Effect of several compounds on the clotting time of whole blood

Test compound[a]	Rat	Guinea-Pig	Human			
			F.M.	D.H.	B.St.	S.H.
	(%)	(%)	(%)	(%)	(%)	(%)
Nil	100[a]	100	100	100	100	100
Methylmercuric chloride	60	55	>500[c]	>500[c]	140	>500[c]
Meralluride	100	100	100	111	150	250
Ethacrynic acid	100	100	70	100	70	80
Mersalyl	>500[b]	>500[b, c]	>500	>500	>400	>500
PCMB	100	100	92	96	104	100
$HgCl_2$	—	93	100[b]	—	—	100[b]

[a] All values expressed relative to control time designated as 100%; [b]Hemolysis of red blood cells was observed; [c] Slight partial incomplete clots observed; contents could move easily upon inversion of tubes and complete clotting of test tube contents resulting in stoppage of movement of contents did not occur; [d] All test compounds were present at a final concentration of 3.3×10^{-3} *M* except PCMB which was present at a concentration of 8×10^{-4} *M*.

p-chloromercuribenzoate and mercuric chloride were ineffective in altering clotting times of all species studied.

No definite conclusion as to the mechanism of action whereby these compounds affect clotting times is possible from the preliminary results presented here because of the complex sequence of events and the many co-factors involved in the blood coagulation process. For example, although blood clotting may be regarded to consist of 3 basic reactions involving autoprothrombin C, thrombin or fibrin formation, many other accessory complexities are essential in order to permit the smooth progression or retardation of these reactions[5]. Moreover, sulfhydryl reagents have been shown to prevent the release of various platelet constituents which are known to be invoved in blood clotting[6, 7] and thus may account for the inhibition of the clotting mechanism by some of the compounds tested. Conversely, acceleration of the clotting times might be due to stimulation of the release reaction. In addition, Factor XIII which requires free thiol groups for activity has been shown to be inactivated by mercuric compounds[8, 9] thus offering another possible mode of action in order to account for the delayed clotting times obtained with several of the compounds. Quantitative differences in clotting times between individuals or species might also be due to differences in plasma levels of mercaptalbumin (major source of protein sulfhydryl groups in plasma) which is known to protect clotting factors from inactivation by sulfhydryl reagents[10]. However, a lack of information on the plasma levels of mercaptalbumin in the various species and its relative effectiveness in preventing the drug-induced inactivation of clotting factors prevents a plausible speculation on its contribution towards the effects on the clotting process of those drugs reported in this communication.

Résumé. On étudie l'effet d'une série de composés (capables de réagir avec les groupes sulfhydryle des protéines) sur la vitesse de coagulation du sang recalcifié du rat, du cobaye et de l'homme. Le chlorure méthylique de mercure accélère la coagulation chez le rat et le cobaye, mais la retarde chez l'homme. Le méralluride accélère et l'acide éthacrinique retarde la coagulation du sang chez certains sujets humains, mais n'ont aucun effet chez le rat et le cobaye. Le mérsalyle prolonge de beaucoup le temps de coagulation chez le rat, le cobaye et l'homme, mais le PCMB et le $HgCl_2$ n'ont pas d'effet.

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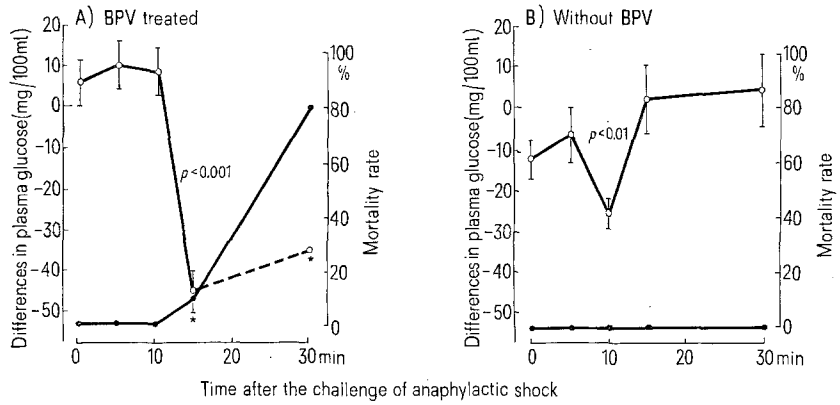
Changes of Blood Glucose During Anaphylaxis in Pertussis Sensitized Rats

It is well known that the injection of *Bordetella pertussis* vaccine (BPV) given along with the sensitizing dose of antigen decreases the resistance of rats to anaphylactic shock[1, 2]. The severity of anaphylactic shock is considerably influenced by alterations in blood glucose, i.e., increased anaphylactic sensitivity could be induced by agents causing hypoglycemia at the time of challenge, and hyperglycemia results in a decreased susceptibility[3–5]. After BPV treatment a prolonged decrease of blood glucose[5, 6] and disturbances in blood glucose regulation[7–9] could be demonstrated in rats. To verify the possible role of these findings in the pathomechanism of severe anaphylactic reactions, blood glucose alterations were followed during the time course of anaphylactic shock in BPV pretreated and untreated rats.

200 female rats of Wistar strain (150–200 g) were devided into 4 groups and treated i.p. as follows: 1.1 ml of phys. saline, 2. 1 ml of horse serum, 3. 0.1 ml of BPV (3×10^{10} organism) + 1 ml of phys. saline, 4. 0.1 ml of BPV + 1 ml of horse serum. 12 days later as a challanging antigen dose, 1 ml of horse serum was administered i.v. to each of the groups of animals. Thus, groups 2 and 4



Effect of anaphylactic challenge on plasma glucose levels in rats sensibilized with (A) or wihtout (B) *Bordetella pertussis* vaccine. ● — ●, mortality rate (%) in groups of rats undergoing anaphylactic shock; ○—○, plasma glucose differences between unsensibilized controls and sensibilized groups, i.e. between groups 3 and 4 (A) or between groups 1 and 2 (B). Vertical lines represent the standard errors. Values of significance were related to the results obtained before anaphylactic challenge. *Results indicate the average of 9 and 2 animals corresponding to the mortality rate.

were submitted to anaphylactic shock and groups 1 and 3 served as their controls. 10 rats from each groups were killed before or at various intervals (see in the Figure) after the i.v. challenge if they did not die earlier. Plasma glucose levels were assayed by the glucose oxydase method of CAWLEY et al.[10]. Animals were fasted 18 h before killing them. Mortality ensued in various groups during anaphylaxis is shown in the Figure.

Challenge of anaphylaxis induced only a slight transitional decrease of blood glucose followed by a rapid recovery (Figure1B).This finding suggests that the increased peripheral glucose utilization of anaphylactized animals will be compensed by counterregulatory mechanisms such as the hyperglycemic action of released epinephrine, histamine and 5-hydroxytryptamine[7–9]. In these groups death did not occure and only moderate anaphylactic symptomps developed in the first 30 min of anaphylactic shock.

During the first 10 min blood glucose of BPV treated sensibilized and unsensibilized groups did not differ significantly from each other (Figure 1A). However, after this period, blood glucose dropped sharply and 10% of the anaphylactized rats died between the 10th and 15th min and another 70% from the 15th to 30th min.

In previous works, it has been shown that hyperglycemic effect of epinephrine[7,8] as well as that of histamine and 5-hydroxytryptamine[9] were considerably inihibited by BPV pretreatment. Thus, during anaphylaxis of BPV treated rats there is an inhibition in the above mentioned hyperglycemic counterregulation, and the prevailing hypoglycemic mechanisms results in a decrease of blood glucose, which may play an important role in the development of fatal anaphylactic symptomps. The latter hypothesis is supported by the fact that death had occured only after blood glucose dropped.

Zusammenfassung. Nachweis, dass der Blutzuckerspiegel von mit *Bordetella pertussis* vorbehandelten Ratten durch anaphylactischen Schock wesentlich vermindert ist, obwohl er in nicht vorbehandelten Tieren unverändert blieb.

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Androgen Receptor in the Thumb Pads of *Rana esculenta*

The thumb pad of *Rana esculenta* is a male secondary sexual character dependent upon gonadal hormones (LOFTS[1]). D'ISTRIA et al.[2] have demonstrated that the administration of small amounts of testosterone propionate to castrate adults induces hypertrophy in both the epidermal and glandular layers of the thumb pads and an increase in RNA and protein content. When labelled testosterone was injected into adult castrates (D'ISTRIA et al.[3]), the radioactivity showed a tendency to concentrate in the thumb pads. The study of in vitro metabolism indicated a conversion of testosterone into androstanolone and 11-Ketotestosterone. Both these metabolites stimulate the thumb pads when injected into castrate males.

In order to establish the presence of androgen receptor in thumb pads of *Rana esculenta*, the following experiments were done. Thumb pads, obtained from one month-castrates, were minced and homogenized in 1.5×10^{-3} *M* EDTA – 2×10^{-2} *M Tris*-HCl buffer pH 7.5, in an all-glass