time of 12.5% plasma, of oral and intravenous administration of sodium salicylate, and of dicumarol has been compared in rabbits on a normal diet.

The oral and intravenous administration of dicumarol gave the same prolongation of the prothrombin time. However, while the oral administration of 0.5 g/kg of sodium salicylate definitely prolonged the prothrombin time, the intravenous administration of the same dose in the same animal did not result in any change in the prothrombin time. After the oral administration of sodium sulfasuccidine, the oral administration of sodium salicylate did not affect the prothrombin time.

It is suggested as an explanation of these results that salicylate may be converted to dicumarol or a substance with similar prothrombinopenic properties by bacterial action in the intestinal tract.

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Quantitative Aspects of the Inhibition of Anaphylactic Shock in Guinea Pigs.

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While the modifying action of the proprietary compounds β-dimethylaminoethyl benzhydryl ether hydrochloride (benadryl) and N′-pyridil-N′-benzyl-N-dimethylethylenediamine (Pyribenzamine) on histamine and anaphylactic shock has been vigorously investigated,[1] some conflicting data have appeared in the literature with regard to the action of these drugs on true anaphylactic shock in guinea pigs. Mayer, *et al.*,[2] demonstrated that 5 guinea pigs injected with horse serum 21 days before, were protected against an intracardial shock dose of 0.5 cc of horse serum when they were previously treated subcutaneously with 1.0 mg/kg of pyribenzamine. Campbell, *et al.*,[3] reported that although benadryl offered effective protection against histamine shock in the rabbit, it was ineffective in controlling anaphylaxis in actively sensitized (hen egg white) rabbits and guinea pigs. These latter workers used shock doses of 0.75 cc of the antigen for guinea pigs, given intraperitoneally. They conclude that their results seem at direct variance with those of Loew and Kaiser[4] and of Friedländer, *et al.*,[5] since these groups had found, similarly to Mayer's group,[2] that benadryl offers marked protection against anaphylaxis in the guinea pig.

Since in none of this previous work on benadryl and pyribenzamine cited was the attempt made to put protection against anaphylaxis in the actively sensitized animal on a quantitative basis, similar experiments were repeated with this view in mind.

Materials and Methods. The whites of 3 hen eggs were separated from the yolks, strained through cheesecloth into a beaker and then stored in a rubber stoppered vaccine bottle in the refrigerator. This material (Pro-

* The Rackham Arthritis Research Unit is supported by the Horace H. Rackham School of Graduate Studies of the University of Michigan.

1 Feinberg, S. M., *J. A. M. A.*, 1946, **132**, 702.

2 Mayer, R. L., Huttrer, C. P., and Scholz, C. R., *Science*, 1945, **102**, 93.

3 Campbell, B., Baronofsky, I. D., and Good, R. A., PROC. SOC. EXP. BIOL. AND MED., 1947, **64**, 281.

4 Loew, E. K., and Kaiser, M. E., PROC. SOC. EXP. BIOL. AND MED., 1945, **58**, 235.

5 Friedländer, S., Feinberg, S., and Feinberg, A. R., PROC. SOC. EXP. BIOL. AND MED., 1946, **62**, 65.

tein = 11.53 g/%; NPN = 21.66 mg/%†) served as the source of sensitizing and shocking antigen throughout these experiments. The guinea pigs used weighed between 300 and 400 g. They received sensitizing injections of 0.1 and 0.2 ml of the egg white intraperitoneally and were kept in large common cages until ready for use. Intracardial injections were made using tuberculin syringes with No. 20 or 21, one or one and one-half inch needles. No anesthetic was employed. After proving the presence of the hypodermic needle in the heart by withdrawing 0.2 to 0.4 cc of blood, the egg white and blood mixture was reinjected slowly, within 30 seconds.

Dilutions of the egg white were made after centrifuging 3 to 4 cc of the refrigerated stock supply. The dilutions were made in saline with 1.0 ml serological pipettes graduated in hundredths and were in the range of the probable lethal dose (L. $D._{100}$) as determined by earlier experience.

The pyribenzamine and benadryl used were the commercial preparations put up for oral use and were incompletely soluble in 0.9% sodium chloride solution. These preparations were kept in test tubes and shaken to insure homogeneous suspension before placing in a syringe for intraperitoneal injection.

TABLE I

Determination of LD_{100} and Maximum Non-Lethal Dose of Egg White Required to Shock Actively Sensitized Guinea Pigs (19–21 days; 300–400 g) by Intracardial Injection.

No. of Animals	Antigen Dilution ml	Result*
1	.1 of 1:16	0
8	.1 1:8	1, 1, 1, 1, 1, 1, 1, 2
3	.2 1:8	3, 3, 3
6	.1 1:4	3, 3, 3, 3, 3, 3

* Results of intracardial injection were numbered as follows:
0, no symptoms noted.
1, mild symptoms. Face washing, coughing or sneezing, animal survives.
2, more severe symptoms, strong inspiratory movements, incontinence, animal survives.
3, severe symptoms and death within 3–8 minutes. Massive emphysema of lungs at autopsy.
4, severe symptoms, prostration followed by death after a delayed period of 15 to 60 minutes or more. Lungs not markedly emphysemic, but show some congestion. Congestion of kidneys, adrenals and mesenteric blood vessels. Beet-red appearance of stomach and intestinal outer wall. Lining of peritoneum dark pink to red.

† Nitrogen determinations (Kjeldahl) were made after concluding the experiments. The egg white was inadvertently frozen before the nitrogen determinations were made.

Results. Table I illustrates a typical titration to determine the L. $D._{100}$ of the egg white antigen used in these experiments. Among this group of animals sensitized 19, 20, or 21 days previously, none survived the intracardial injection of 0.1 ml of a 1:4 dilution or its concentration equivalent and none died of the effects of 0.1 ml of a 1:8 or higher dilution. The conclusion reached was that 0.1 ml of a 1:4 dilution represented an L. $D._{100}$ dose.

Table II shows the results obtained when sensitized guinea pigs, protected by pyribenzamine were injected with the same dilutions of egg white on the same day that the control animals were injected. These animals were injected intraperitoneally with the pyribenzamine 10 to 30 minutes preceding the shocking dose of egg white. They received either 5 or 10 mg of the drug with no attempt being made to vary the drug on a weight basis. Among 8 guinea pigs receiving 8 L. $D._{100}$ doses of egg white, 2 died after a delayed period and necropsy showed what appeared to be the systemic result of anaphylaxis in the guinea pig. The other 6 animals exhibited mild to severe symptoms, but lived in each case. Among 4 animals receiving less than 8 L. $D._{100}$ doses (*i. e.*, 1, 4, and 6) none exhibited symptoms of any type. Among 3 animals receiving more than 8 L. $D._{100}$ doses (*i. e.*, 20, 12, 10) all died within 7 minutes, in every way similarly to the control animals.

Table III illustrates the results obtained with sensitized guinea pigs protected against shock by benadryl which was intraperitoneally injected 15 to 30 minutes preceding the dose of egg white. Again, 5 or 10 mg of the drug was given with no effort being made to vary dosage according to weight of the animal. Among 4 animals receiving 8 L. $D._{100}$ doses none died although all exhibited mild to severe symptoms. One animal injected with 10 L. $D._{100}$ doses 20 minutes after receiving 10 mg of benadryl had violent symptoms but survived the shock and lived. Five others

TABLE II
Effect of Pyribenzamine on Amount of Egg White Required to Shock Actively Sensitized Guinea Pigs (300–400 g) by Intracardial Injection.

Animal No.	Days after Sensitization	Antigen Dilution ml	No. of Doses LD_{100}	Premedication: Intraperitoneal Dose mg	Premedication: Minutes Shocking Before	Result*
1	19	.1 of 1:4	1	5	10	0
2	19	.4 1:4	4	5	15	0
3	19	.4 1:4	4	5	20	0
4	19	.8 1:4	8	5	15	2
5	19	.8 1:4	8	5	15	4
6	19	.4 1:2	8	5	15	1
7	19	.4 1:2	8	10	15	4
8	19	.8 1:4	8	10	20	1
9	19	.4 1:2	8	10	20	1
10	19	.5 1:2	10	5	30	3
11	19	.6 1:2	12	10	15	3
12	19	1.0 1:2	20	5	20	3
13	20	.6 1:4	6	10	30	0
14	20	.8 1:4	8	10	15	1
15	20	.4 1:2	8	10	20	1

*Code same as that used in Table I.

TABLE III
Effect of Benadryl on Amount of Egg White Required to Shock Actively Sensitized Guinea Pigs (300–400 g) by Intracardial Injection.

Animal No.	Days after Sensitization	Antigen Dilution ml	No. of LD_{100} Doses	Premedication: Intraperitoneal Dose mg	Premedication: Minutes Before Shocking	Result*
1	20	.1 of 1:4	1	5	20	0
2	20	.4 1:4	4	5	25	0
3	20	.5 1:2	10	5	35	3
4	21	.4 1:4	4	10	20	0
5	21	.6 1:4	6	10	20	0
6	21	.4 1:2	8	5	15	2
7	21	.4 1:2	8	10	25	1
8	21	.4 1:2	8	10	30	1
9	21	.4 1:2	8	10	15	1
10	21	.5 1:2	10	10	20	2
11	21	.5 1:2	10	10	25	3
12	21	.6 1:2	12	5	25	3
13	21	.6 1:2	12	5	25	3
14	21	.6 1:2	12	10	25	3

*Code same as that used in Table I.

which received more than 8 L.D.$_{100}$ doses (*i. e.*, 12 or 10) all died within 7 minutes, exactly as had the control animals. Four animals receiving less than 8 L.D.$_{100}$ doses (1, 4, 6) exhibited no symptoms of shock.

Summary and Conclusions. The L. D.$_{100}$ dose of a solution of hen egg white by intracardial injection was determined for a group of actively sensitized guinea pigs. It was found that 5 to 10 mg of pyribenzamine or benadryl injected intraperitoneally between 10 and 30 minutes before giving the shocking dose protected the animals against approximately 8 L. D.$_{100}$ doses of the antigen while injection of 10 or more L. D.$_{100}$ doses resulted in rapid death with symptoms indistinguishable from the control animals. If 6 or less L. D.$_{100}$ doses were given, no symptoms were noted in the protected group.

These results seem to indicate that the so-called anti-histaminic drugs do exert a protective influence, quantitative in nature, against anaphylaxis in the actively sensitized guinea pig. This fact might well be expected since a similar quantitative protection is obtained with these drugs against histamine

shock.[1,2,3] The modifying action of these drugs again seems in accord with the theory that histamine plays a major role in anaphylaxis. It is of interest that approximately the same degree of protection was offered, in these experiments, by both the compounds used.

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Immunization of Mice Against Viruses of the Psittacosis Group with Ultraviolet-Inactivated Vaccines.*

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Various workers have demonstrated[1-4] that formalinized suspensions of psittacosis virus possess antigenicity as judged by their capacity to elicit active immunity in mice and avian species. No report has appeared, to our knowledge, of the effect of ultraviolet rays on this or other viruses of the psittacosis group. Levinson, Milzer, and co-workers, employing a new method of inactivating turbid suspensions of viruses and bacteria by ultraviolet rays, have found that it is possible to prepare experimental vaccines[5-7] with dysentery bacilli, and rabies, St. Louis encephalitis, and poliomyelitis viruses with a minimal loss of antigenicity. Briefly, this technic consists of exposing a continuously flowing thin film of suspension to the Oppenheimer-Levinson type lamp which is a powerful source of total and extreme (below 2000 Angstroms) ultraviolet energy. This method of inactivation was employed in the present study with the viruses of psittacosis (6BC), human pneumonitis (SF), and ornithosis (207).

Previous experiments[8] have indicated that these viruses can be completely inactivated by this method and still retain antigenicity as determined by the production of neutralizing antibody in chickens repeatedly inoculated with these preparations. It was our purpose here to make a preliminary investigation of the possibility of using such preparations for the induction of resistance against challenge injections of active virus.

Materials and Methods. The strains of virus employed have been used in previous work in this laboratory and their sources have been recorded elsewhere.[9] Earlier studies[10-12] have shown that large amounts of virus in relatively clear suspensions can be obtained by harvesting the allantoic fluid from embry-

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