juice studied *in vitro* showed that the lower the pH of the specimen, the greater the inactivating effect. That pepsin was not responsible for the destruction of the penicillin was shown by its failure to cause a decrease in the concentration of the active substance in concentrations of 0.2%.

These studies suggested, then, that penicillin should be absorbed when administered by mouth to subjects with achlorhydria. In 2 patients with pernicious anemia reported herein, the concentration obtained in the serum and the amount excreted in the urine showed that penicillin was absorbed readily. The curve of the plasma concentrations in subject A was not unlike that obtained after intramuscular and intraduodenal administration.[1]

Saliva, bile, and succus entericus did not exhibit any inactivating effect on penicillin, suggesting that oral administration of penicillin might be accomplished effectively by the administration of large amounts of alkali or by the use of enteric coated capsules. The latter method is not likely to prove effective since unless the capsule is dissolved in the upper gut the bacteria in the lower intestines may inactivate the penicillin.[1]

Conclusion. Saliva, bile, and succus entericus obtained from man do not inactivate penicillin. Gastric juice destroys this antibacterial substance rapidly especially at body temperature. Hydrochloric acid, and not pepsin, is apparently responsible for this action of gastric juice.

In 2 subjects with pernicious anemia the absorption of penicillin, when administered by mouth, was greater than that observed in normal subjects.

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Vascular Fragility and Permeability. II. Influence of Fasting and Bile Salts on Induced Pulmonary Hemorrhage.*

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When normal mice on a regular laboratory diet are subjected to sudden and extreme reduction of atmospheric pressure, they die, and their lungs show extensive hemorrhage. This fact was utilized in testing the ability of certain mixtures derived from lemon or orange peel to decrease capillary fragility.[1]

While comparing the effects of several of these mixtures, some of the mice were left for 24 hours to observe any delayed effect they might exhibit. It was noted, however, that the mice in one lot refused food. When these were killed and post mortem examination revealed absence of hemorrhage, it appeared that the "protection" might be due to fasting. Accordingly, one lot of mice was set aside and deprived of food, but allowed free access to water. At the end of 24 hours they were subjected to the partial vacuum, along with others which had received the regular diet. The fed mice showed the usual hemorrhage of the lungs, but the fasted mice showed much less. By extending the fasting time to 48 hours, the difference between the fasting and fed animals was increased.

The fact that the animals which had been taking food showed hemorrhage, suggested that the effect might be associated with the process of digestion, and prompted an investigation of the role which bile salts might play.

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1 Majovski, George J., Lesser, A. J., Lawson, Homer C., Carne, Herbert O., and Thienes, C. H., *J. Pharm. and Exp. Therap.*, in press.

The following experiment shows the effect of bile salts on the degree of hemorrhage produced in fasted animals.

Method. The mice selected were adults weighing about 20 g. One lot was fasted for 24 hours, and later lots for 48 hours. After fasting, one-half the lot were given bile salts† in the dosage of 0.1 mg/g body weight, in a 1% solution in distilled water, by stomach tube. The remainder were given an equivalent amount of distilled water. This dosage of bile salts is in accord with the figures of Whipple and Smith[2] for the amount of bile salts circulating in the body. The observation of these authors that there is a lag in the decrease of bile salts after starting a fasting regime, is borne out by the greater effect on hemorrhage of extending the fasting period to 48 hours.[3]

One hour after administering the solution, the mice were placed in the killing chamber. This was a small glass chamber provided with a stop-cock to open and shut a communication with a 5-gallon bottle evacuated by a water pump. A mercury manometer measured the pressure in the system. When the stop-cock was opened, the air pressure in the chamber promptly dropped to about 70 mm of mercury, which is the atmospheric pressure at an elevation of about 54,000 feet above sea level. After 2 minutes they were removed, and the lungs examined. The degree of hemorrhage was estimated, and evaluated in the following manner: lungs showing a normal appearance, of pale pink color, with no bleeding points, no hemorrhagic areas, and no evidence of petechiæ nor extravasation of blood into the pleural cavity, were rated "zero." Lungs showing petechiæ, but no bleeding points nor hemorrhagic areas, were rated "plus-minus." Lungs showing any bleeding points, or hemorrhagic areas involving not more than one-tenth of the total lung area were rated "one plus." Lungs showing hemorrhagic areas involving less than one-half of the total lung area, but of a greater extent than those rated one plus, were rated "two plus." Lungs showing hemorrhagic areas involving one-half or more of the total lung area, but less than virtually the entire lung area, were rated "three plus." Lungs showing hemorrhage involving the entire lung, or all but a minimal area of the lung tissue, were rated "four plus." The average degree of hemorrhage in a series was determined by adding the number of "plusses" (evaluating "plus-minus" as ½), and dividing the total by the number of mouse lungs represented in the series. This figure was multiplied by 25, so that the degree was expressed as per cent, *e.g.*, "four plus" represented total hemorrhage, or 100%.

The abdominal contents were likewise examined. In the fasted animals, the stomach was empty; the gall bladder was distended with

TABLE I.
Pulmonary Hemorrhage in Fasted Mice.

	Bile salts, 0.1 mg/g	No bile salts
24-hr fast	++++	++
	++	+
	++	±
	—	—
% hemorrhage	67	26
48-hr fast	++++	+
	++++	+
	++++	+
	++++	+
	+++	+
	+++	±
	+++	±
	+++	±
	+++	±
	++	±
	++	0
	++	0
	+	0
		0
		0
	—	—
% hemorrhage	73	13
	Bile salts, 0.01 mg/g	
48-hr fast	++++	+++
	++++	++
	+++	±
	+	±
	+	±
	+	±
	±	0
	±	0
	±	0
	±	0
	—	—
% hemorrhage	40	18

† Difco Laboratories.

2 Whipple, G. H., and Smith, H. P., *J. Biol. Chem.*, 1928, **80**, 697.

3 Smith, H. P., Groth, A. H., and Whipple, G. H., *J. Biol. Chem.*, 1928, **80**, 659.

bile, but the intestines showed no color that would indicate the presence of bile.

The results of the experiment are shown in Table I. It is apparent that 0.1 mg of bile salts per gram body weight resulted in the appearance of a much greater degree of hemorrhage in the fasted animals, and that even one-tenth this amount had a definite effect.

Summary. 1. Sudden reduction of atmospheric pressure to 70 mm Hg caused pulmonary hemorrhage and death, in mice. 2. Fasting had the effect of reducing the degree of hemorrhage. 3. Bile salts, given to the fasted animals increased the degree of hemorrhage.

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Induction of Decidua-Placental Hemorrhage in Mice by the Endotoxins of Certain Gram-Negative Bacteria.

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Goldman[1] observed that trypan blue intravenously injected localized in parts of the placenta and in certain of the foetal membranes of mice, but not in the embryos. These observations were in general confirmed by Burrows[2] and by Everett.[3] Paralleling these reports many studies have indicated that colloidal acid-azo dyes in the same class as trypan blue, when injected intravenously, localize in traumatized areas, as well as in certain induced or naturally-occurring lesions. Among these studies was the observation that such dyes localize in neoplasms.

Zahl and Waters[4] made a histological study of the localization of various dyes in mouse sarcoma 180, extending similar earlier observations by Ludford,[5] Duran-Reynals,[6] Brunschwig, Schmitz and Clarke.[7] In the course of this study it was observed that certain bacterial products would, when injected into mice bearing implanted sarcoma, induce hemorrhage of the tumor, often leading to its regression.

Initiated by this observation, a series of studies by Zahl, Hutner and co-workers[8] demonstrated that the endotoxins of almost all gram-negative bacteria when injected into tumor-bearing mice would induce tumor hemorrhage, and that this effect was consistent with the view that endotoxins of gram-negative bacteria are primarily vascular poisons.

Stemming from the apparent resemblance between the localization of colloidal dyes in tumors and the localization of such dyes in placental structures, the following study was undertaken to determine whether a parallel also existed in respect to hemorrhage induction in tumors and hemorrhage induction in placental tissues of mice following injection of the endotoxins of gram-negative bacteria.

Experimental. Three representative gram-negative bacterial species were selected as sources of endotoxin.* These were *Shigella*

1 Goldman, E. E., *Beitr. z. klin. Chir.*, 1909, **64**, 192.

2 Burrows, Harold, *Some Factors in the Localization of Disease in the Body*, Bailliere, Tindall and Cox, London, 1932.

3 Everett, J. W., *J. Exp. Zool.*, 1935, **70**, 243.

4 Zahl, Paul A., and Waters, L. L., PROC. SOC. EXP. BIOL. AND MED., 1941, **48**, 304.

5 Ludford, R. J., *Proc. Roy. Soc. London*, Ser. B, 1929, **104**, 493.

6 Duran-Reynals, F., *Am. J. Cancer*, 1939, **35**, 98.

7 Brunschwig, A., Schmitz, R. L., and Clarke, T. H., *Arch. Path.*, 1940, **30**, 902.

8 Zahl, Paul A., Hutner, S. H., Spitz, S., Suguira, K., and Cooper, F. S., *Am. J. Hyg.*, 1942, **36**, 224; Zahl, Paul A., and Hutner, S. H., PROC. SOC. EXP. BIOL. AND MED., 1942, **51**, 285; Zahl, Paul A., and Hutner, S. H., PROC. SOC. EXP. BIOL. AND MED., 1943, **52**, 116; Hutner, S. H., and Zahl, Paul A., PROC. SOC. EXP. BIOL. AND MED., 1943, **52**, 364.

* As many workers now agree that the toxic effects of extracts of gram-negative bacteria are