cells from spleens and lymph nodes, respectively, as compared to 30–60 mg for heart, lung and kidney. Liver differed from these tissues in the low contents of both fatty acids and triglycerides. Table II shows a representative of cytotoxicity tests in which the neutral lipid classes and the phospholipids from lymphoid cells and the livers were examined for their cytotoxic activity against the tumor cells as described above. It can be seen that all, or nearly all, groups of the mice receiving the tumor cells preincubated with fatty acids, irrespective of the original tissues, at a concentration of 2 mg per ml of cell suspension failed to develop tumors at the end of the 60-day period, while control mice invariably died from ascitic tumors in less than 20 days. In contrast to this, all other lipid classes were practically ineffective to suppress the growth of tumor cells. From the foregoing it may be concluded that lymphoid cells are sharply distinguishable from other mammalian cells in the exceptionally high contents of the cytotoxic free fatty acids.

Although fatty acids and their esters from various sources[5,8–13] have long been known to be highly cytotoxic to mammalian cells including tumor cells, OKUDAIRA et al.[14] were the first to suggest the possibility of fatty acids isolated from lymph node extract as a cytotoxic factor of lymphoid cells. Very recently, TURNELL et al.[15] have presented evidence that accumulation of free fatty acids is involved in corticosteroid-induced lymphocytolysis. The present results, demonstrating the occurrence of high levels of cytotoxic fatty acids concomitant with the increase of sterol esters in lymphoid cells, point to characteristic features of fatty acid metabolism in lymphoid cells, presumably underlying the physiological function of lymphoid cells as a surveillance mechanism.

Résumé. On a trouvé, chez le cobaye, que la teneur en acides gras des cellules lymphoïdes isolées de la rate et des ganglions est 5–6 fois plus abondante que celle d'autres tissus. Ces acides sont fortement cytotoxiques pour la cellule cancéreuse d'Ehrlich.

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A Comparison of the Effects of Several Diuretics and Sulfhydryl Reagents on the in vitro Clotting Time of Rat, Guinea-Pig and Human Blood

In a recent communication GADD et al.[1] demonstrated that the organomercurial compounds meralluride, mersalyl and *p*-chloromercuribenzoic acid (PCMB), all capable of reacting with protein sulfhydryl groups, inhibit platelet aggregation induced by adenosine diphosphate (ADP), while ethacrynic acid, a non-mercurial diuretic but also capable of reacting with protein sulfhydryl groups, had no effect. Conversely methyl-mercuric chloride, an organomercurial sulfhydryl reagent, caused aggregation in the absence of exogenous ADP. Because of the central role occupied by platelets in blood coagulation, thrombosis formation and the hemostatic mechanism[2], the effect of these compounds on the clotting time of recalcified whole blood from rat, guinea pig and human was investigated. The results of this study are reported in the present communication.

Materials and methods. Blood was collected from Wistar strain rats and Syrian Random strain guinea pigs by cardiac puncture and from human male volunteers by venipuncture into siliconized glass tubes containing 3.8% sodium citrate. Final ratio of blood/citrate was 9:1.

The silicone clotting time was determined according to the method of CONSTANTINE et al.[3]. The reaction was started by the addition of 0.2 ml of 2.8×10^{-2} *M* $CaCl_2$ (final concentration 9.5×10^{-3} *M*) to siliconized test tubes (10×75 mm) containing 0.2 ml citrated blood and 0.2 ml of the test compound prepared in modified tyrodes solution (Ca^{++}, Mg^{++}-free). Final volume was 0.6 ml. Mixing was achieved by inversion of the test tubes every 30 sec. Clotting time was noted when the blood was unable to flow upon inversion of the test tube[4].

Compounds were obtained from the following sources: Methylmercuric chloride (Alpha Inorganics), *p*-chloromercuribenzoic acid (Mann), mersalyl (Sigma), ethacrynic acid (Merck Sharp and Dohme), meralluride (Lakeside Lab.) and mercuric chloride (Mallinckrodt Chemicals).

Results and discussion. The results of this study are shown in the Table. In contrast to the shortening of the silicone clotting time of rat and guinea pig blood, methylmercuric chloride prolonged the clotting time in all human subjects. In the case of B. St. only a 40% increase was observed as opposed to the greater than 500% increase for all other volunteers. The mercurial diuretic, meralluride prolonged the clotting time in 2 of 4 human subjects, but had no effect on blood from either rat or guinea-pig. A greater than 5-fold increase in the clotting time of blood from all three species was observed in the presence of the other mercurial diuretic, mersalyl. In addition to lengthening the clotting time, mersalyl also caused hemolysis of erythrocytes from rat and guinea-pig blood but had no effect on any of the 4 human blood samples. In contrast to the effects of the 2 mercurial diuretics, the non-mercurial diuretic agent, ethacrynic acid, caused a slight acceleration in the clotting time in 3 out of 4 of the human subjects but had no effect on either of the 2 animal species. The sulfhydryl reagents,

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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