trations in blood varying from less than 0.1 mg. to more than 1 mg. of lead per 100 cc. Normal values are definitely below 0.1 mg. per 100 cc. of serum.

TABLE I.		
Sample Determinations of Lead in Human	Blood S	era.
Values expressed as mg. Pb ⁺⁺ /100 cc.	Serum.	

Dia	agnosis	Subject	Lead Found
Lead F	Poisoning	Adult	.7 .3
,,	,,	,,	.ī
,,	"	,,	1.0
,,	,,	,,	.2
,,	,,	,,	.5
Normal	+ 0.5 mg. Pb in 100 cc.	Child	.03 appr.
,,	+ 0.3 mg. Pb in 100 cc.	,,	.3
"	+ 0.2 mg. Pb in 100 cc.	,,	.2

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Demonstration of Hemorrhagins in Snake Venom by Means of the Chicken Embryo.

ERNST WITEBSKY, SAMUEL PECK, AND ERWIN NETER. (Introduced by Louis Gross.)

From the Laboratories of The Mount Sinai Hospital, New York City.

The venoms of various species of snakes differ in their content of toxic principles such as hemorrhagins, hemolysins and neurotoxins. While the hemolysins and neurotoxins can easily be determined, a reliable method for the demonstration of hemorrhagins has been lacking.

In the course of experiments with the chicken embryo it occurred to us that such a preparation might lend itself as a suitable test object for the study of hemorrhagins in venoms.

Fertilized chicken eggs are placed in an incubator at 42°C. for 3 days. The egg is opened preferably at the small pole; one-third or half of the shell is carefully removed and the egg poured into a beaker. Then, the beaker is covered with a watch glass and kept in the incubator at 37°-38°C. The body of the 3-day-old chicken embryo is surrounded by a vascular network of 3-4 cm. in diameter (Fig. 1).

A 1% solution of moccasin venom (Ancistrodon piscivorus) in physiological saline was prepared. Serial dilutions were then made up to 1.0 cc. with physiological saline. These were placed in the

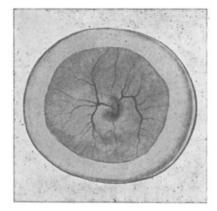


Fig. 1.

incubator at 37°C. for a few minutes and then added drop by drop to the chicken embryo. The embryo was returned to the incubator.

The addition of a sufficient concentration of moccasin venom to the chicken embryo causes petechial hemorrhages at various sites of the vascular network (Fig. 2). This phenomenon manifests itself in a few minutes. The hemorrhagic points rapidly increase in size and number (Fig. 3). Occasionally complete exsanguination of the

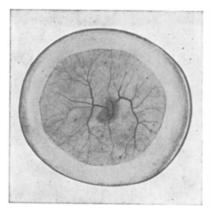




Fig. 2. 3 minutes.

Fig. 3. 7 minutes.

embryo was observed from only one large hemorrhage. With the increase of hemorrhages the blood vessels lose their bright red color, their contours become less distinct and very soon the whole embryo assumes a dull glazed appearance which is followed by death.*

^{*} Addition of one of the moccasin venoms (1%) caused a damage to the membrane of the egg yolk, so that its content ran out.

		The	hemori	rhagie e	TABLE 1. The hemorrhagic effect of moccasin venom on the chicken embryo.	TAE	TABLE I. noceasin ver	nom on	the ch	icken	embryo				
Venom I Amounts (Vol. 1.0 cc.)	63	4	9	∞	Tim 10	le in m	in. afte	Time in min. after addition of venom dilutions. 10 12 14 16 18 20 22 24	ion of 18	venor 20	diluti 22	ons. 24	56	88	30
(1) 1 (2) 1/3 (3) 1/9 (4) 1/27 (5) 1/81	*	+	++ ;;	++	++ ++ +		+	+		++!	++	++	++	++ ++ ++	
Venom II															
(1) 1 (2) 1/2 (3) 1/4 (4) 1/8 (5) 1/16	1111	11111	+	++111	+ + + + + + + + + + +	+ ++ +	++11	+ + +	+1	+1	+ + + + +	+ + +	l		
*—= no hemorrhage. ++= strong hemorrhage.	rrhage.	hage.	ap	pearan ++=	+ = appearance of the first petechiae. +++ = death of the embryo.	e first f the	petechi embryo	ae.							

Table I gives 2 illustrations of a series of 11 similar experiments. The data in the table represent observations on duplicate test objects. Physiologic saline in equal amounts, as well as a 1% solution of Bothrox atrox was added to the chicken embryos as control. Bothrox atrox did not give a purpuric reaction.

Furthermore, it can be seen from the table that there is a relationship between the concentration of the venom solution and the rapidity with which petechiae first appear. The latent period increases with the decrease of venom concentration. Once the hemorrhage is initiated, however, there is no striking difference to be observed as far as the degree and the number of hemorrhagic manifestations are concerned. Higher dilutions of the moccasin venom which just fail to produce hemorrhages in the chicken embryo sometimes caused bradycardia and death.

Summary. The addition of moccasin venom in sufficient concentration to 3-day-old chicken embryos produces hemorrhages in the vascular network. This method may be used for the demonstration of hemorrhagins in venoms. Further studies should be made to determine whether this method is applicable for the quantitative determination of hemorrhagins.

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Influence of Drugs on Chicken Embryo and on Its Reaction to Sera Containing Forssman Antibodies.

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The addition of sera containing Forssman antibodies to 3-day-old chicken embryos which are taken out of the eggshell and kept in beakers, causes the following phenomenon: The vascular network contracts, the embryo turns around, sinks into the yolk and finally dies (Baumann and Witebsky¹). This phenomenon is to be interpreted as the sequela of a reaction between the Forssman antigen of the chicken embryo and the corresponding antibodies of the added serum. It can be produced only by sera containing Forssman antibodies (normal "rabbit type" sera and Forssman antiserum). Thus, we are dealing with a phenomenon which parallels the so-called inverted anaphylactic shock of guinea pigs following the injection of Forssman antiserum.

The nature of the mechanism of anaphylactic phenomena is still a moot question. Some authors claim that they are due to physicochemical changes of the cells following antigen-antibody reaction;

¹ Baumann, A., and Witebsky, E., Compt. rend Soc. Biol., 1934, 116, 10; Annales de l'Inst. Pasteur, 1934, 53, 282.