberellin $A_3$ in höheren Pflanzen ist unseres Wissens bisher nicht sicher festgestellt worden. Hingegen wurden stark polare, gibberellinwirksame Substanzen sowohl in Kartoffelknollen als auch in Bohnen- und Salatpflanzen bereits nachgewiesen[8,9]. Über die Bildung eines Gibberellinsäureglucosids *in vitro* ist gleichfalls berichtet worden[10]. Die Existenz gebundener Gibberelline wurde gelegentlich diskutiert[8,11].

*Summary.* In addition to gibberellins $A_1$, $A_5$, $A_6$ and $A_8$ previously isolated from *Phaseolus coccineus* L., the occurrence of gibberellin $A_3$ and of 5 new gibberellin-like substances «Phaseolus α–ε» was demonstrated. «Phaseolus ε» was proved to be a gibberellin bound to carbohydrates and ninhydrin-positive compounds.

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## The Effects of Intraperitoneal Adaptation to Histamine on the Anaphylactic Shock in the Guinea-Pig

In a previous paper dealing with the influence of inhalatory adaptation to histamine (H) on anaphylactic shock, we found histamine-adapted animals to show increased resistance only when they had been subjected to an additional exposure to histamine aerosols immediately before the lethal H dose or shock-producing dose of antigen was given[1]. On the other hand, it is known that adaptation to H produced by its inhalatory application is first of all a kind of local adaptation of the lungs[2]. In this paper we present the data concerning the influence of intraperitoneal adaptation to H on anaphylactic shock in guinea-pigs.

*Methods.* Experiments were made on male guinea-pigs 300–420 g in weight. Animals were adapted to H for 8 weeks by daily intraperitoneal injections of 0.1 mg histamine dihydrochloride. Before and after adaptation guinea-pigs were tested one by one in histamine aerosols in order to obtain the curve of susceptibility to H. After adaptation had been produced, we calculated adaptation capacity by a method described elsewhere[3]. D-30 aerosol generator was used to produce H aerosols. Sensitization of animals and the anaphylactic shock were produced by HERBERTS's method[4] with hen's egg albumin.

*Results.* Sensibility to H before and after adaptation is presented in the Figure. Adaptation capacity is for 0.5% of H – 3.0, 0.8% of H – 2.4, 1.6% of H – 1.2. These values show that animals, after their adaptation by intraperitoneal injections of H, are 3 times less sensitive to aerosol

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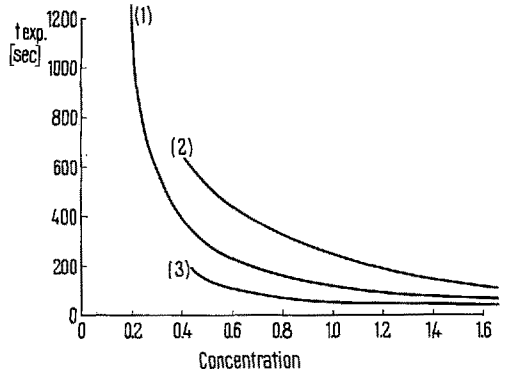
of 0.5% of H, 2.4 times less sensitive to that of 0.8% of H and 1.2 times less sensitive to that of 1.6% of H as compared with sensitivity before adaptation. Animals which had not been adapted did not change their sensitivity to H. The results of experiments on the course of the anaphylactic shock in animals adapted to H are presented in the Table.




Sensitivity to histamine before and after adaptation. (1) Initial sensitivity to histamine (group I, II, III, IV, V). (2) Final sensitivity to histamine – after adaptation (group I, II, III). (3) Final sensitivity to histamine in non-adapted guinea-pigs (group IV, V).

| Group | | Number of animals | Survived | Died |
|---|---|---|---|---|
| I | Adapted to H, sensitized | 7 | 3 | 4 |
| II | Adapted to H, sensitized, exposure to H aerosol immediately before shock-producing dose of antigen | 7 | 6 | 1 |
| III | Adapted to H, non-sensitized | 7 | 7 | 0 |
| IV | Non-adapted to H, sensitized | 6 | 1 | 5 |
| V | Non-adapted to H, sensitized, exposure to H aerosol immediately before shock-producing dose of antigen | 4 | 1 | 3 |

Table of statistical significance

| | I | II | III | IV | V |
|---|---|---|---|---|---|
| I | | $0.2<p<0.3$ | $0.05<p<0.1$ | $0.1<p<0.2$ | $0.2<p<0.3$ |
| II | | | $p>0.99$ | $p<0.01$ | $p<0.01$ |
| III | | | | $p\ll 0.01$ | $p\ll 0.01$ |
| IV | | | | | $0.5<p<0.7$ |

*Discussion.* The data presented above indicate that animals adapted to H by intraperitoneal injections show increased resistance to anaphylactic shock. This resistance is more pronounced when animals have been submitted to an additional exposure to histamine aerosol immediately before the shock-producing dose of antigen is applied. A single exposure of control animals to H did not alter their sensitivity to the anaphylactic shock.

Adaptation to H injected intraperitoneally increases resistance to anaphylactic shock more significantly than that produced by inhalatory H application. This finding supports our previous data that adaptation to H produced by its inhalatory application is rather a local phenomenon, and not a general one. In fact, the general symptoms of decreased susceptibility take place in animals adapted to H by its inhalatory application, too. But they are less pronounced than those from lungs. In both cases adaptation to H increases resistance to anaphylactic shock irrespective of the way in which H has been applied.

What is the mechanism of resistance? From the data obtained in this laboratory, it is evident that resistance to H in the adapted guinea-pigs does not depend on histaminase, because histaminase activity in blood, liver, kidneys and brain of these animals does not increase, and, as to the lungs, histaminase activity even shows a slight decrease[5]. It is possible that resistance to the anaphylactic shock depends on a decreased lung sensitivity to H. We have demonstrated that lungs and ileum isolated from guinea-pigs adapted to H were significantly less sensitive to H than those from the controls[6]. It seems probable that it is peripheral tissue resistance to H and anoxia which are responsible for increased general resistance. We have demonstrated that guinea-pigs adapted to H by inhalatory application were more resistant to KCN than the controls[7].

The death of a guinea-pig in anaphylactic shock is an asphyctic one. The final effect is determined by oxygen. The battle for oxygen is a battle for time. By means of an increased resistance to the lack of oxygen, it is possible to increase general resistance to the anaphylactic shock. This is a possible explanation of the mechanism of the influence of adaptation to H on anaphylactic shock in the guinea-pig.

*Résumé.* Les cobayes adaptés à l'histamine par voie intrapéritonéale sont protégés contre le choc anaphylactique. Les mécanismes périphériques de la résistance sont discutés.

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