corpus luteum, number of corpora lutea per ovary or stage of pregnancy. An increase in lutein cell size from 30 to 90 days of pregnancy as reported by Foley and Reece(1) and Foley *et al*(17) was not observed. Hofliger(4) reported that the corpus luteum at 30 days of pregnancy contained equally-sized colloid inclusions and granulosa cells filled with fatty droplets.

Summary. Nulliparous heifers were injected with 1500 or 3000 i.u. of PMS on day 16 of estrus, bred to a fertile bull and autopsied 3, 30, 45, 60 and 90 days *post ovulation.* The corpora lutea were dissected and the diameter and weight of each were recorded. A portion of each corpus luteum was sectioned serially and examined microscopically. At 3 days *post ovulation* the weight of corpus luteum in ovaries possessing one ovulation, ranged from 453 to 820 mg with an average of 657 mg. There was wider variability in the weight of the corpus luteum in ovaries possessing multiple corpora lutea than in those with single corpus luteum. There was no significant correlation between corpus luteum weight and number of corpora lutea per ovary. The weight of corpus luteum was decreased when the number of corpora lutea increased over 20.

At 30 days of pregnancy corpus luteum weight in ovaries possessing a single corpus luteum, ranged from 4005 to 5240 mg with an average of 4752 mg. This weight was unaffected by stage of pregnancy from 30 to 90 days. There was a marked decrease in weight of the corpus luteum with the increase in number of corpora lutea with advancing pregnancy. There was a progressive increase in weight of luteal tissue per ovary with increase in number of corpora lutea. The histology of multiple corpora lutea was similar to that of single corpus, and the size of lutein cells was not correlated with the weight of individual corpora lutea, number of corpora lutea per ovary or stage of pregnancy.

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Effect of Cholestyramine, a Bile Acid Binding Polymer, on Vitamin K_1 Absorption in Dogs. (28845)

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It has been shown that cholestyramine, a quaternary ammonium anion exchange resin of high molecular weight, will bind bile acids *in vitro*(1). Several experimental and clinical reports indicate that this resin will lower plasma cholesterol levels in several animal species and in man(2-5). It has been suggested that the lowered plasma cholesterol

concentrations found in cockerels, dogs and man treated perorally with cholestyramine are due to bile acid sequestration in the intestinal lumen(6). Since bile acids are known to aid in absorption of fat-soluble vitamins, it seemed of interest to determine whether or not cholestyramine had any influence on the intestinal absorption of Vit. K_1.

Materials and methods. Oral absorption of Vit. K_1 can be readily quantitated by measuring the rate and efficiency of the orally administered vitamin in reversing Dicumarol [3,3′-methylenebis(4-hydroxycoumarin)] induced hypoprothrombinemia in dogs(7). In the present study 4 separate experiments were performed. In the first 3 experiments, 17-19 adult beagle dogs of both sexes were employed in each experiment. The animals weighed between 9 and 11 kg and were housed in individual cages in an air-conditioned room and maintained on a nutritionally adequate stock diet. The commercial stock diet contained 27% protein, 9% fat, 4.5% fiber, 10% ash, and 12% moisture. This diet was supplemented with canned horse meat. The animals were fed in the late afternoon and were allowed to fast overnight. Each study was initiated in the morning, therefore all studies were performed in fasted animals. Water was given *ad libitum*. Prothrombin time values were determined on citrated plasma by the one-stage method, essentially according to Quick(8). The dogs were rendered hypoprothrombinemic by oral administration of Dicumarol. The anticoagulant was administered in gelatin capsules on each of 2 consecutive days at a dose level of 3 mg/kg. Two days after the second dose of Dicumarol, when prothrombin time values reached a peak but plateau level, in each of the first 3 experiments the dogs were divided into 3 groups and given Vit. K_1,* Vit. K_1 and cholestyramine or no additional treatment respectively. More specifically, in the first experiment (Fig. 1), one group of 6 fasted hypoprothrombinemic dogs was given a single oral dose of 10 mg/kg of Vit. K_1 administered in gelatin capsules. The second group, which received both the vitamin and the resin, was fed the first dose of resin (67 mg/kg), suspended in water, one-half hour before oral administration of 10 mg/kg of Vit. K_1. The same amount of cholestyramine was given 3 and 5½ hours after administration of Vit. K_1 thereby making the total dose of the resin 200 mg/kg. Under these conditions the resin had an optimal opportunity to bind bile acids before and during the passages of Vit. K_1 along the intestinal tract. The third group of 6 hypoprothrombinemic dogs received no additional treatment and served as controls. Prothrombin times were determined just prior to, and 1, 3, 6, and 24 hours after Vit. K_1 treatment. After a suitable time interval of at least one week, the second and third experiments were performed in the same dogs under identical conditions except that the total dose of cholestyramine was increased to 1.0 and 3.0 g/kg respectively as shown in Fig. 2 and 3. Finally, in the fourth experiment 3.0 g/kg of the resin was given 17 hours before administration of the vitamin. A dose of 1.0 g/kg of cholestyramine was fed at 2 hourly intervals for 3 doses over a 6-hour period. The following morning 10 mg/kg of Vit. K_1 was given orally to the dogs and prothrombin time determined at the time intervals described in the foregoing experiments.

Results. In agreement with data published previously(7), it is evident that 3 mg/kg of Dicumarol given alone to dogs induced a striking hypoprothrombinemia which persisted uniformly throughout the 4-day observation period (Fig. 1-4). Moreover, a single oral dose of 10 mg/kg of Vit. K_1 rapidly reversed the hypoprothrombinemia and in most instances prothrombin time returned to a normal range within 3 to 6 hours following Vit. K_1 administration. Cholestyramine administered in a total dose of 200 mg/kg (67 mg/kg × 3) had no effect on absorption of Vit. K_1 (Fig. 1). Prothrombin time value rapidly returned to a normal range 3 hours after the vitamin was given. Larger doses of 1.0 g/kg of resin decreased and delayed absorption of the vitamin somewhat, as evidenced by the

* Mephyton is the registered trademark of Merck & Co., Inc., for (natural) Vit. K_1.

EFFECT OF CHOLESTYRAMINE ON VITAMIN K_1 ABSORPTION IN DOGS

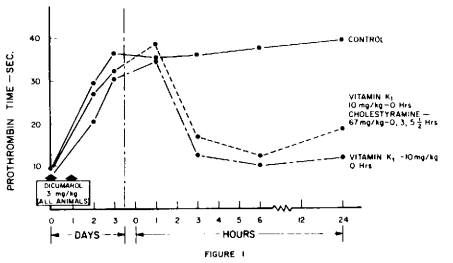

FIGURE 1

EFFECT OF CHOLESTYRAMINE ON ABSORPTION OF VITAMIN K_1 IN DOGS

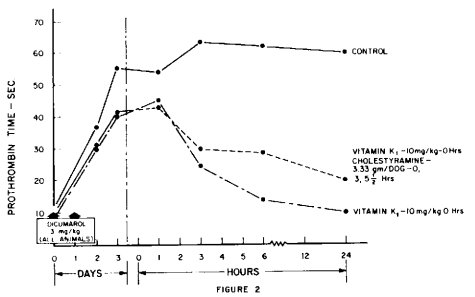

FIGURE 2

EFFECT OF CHOLESTYRAMINE ON ABSORPTION OF VITAMIN K_1 IN DOGS

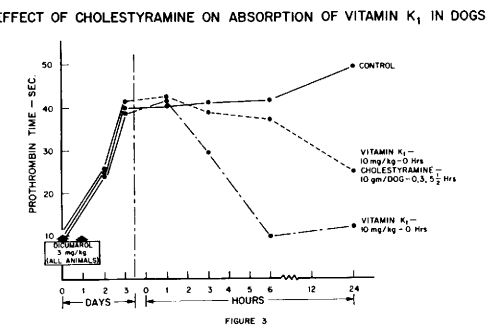

FIGURE 3

EFFECT OF CHOLESTYRAMINE ON ABSORPTION OF VITAMIN K_1 IN DOGS

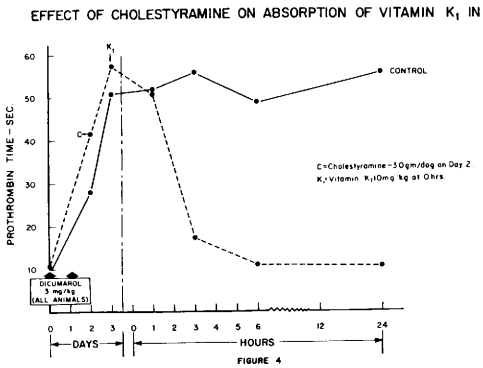

FIGURE 4

elevated prothrombin time values 3 to 6 hours following administration of Vit. K_1 (Fig. 2). However, within 24 hours prothrombin time returned to a normal range indicating that Vit. K_1 was ultimately absorbed during this time interval (Fig. 2). When a massive dose of 3.0 g/kg was given orally simultaneously with the vitamin, there was a definite decrease in absorption of Vit. K_1 as evidenced by the prolonged prothrombin time in all dogs treated with this dose of cholestyramine (Fig. 3). However, even this large amount of resin had no effect on Vit. K_1 absorption, if the resin was given well before administration of the Vit. K_1, as illutrated in the data from the fourth experiment (Fig. 4).

Discussion. It is apparent from the data obtained in these studies that large doses of cholestyramine, administered essentially simultaneously with Vit. K_1, will interfere with the absorption of this vitamin. However, when the resin is administered well before the vitamin or given in amounts more in line with the practical clinical utility of the resin, the latter had no apparent effect on Vit. K_1 absorption. Since the recommended dose of cholestyramine for man for treatment of hypercholesterolemia is in a range of 200 mg/kg(6), it would appear unlikely that this resin will induce a deficiency of this vitamin in man. Presumably similar conclusions may be drawn for the other fat-soluble vitamins although Vit. A, D or E were not studied in this investigation.

It should be noted that the first 3 experiments were designed to afford optimal conditions for cholestyramine to inhibit the absorption of Vit. K_1. The resin was given 30 minutes before oral administration of Vit. K_1 and 2 additional doses of cholestyramine were fed 3 and 5½ hours after administering the vitamin. Under these experimental conditions the resin had a maximal opportunity to sequester the bile acids present in the intestinal lumen before, or secreted into the lumen during the time Vit. K_1 entered the small intestine. Under practical clinical conditions of use, where the resin is given during the waking hours, cholestyramine would not be present in the intestinal tract constantly and therefore, at times the bile acids

would be freely available to aid in the absorption of fat-soluble vitamins. This conclusion is supported by the data obtained in the fourth experiment of this study. These are added reasons for concluding that it is highly unlikely that cholestyramine will induce a Vit. K deficiency in man. From the experimental point of view, however, it is evident that large doses of cholestyramine may serve as a useful tool in the study of bile acid function and their effect on the absorption of fat-soluble vitamins.

Summary. Studies were undertaken to determine whether or not cholestyramine, a bile acid binding resin, would interfere with the intestinal absorption of fat-soluble vitamins. The studies were designed to give the resin an optimal opportunity to interfere with Vit. K_1 absorption. Administration of cholestyramine in a dose range recommended for human use had no effect on Vit. K_1 absorption. Large doses of the resin, administered before and shortly after feeding the vitamin, did delay and decrease the absorption of this vitamin. However, very large doses of cholestyramine given 17 hours before feeding Vit. K_1 had no affect on the availability of the vitamin. These studies suggest that cholestyramine would not be expected to interfere with the absorption of Vit. K when the resin is used under practical clinical conditions in man. However, the resin may be a useful experimental tool to investigate the role of bile acids in fat absorption.

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Absence of Cross Reactivity Between Heterophile and Dye Test Antibodies.* (28846)

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One of the recognized clinical syndromes resulting from acute infection with *Toxoplasma gondii* closely resembles infectious mononucleosis(1,2,3). Although the laboratory findings in patients with these infectious diseases are similar, the presence of a positive heterophile antibody test titer, which is considered diagnostic of infectious mononucleosis, is seldom found in cases of acute toxoplasmosis(1). Of interest in this regard is a report of a case in which Toxoplasma organisms were isolated at a time when the serum was positive in both the dye and heterophile tests(4).

During recent studies performed in an attempt to determine the incidence of acute Toxoplasma infection among students with symptoms and signs suggestive of infectious mononucleosis, a number of sera were noted to be positive both in the dye test and het-

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