not be maintained by cortical hormone alone in the one hypophysectomized animal thus treated from Series III.

Series VII. Lactation could not be maintained (as judged by the death of suckling litters) in 3 hypophysectomized, lactating rats treated with cortical and purified lactogenic hormones. This result was expected in view of the fact that even with crude pituitary extracts we could not maintain lactation in 12 such rats.

Summary. A purified lactogenic hormone, unlike crude pituitary extracts, has failed to initiate lactation in hypophysectomized guinea pigs. Lactogenic hormone plus the adrenal cortical hormone (Swingle-Pfiffner extract) has initiated lactation in such animals in every case.‡

8746 C

Production of Shwartzman Reaction in Rabbits with Purified Fraction of *B. coli* Filtrate.

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Previous publications[1, 2] from this laboratory described a method for the separation and concentration of the factor in *B. coli* filtrates which produces hemorrhage in mouse tumors. Since *B. coli* filtrates are known to produce the Shwartzman reaction in rabbits, it was of interest to ascertain whether the purest fraction of the tumor-affecting agent obtained thus far[2] was also capable of eliciting hemorrhage in prepared sites in the skin of rabbits. It was also desirable to make quantitative comparisons between the original filtrate and the purified fraction as to their ability to produce hemorrhage in tumors of mice as well as the Shwartzman reaction in the rabbit.

Equivalent tumor-affecting doses were determined by performing

‡ Judging by gross observation at autopsy, anterior lobe removal was complete in all of these cases. To verify this, however, sections of the sella turcica contents have been made in most cases, although not yet complete in a few cases. We have omitted in this account those animals, a considerable number, that failed to survive the operation long enough to allow for the necessary observations.

1 Shear, M. J., and Andervont, H. B., PROC. SOC. EXP. BIOL. AND MED., 1936, **34**, 323.

2 Shear, M. J., *ibid.*, 1936, **34**, 325.

a preliminary titration in mice bearing cutaneous tumors, according to a technic described elsewhere.[3] Strain M mice bearing 6-day-old cutaneous growths of sarcoma 37 were given intra-abdominal injections of diluted filtrate or dilutions of the purified fraction. All dilutions were made with 0.9% NaCl solution. It was found that 0.2 cc. of a 1:50 dilution of the original filtrate produced hemorrhage in tumors, but 0.2 cc. of a 1:100 dilution failed to do so. The purified fraction was active down to 0.2 cc. of a 1:1500 dilution of a solution which contained 2 mg. of the purified fraction per cc. Thus the minimal tumor-affecting dose of this purified fraction was 0.26γ.

Comparable doses of the two materials were then injected into rabbits according to Shwartzman's technic. The endermal injections consisted of 0.2 cc. doses. Each of 4 rabbits received preparatory injections of 3 dilutions of the original filtrate and of 3 dilutions of the purified fraction, as shown in Table I. Twenty-four hours later 2 of the rabbits each received 0.5 cc. of undiluted filtrate intravenously while the other 2 received 0.5 cc. of a 1:10 dilution of the purified fraction. The results are shown in Table I.

It is seen that both the filtrate and the purified fraction elicited

TABLE I.

Response of Cutaneously Prepared Rabbits to Intravenous Injection of *B. coli* Filtrate and of a Purified Fraction Obtained from the Filtrate.

		Preparatory endermal injection (0.2 cc.) in all 4 rabbits					
Rabbit No.	Intravenous injection	Original filtrate (undil.)	Original filtrate (1:10)	Original filtrate (1:50)	Purified fraction, 0.04 mg.	Purified fraction, 0.002 mg.	Purified fraction, 0.0005 mg.
		Results					
467	Purified fraction, 0.1 mg.	+++	+++	+++	++++	+++	++
468	Purified fraction, 0.1 mg.	++	+	—	+++	++	+
469	Original filtrate, 0.5 cc. (undiluted)	++	—	—	+++	—	—
470	Original filtrate, 0.5 cc. (undiluted)	+++	—	—	++++	—	—

Note: In the above table + denotes a positive Shwartzman reaction and — a negative Shwartzman reaction. The number of + signs are used to denote the relative size of the areas covered by the reaction.

[3] Andervont, H. B., *Am. J. Cancer*, 1936, **27**, 77.

the Shwartzman reaction in rabbits. However, the purified fraction appears to be more potent than the filtrate in producing the reaction, since intravenous injection of a 1:10 dilution of the stock solution of the purified fraction produced the reaction at practically all sites of endermal injection of both filtrate and of purified fraction while intravenous injection of the undiluted filtrate elicited the reaction only in those sites that had received the highest concentration of filtrate or of purified fraction.

It is of interest to note that, so far as the purified fraction is concerned, the greater the amount of tumor-affecting agent employed in the preparatory injections the more pronounced is the Shwartzman reaction. It may also be pointed out that, in this experiment, a positive Shwartzman reaction was obtained with an amount of purified fraction (0.0005 mg.) that is close to the minimal amount (0.0003 mg.) effective in eliciting hemorrhage in mouse-tumors.

8747 P

Chemical Composition of *Bacterium tumefaciens.*

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Bacterium tumefaciens is the etiological agent of plant tumors, known as crown-gall disease.[1] The importance of investigating the chemical composition of this organism and the pathological effects of its various constituents is apparent. Although the etiology of plant tumors probably does not parallel that of animal cancer, the fact that a chemical stimulant of cellular proliferation apparently is synthesized by this microörganism is reminiscent of certain results obtained with chemically well-defined carcinogenic agents. Doubtless, a deeper knowledge of the mechanism underlying the development of plant cancer would also be of value for a better understanding of malignant growth in general.

A virulent strain of *B. tumefaciens* (strain 6NIS[6]) was cultivated on sterile bean-broth in the dark at room temperature for 14 days. The washed bacteria were suspended in a mixture of equal amounts

[1] For discussion and literature compare M. Levine, *Am. J. Cancer*, 1931, **15**, 1410.