

inhibition of streptomycin activity may be associated with the blocking of an active grouping in the molecule of the streptomycin. To explain the inactivation of the antibacterial properties of streptomycin by the blocking of a single group in its molecule cannot

be attempted at present due to its rather complicated structure. Until the chemistry of streptomycin is more clearly elucidated, it is difficult to present a suitable theory that would account for the various effects of streptomycin inactivation.

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Blood Platelets in Heterophil Anaphylaxis.

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The serum of rabbits into which erythrocytes of sheep have been injected produces toxic symptoms and death when injected into guinea pigs. The reason for this was explained when Forssman¹ and others^{2,3} announced the discovery and distribution of the so-called heterophil antigen. At this time it was found that the same antigenic substance which is present in erythrocytes of sheep is also present in the tissues of the guinea pig, dog and other species. Hence, the antibody produced in the rabbit in response to erythrocytes of sheep would also react with the antigen present in the tissues of the animal injected.

As the reactions exhibited by the animals following such injections of serum are almost identical with those of anaphylaxis, the phenomenon was designated heterophil anaphylaxis.

Later Taniguchi^{4,5,6} observed that the lungs of guinea pigs killed by such injections of serum were markedly hemorrhagic and both

lungs and trachea were filled with frothy fluid but were not markedly emphysematous as in true anaphylaxis. Furthermore Redfern⁷ observed that no contraction is produced in the perfused uterine muscle following the addition of anti-Forssman serum. I have confirmed these facts shown by previous investigators. Moreover I found in collaboration with Recarte⁸ that no increase of blood histamine was present in the acute shock induced in guinea pigs by the injection of anti-Forssman serum and no histamine was liberated when lungs of guinea pigs were perfused with such serums.

The experiments reported in the present paper were made with the aim of studying the modifications of the blood platelets in this particular type of shock. For this the volume of blood platelets was determined in guinea pigs and dogs before and after the injection of rabbit serum with high titer of antishoop hemolysin. At the same time the influence of injections of hepatic glycogen on the course of this type of shock was observed, since this substance in certain doses produces a temporary disappearance of the platelets of the blood.⁹

Methods. Thirteen unanesthetized male guinea pigs weighing from 500 to 1,030 grams were injected in the vein of the penis with vari-

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¹ Forssman, J., and Hintze, Assar, *Biochem. Z.*, 1912, **44**, 336.

² Doerr, R., and Pick, R., *Biochem. Z.*, 1913, **50**, 129.

³ Sachs, H., and Nathan, E., *Z. f. Immunitätsforsch. u. exp. Therap.*, 1913, **19**, 235.

⁴ Taniguchi, Tenji, *J. Path. and Bact.*, 1920, **23**, 364.

⁵ Taniguchi, Tenji, *J. Path. and Bact.*, 1921, **24**, 217.

⁶ Taniguchi, Tenji, *J. Path. and Bact.*, 1922, **25**, 77.

⁷ Redfern, W. W., *Am. J. Hyg.*, 1926, **6**, 276.

⁸ Graña, A., and Recarte, P., *Rev. Soc. argent. de biol.*, 1945, **21**, 202.

⁹ Rocha e Silva, M., Graña, A., and Porto, A., *Proc. Soc. Exp. Biol. and Med.*, 1945, **59**, 57.

TABLE I.
Blood Platelets and Leukocytes in Heterophil Anaphylaxis of the Guinea Pig.

Guinea pig No.	Weight, g	Antisheep rabbit serum, cc/100 g body wt.	Grade of shock	Blood platelets, vol. in 100 cc of blood		Leukocytes, thousands per cu mm of blood	
				Before inj.	After inj.	Before	After
1	500	0.10	Death in 2 min.	1.20	0	—	—
2	510	0.02	No shock	1.30	0	12.4	10.8
3	570	0.07	Death in 1 min.	1.20	0.05	14.8	5.6
4	535	0.01	No shock	0.85	0.85	12.3	11.8
5	530	0.02	" "	0.85	0.20	14.4	5.0
6	500	0.02	Slight dyspnea	0.70	0	18.8	7.8
7	490	0.10	Death in 1 min.	0.80	0	—	—
8	510	0.015	No shock	0.60	0.05	12.0	11.2
9*	590	0.10	Death in 1 min.	0	0	—	—
10*	940	0.05	No shock	0.70	0	—	—
11*	1,030	0.10	Death in 1 min.	0.40	0	—	—
12*	1,000	0.15	Death in 2 min.	0	0	—	—
13*	900	0.10	Death in 1 min.	—	—	—	—

* The last 5 guinea pigs were injected with 1 g of glycogen in the vein of the penis before the injection of antisheep rabbit serum and the platelets were determined after the injection of glycogen and after the injection of serum.

able amounts of rabbit serum containing anti-sheep hemolysin. These serums were prepared by injecting erythrocytes of sheep into the rabbits, standard technics being followed. The hemolytic titer of the serum used in these experiments was 20,000 (that is, 0.00005 ml hemolyzed 0.5 ml of 2% sheep cells in the presence of 0.05 ml of pooled guinea pig serum as complement). Following intracutaneous injections of 0.10 ml of this serum into guinea pigs and dogs a notable hemorrhagic lesion appeared locally.

Five of the 13 guinea pigs used in these experiments were injected in the vein with 1 g of hepatic glycogen, dissolved in 5 ml of water, 3 minutes before the injection of the immune rabbit serum.

Four guinea pigs were injected with normal rabbit serum as controls and the volume of the platelets was determined before, and from 3 to 10 minutes after, the injection.

Different amounts of the immune rabbit serum were injected into 4 dogs anesthetized with pentobarbital sodium and with the femoral artery cannulated in order to determine the blood pressure. The volume of

platelets was determined before and after the injection.

All specimens of blood were drawn directly from the heart of the guinea pigs and from the jugular vein of the dogs. In all the guinea pigs that manifested shock the blood samples were taken immediately following the appearance of symptoms. In animals without shock the samples were taken from 3 to 10 minutes after the injection of serum.

For the determinations of the platelets the Van Allen method¹⁰ was used; in guinea pigs 2 ml of blood were taken in 3 ml of 3.8% solution of sodium citrate with 0.1% of formalin.

In several animals the modification of the leukocytes was studied and standard microscopic methods were used.

Results. From the data presented in Table I it may be observed that the immune rabbit serum employed in these experiments is highly toxic for the guinea pig, since 0.07 ml for each 100 g of body weight injected into the

¹⁰ Van Allen, C. M., *J. Lab. and Clin. Med.*, 1926, 12, 282.

TABLE II.
Blood Platelets in Guinea Pigs Injected with Normal Rabbit Serum.*

Guinea pig No.	Weight, g	Normal rabbit serum, cc/100 g body wt.	Blood platelets, volume in 100 cc of blood	
			Before	After
1	1,030	0.10	1.20	1.0
2	1,000	0.05	1.20	1.0
3	1,170	0.10	1.20	1.20
4	960	0.02	1.40	1.40

* None of these guinea pigs exhibited shock.

TABLE III.
Blood Platelets and Leukocytes in Heterophil Anaphylaxis of the Dog.

Dog No.	Weight, kg	Antisheep rabbit serum, cc/kg body wt.	Grade of shock	Blood platelets, volume in 100 cc of blood		Leukocytes, thousands per cu mm of blood	
				Before inj.	After inj.	Before	After
1	10	0.5	Death in 1 min.	0.60	0	—	—
2	10.3	0.20	Severe shock; death after 90 min.	0.90	0 (4 min.) 0.2 (10 ") 0.5 (90 ")	17.0	1.6 2.6 2.8
3	18	0.10	Slight shock; blood pressure decreased 15 mm Hg	0.60	0.17	7.2	0.6
4	6.8	0.10	Moderate shock; blood pressure decreased 90 mm Hg; recovery in 90 min.	0.55	0.05 (2 ") 0.12 (30 ") 0.35 (45 ") 0.45 (5 hr.)	3.0	0.4 1.0 1.4 4.2

vein caused the death of the animal in one minute.

The lungs of guinea pigs and dogs that died in the course of these experiments showed deep macroscopic hemorrhagic lesions. The microscopic examination showed bronchial exudate in the terminal bronchi and striking adventitial hemorrhage around the larger vessels.

The volume of platelets was always drastically reduced after the injection of 0.02 ml or more per 100 g of body weight of this immune rabbit serum in spite of the fact that injections of small quantities of serum did not produce shock (Table I).

On the other hand, as shown in Table I, 4 of the last 5 guinea pigs which were injected intravenously with 1 g of glycogen, which eliminated all or nearly all of the blood platelets, were killed by an injection, 3 minutes later, of immune rabbit serum.

The experiments described show that in this special type of anaphylaxis the decrease of the blood platelets does not parallel the degree of shock as Kopeloff and Kopeloff¹¹

found in true anaphylaxis.

The reduction of blood platelets under these circumstances raises the question whether the phenomenon observed is specific or nonspecific. As may be seen from Table II, there was no significant change in volume of the blood platelets when normal rabbit serum was injected into guinea pigs.

The sharp fall of the blood platelets after the injection of the immune rabbit serum perhaps depends on a reaction produced in the blood between the antisheep hemolysin injected and the Forssman antigen present in the platelets of the guinea pig.

As is shown in Table III, the same drastic reduction of platelets was observed in dogs after the injection of antisheep hemolysin, although the shock was slight or moderate in dogs 3 and 4. Five hours after the injection of 0.10 ml of immune rabbit serum for each kilogram of body weight the volume of platelets reached approximately the normal

¹¹ Kopeloff, Nicholas, and Kopeloff, Lenore M., *J. Immunol.*, 1941, **40**, 471.

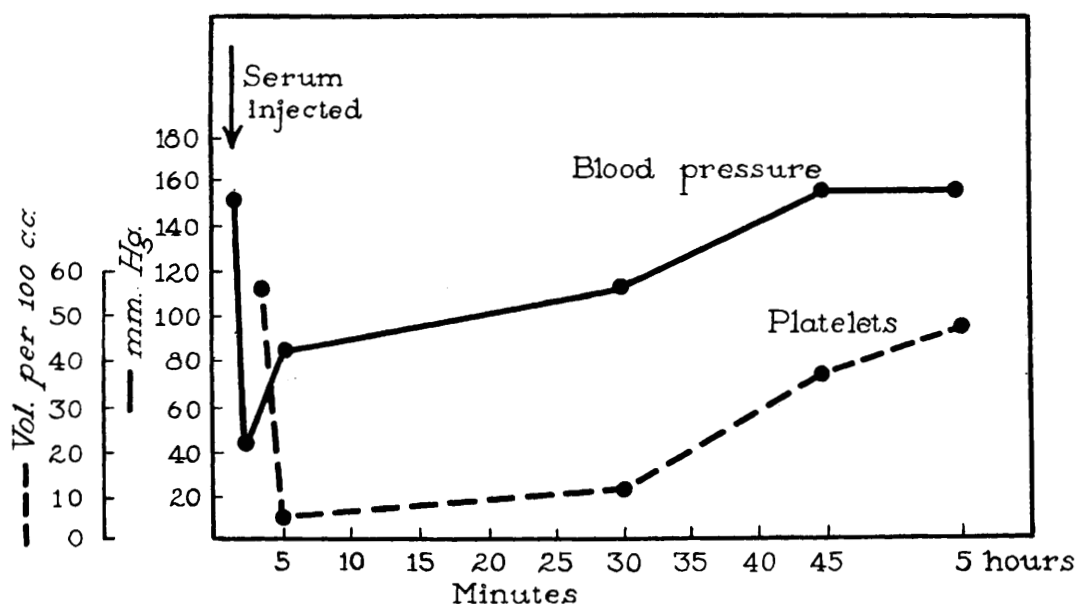


Fig. 1.

Platelets and blood pressure of a dog followed for 5 hours. As seen, 5 hours after the injection of 0.10 ml of immune rabbit serum for each kilogram of body weight the volume of platelets reached approximately the normal values.

values (Fig. 1). In addition to the reduction of platelets there was a drastic reduction of the number of leukocytes in the dog (Table III).

Summary. Serum of rabbits immune to sheep erythrocytes injected intravenously in appropriate doses in guinea pigs and dogs produced a drastic reduction of the concentration of blood platelets even though these quantities of serum provoked only moderate shock or no shock.

When the shock was deep, generally the platelets disappeared from the blood.

The injection of glycogen in doses that reduced the blood platelets in the guinea pig did not prevent the production of shock following the injection of the immune serum.

The experiments here presented show that there is not always a parallel relation between the decrease of blood platelets and the intensity of the pulmonary symptoms in the guinea pig or the decrease of arterial pressure of the dog in heterophil anaphylaxis.

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Subacute and Chronic Toxicities of Ascorbyl Palmitates.

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The fatty acid monoesters of l-ascorbic and d-isoascorbic acid prepared by Swern *et al.*¹ were suggested for use as antioxidants for fats and oils by Riemenschneider *et al.*² In

cooperation* with these investigators we have made a study of the subacute and chronic oral toxicities of the palmitates of these acids.

¹ Swern, D., Stirton, A. J., Turer, J., and Wells, P. A., *Oil and Soap*, 1943, **20**, 224.

² Riemenschneider, R. W., Turer, J., Wells, P. A., and Ault, W. C., *Oil and Soap*, 1944, **21**, 47.

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