reported an increased rate of gain in rats fed lyxoflavin as compared to the basal group. From the results of the experiments reported herein, it appears that lyxoflavin also has growth promoting activity in baby pigs when the basal ration contains a minimum amount of fat and contains protamone to enhance the deficiency state.

It appears possible that lyxoflavin may be a new member of the vit. B complex with a biological role different from that of riboflavin and other known vitamins. However, the fact that the baby pigs do quite well on the basal ration which is devoid of any known lyxoflavin might indicate that lyxoflavin exerts its effect in a manner similar to that of the antibiotics or of the surfactants(9) or other growth stimulants, rather than due to the vitamin nature of the compound. The high amount of the lyxoflavin fed which was excreted in the urine might indicate tissue saturation and implies that a lower level would have been as effective as the level used. In the case of riboflavin approximately 30% of an intake of 11 μg per g was excreted in the urine as compared to 68.1% of 4 μg per g for lyxoflavin.

Lyxoflavin is apparently not converted into riboflavin in the animal body as Emerson and Folkers(8) report that synthetic lyxoflavin is devoid of riboflavin activity in rats by the standard assay. We have confirmed this by *in vitro* studies and have also confirmed their report that lyxoflavin is inactive for *Lactobacillus casei*.

It appears that some lyxoflavin may be stored in the heart, however, the amount is small and with a differential assay it is more difficult to determine these minute quantities than with a direct assay.

Summary. 1. The addition of 4 μg of synthetic l-lyxoflavin per g of dry matter consumed in 3 experiments significantly increased the rate of gain and efficiency of feed utilization of baby pigs fed a low fat basal ration including 0.01% protamone. 2. Using a differential assay procedure for lyxoflavin it was found that approximately 67% of the lyxoflavin intake was excreted in the urine. 3. *In vitro* studies of heart and liver tissue did not indicate any conversion of riboflavin into lyxoflavin.

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Shwartzman Phenomenon II. Suppressive Action of HN_2 on Antigen-Antibody Provocation. (19470)

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The basic experiment demonstrating the phenomenon of local tissue reactivity (Shwartzman phenomenon) consists of the intradermal injection of a small amount of a suitable bacterial filtrate (preparatory injection), followed after some 24 hours by an intravenous injection of a quantity of the same material (provocative injection). In susceptible rabbits, a zone of hemorrhagic necrosis appears at the site of the preparatory injection 3 to 5 hours after the intravenous injection of filtrate(1). The phenomenon can

also be elicited when the intravenous injection of bacterial filtrate is replaced by a challenging intravenous injection of antigen into a sensitized animal (antigen-antibody reaction *in vivo*)(1). The mechanisms through which these agents provoke the phenomenon seem to be different, at least in part, since Shwartzman has shown that ACTH strongly suppresses the reaction of hemorrhagic necrosis when the phenomenon is provoked by an intravenous injection of a bacterial filtrate (2), but has no effect when an antigen-antibody reaction *in vivo* is substituted for the latter(3).

The reported contrast in the reactions to the provocative agents in rabbits treated with ACTH prompted a similar comparison when a nitrogen mustard is used as the suppressive agent. Methyl bis β-chloroethyl amine hydrochloride has been shown by Becker to suppress completely the reaction of hemorrhagic necrosis when 2 mg/kg are given intravenously 4 days before the intravenous provocative injection of bacterial filtrate(4). The observation has been amply confirmed (5,6), but in none of the studies on mustard suppression reported heretofore has the antigen-antibody reaction *in vivo* been used to provoke the phenomenon.

Methods and materials. Rabbits of both sexes and of various strains, weighing 1800 to 3000 g were used. The animals were maintained in separate cages on routine laboratory feed and water *ad lib*. *Skin preparation.* The abdominal skin of the rabbit was lathered and shaved just prior to the preparatory intradermal injection. The preparatory injection consisted of 0.5 ml of *Meningococcus 44B* filtrate,* 0.5 ml of a filtrate of *E. coli*, or 0.25 mg of a partially purified polysaccharide of *S. typhimurium*. The injections were made about 24 hours before the challenging intravenous antigen injection and the preparatory substances were varied to obviate acquired immunity when repeated experiments were done on the same animal. *Nitrogen mustard.* Methyl bis β-chloroethyl amine hydrochloride, (HN_2) was injected intravenously. Solutions were freshly prepared before each group of injections and were made up to 1 mg/ml in isotonic saline or distilled water. Two mg/kg of the agent were injected 4 days before the challenging intravenous injection of antigen. *Antigen-antibody reactions.* One or two sensitizing injections of 2 ml of fresh human serum were given intravenously to each rabbit. An interval of 7-10 days elapsed when two sensitizing injections were used. Seven to ten days after the last sensitizing injection, and 24 hours after the intradermal preparatory injection, a challenging injection of 2 ml of serum was given. Small groups of rabbits served alternately as mustard-treated and control subjects, resting 10-30 days between experiments. Each group was used 2 or 3 times.

TABLE I. HN_2 Suppression of Shwartzman Phenomenon Provoked by Antigen-Antibody Reaction *in vivo*.

	No. of exp.	Pos.	Neg.
Controls	19	14	5
HN_2-treated	15	0	15

Results. The results of the experiments are summarized in Table I.

Conclusion. It is evident from the data presented that HN_2 completely suppresses the reaction of hemorrhagic necrosis when an attempt is made to provoke the Shwartzman phenomenon by an antigen-antibody reaction *in vivo*.

* Kindly supplied by Dr. G. Shwartzman.

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