Further investigation is proceding to elucidate the mechanism of the R.I. phenomenon and to see whether it relates to neoplasia.

Summary. Reducing capacity of erythrocytes has been measured. Followng intravenous infusion of sodium thiosulfate, there occurs a significant diminution in this capacity. A reductivity index (R.I.) was established relating diminution in this capacity with amount of sodium thiosulfate infused. R. I. of erythrocytes of women is more than double that of normal men. Preliminary studies suggest that estrogen and androgen may influence R.I.

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Dietary Vitamin K Requirement of the Rat. (25526)

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Recent reports by Barnes and Fiala(1) and by Mameesh and Johnson(2) have shown that a dietary Vit. K deficiency can be produced in the non-coprophagic rat. Prior to the finding of the role of coprophagy in supplying intestinally synthesized Vit. K to the rat(1,2), it had not been possible to determine the quantitative Vit. K requirement of the rat for maintenance of a normal prothrombin level. The experiments reported herein were conducted to measure the Vit. K requirement of the growing, non-coprophagic, male rat.

Methods. Weanling male albino rats of the Sprague-Dawley strain were randomly divided into groups of 10 rats each. The animals were individually housed in wire-bottomed cages in a temperature-controlled laboratory. Food and water were given *ad libitum.* Daily feed records were kept, and the rats were weighed at weekly intervals. Coprophagy was prevented in all rats by the method of Barnes *et al.*(3). The composition of the basal diet is given in Table I. The diet was mixed every 2 weeks and stored in dark brown glass jars under refrigeration. To reduce bacterial growth, the daily feed refused was discarded and feed dishes and water bottles were washed every other day with hot water and a household detergent. Vit. K_1 (3 - phytyl - 2 - methyl - 1,4 - naphthoquinone, Mann Research Laboratories, Inc., New York) was dissolved in triolein and added to the diet at the time of mixing at predetermined levels. Evidences for inadequate Vit. K intake were: a) hemorrhagic deaths with one or more of the following symptoms ob-

TABLE I. Basal Diet.

Ingredient	g
Sucrose	66.9
Drackett-soy protein (Archer Daniels Midland Co., Cincinnati, O.)	20.0
DL-methionine (Dow Chemical Co., Midland, Mich.)	.5
Triolein	3.0
Vitaminized cerelose*	5.0
Salt mixture No. 446†	4.0
Methyl linoleate urea inclusion compound (25% methyl linoleate, Hormel Foundation, Austin, Minn.)	.6
α-Tocopherol acetate	.012
Vit. A (Nopcay, 250,000 USP units per g, Nopco Chemical Co., Harrison, N. J.)	.008
Vit. D_3 (Super Nopdex 15,000 I units/g, Nopco Chemical Co., Harrison, N. J.)	.013

* Mameesh and Johnson(2). † Spector(5).

TABLE II. Mean 4-Week Gains, Feed Intakes, Incidence of Lethal Hemorrhages and Plasma Prothrombin Times of Groups of 10 Non-Coprophagic Rats Receiving Various Levels of Vit. K_1.

	Vit. K_1 admin. (μg/g diet)	Avg 4-wk gain (g)	Avg 4-wk feed intake (g)	Hemorrhagic deaths	Avg prothrombin time (sec.)
Exp. I	0	103 ±15.6*	279 ±19.5*	5	58.6 ±8.8*
	.2	169 ± 5.3	386 ± 5.8	0	13.2 ± .2
	.4	165 ± 4.3	380 ± 8.6	0	13.8 ± .3
	.6	162 ± 3.3	368 ± 6.7	0	13.9 ± .2
	.8	154 ± 4.8	359 ± 7.8	0	14.2 ± .2
	1.0	159 ± 1.8	379 ± 4.5	0	14.0 ± .1
Exp. II	0	156	366	9	83
	.05	146 ± 4.1	359 ±10.4	2	25.0 ±2.3
	.10	159 ± 3.2	380 ± 6.1	0	14.8 ± .2
	.15	157 ± 3.6	372 ± 5.8	0	13.9 ± .2
	.20	157 ± 3.2	387 ± 6.3	0	13.9 ± .2

* Stand. error of mean.

served at autopsy: unclotted blood in thoracic cavity, epididymal bleeding, nasal bleeding and subcutaneous hemorrhages; b) prolonged whole plasma prothrombin time as determined by the method of Quick *et al.*(4). Two experiments were conducted; in the first, Vit. K_1 was given at 0, 0.2, 0.4, 0.6, 0.8 and 1.0 μg/g diet. Since all levels of Vit. K_1 used in this experiment were found to be adequate, a second experiment was conductd, using the levels of 0, 0.05, 0.10, 0.15 and 0.20 μg Vit. K_1/g diet.

Results. The results are given in Table II. Coprophagy-prevented rats consuming a diet deficient in Vit. K developed severe Vit. K deficiency. This observation confirms previous reports(1,2). The results in Table II indicate that the dietary Vit. K requirement of the non-coprophagic rat was between 0.1 and 0.15 μg Vit. K_1/g of diet. When the reciprocal of the prothrombin time was plotted against Vit. K level (Fig. 1), the 3 values below the requirement fitted in a straight rising line which intersected with the horizontal line of normal prothrombin at a point corresponding to approximately 0.1 μg K/g of diet. The diet provided a growth rate of approximately 5.7 g per rat per day over the 28-day period when Vit. K was supplied, while in the case of the deficient rats (Exp. I, Table II) feed intake and growth rate were depressed.

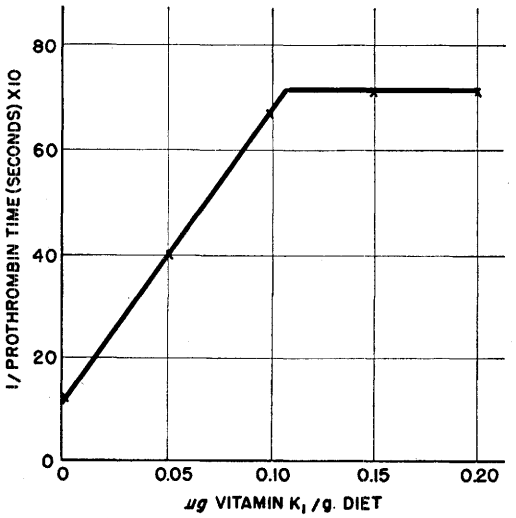

FIG. 1. 1/prothrombin time (sec.) × 10 at different levels of Vit. K_1 in the diet.

The linear relationship between reciprocal of the prothrombin time and Vit. K intake in coprophagy-prevented rats indicates that the amount of the vitamin contributed by intestinal microbial synthesis and direct absorption is insignificant. This confirms the earlier observation of Mameesh and Johnson(2) that coprophagy was the means by which Vit. K synthesized by the intestinal microflora becomes available to the rat. Growth and food intake data in Table II indicate that Vit. K was required for growth in coprophagy-prevented rats. The requirement for growth was found to be the same as that for maintainance of normal plasma prothrombin levels.

Summary. The dietary Vit. K requirement of the coprophagy-prevented growing male rat was found to be 0.1 μg/g of diet fed as Vit. K_1. This requirement satisfied the needs for maintainance of normal plasma prothrombin levels and for growth.

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Synergistic Action of Pneumococci and Leukocytes in Lowering Cerebrospinal Fluid Glucose.* (25527)

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The mechanism responsible for disappearance of cerebrospinal fluid sugar in bacterial meningitis has not been clearly elucidated and the relative importance of cells, bacteria, changes in meningeal permeability and accelerated utilization of glucose by neural tissue in genesis of hypoglycorrhachia remains poorly defined. It had been demonstrated that glycolysis occurs *in vitro* when CSF from dogs with non-bacterial meningitis is incubated at 37°C; the decrease in CSF sugar is proportional to number of leukocytes present (1,2). In contrast, pneumococci incubated in sterile CSF consume glucose at a very slow rate and large numbers of rapidly growing bacteria must be present before a fall in CSF sugar becomes evident. In the intact animal, however, sterile meningitis is not usually associated with a decrease in CSF glucose while bacterial infections in the meninges are almost invariably accompanied by hypoglycorrhachia. These paradoxical observations have led to the suggestion that neither cells nor bacteria can be held entirely accountable for the fall in CSF glucose(2). In our studies, CSF containing leukocytes was inoculated with pneumococci, and the mixture incubated at 37°C. Under these conditions consumption of glucose was greater than when organisms were incubated in cell-free CSF, or when CSF containing leukocytes obtained from dogs with non-bacterial meningitis was tested in absence of bacteria.

Methods. Aseptic meningitis was produced in mongrel dogs by injection of 6 ml of sterile, pyrogen-free, 0.85% NaCl into the cisterna magna. Four hours later CSF, which regularly contained 3000 to 8000 WBC/mm^3 (95-99% polymorphonuclear leukocytes), was removed by cisternal puncture. Fluids from several animals were pooled for each experiment and used immediately. Seed cultures of Type III pneumococci were stored at −70°C. For each experiment a fresh tube was thawed, added to 9 ml of tryptose phosphate broth and incubated at 37°C for 8 hours. Control tubes containing serum and broth but no bacteria were handled in similar fashion. In each experiment 0.1 ml of bacterial inoculum was added to a series of tubes containing 0.9 ml CSF obtained from animals with non-bacterial meningitis. The same number of bacteria added to 0.9 ml of sterile, cell-free CSF and fluid from animals with aseptic meningitis mixed with serum-broth inoculum not containing organisms, served as controls. One tube was immediately removed from each series for baseline bacterial and leukocyte counts and determination of glucose. The remainder were shaken in constant temperature bath at 37°C and were removed at intervals of 30, 60, 120, 180, and 240 minutes after beginning of experiment. The number of bacteria in each tube was determined by colony counts of serial 10-fold dilutions, a single colony being counted as one viable unit. Leu-

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