and massive vacuolation in the cells of the proximal convoluted tubule. Oliguria proceeds rapidly to anuria with destruction of larger blood vessels and hemorrhage. Hume *et al*(9) showed that unless a major vascular insult such as thrombosis or infarction has occurred, tubular rather than glomerular lesions are present early in a rejection crisis.

Lysozymuria was noted in the rejection crises observed in this study. In view of the reported relationship between lysozymuria and renal tubular damage(6) it would appear that this investigation lends further support to the ascribed importance of tubular lesions in the rejection phenomenon.

Summary. Increased lysozyme activity in urine has been shown to be a reliable sign of renal homograft rejection. The ease and rapidity (20 minutes) of the measurement should make this assay a most valuable adjunct in the postoperative management of patients with kidney homotransplants.

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Adrenal Cortical Function in Experimental Alcoholism in Dogs.* (30642)

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Although evidence is available to suggest that ethanol administration stimulates adrenocortical secretion in several animal species (1,2,3), no work involving direct measurement of plasma steroids appears to have been done in dogs under the acute influence of this alcohol. Since some features of the disposition of both ethanol and the corticosteroids seem to occur in common in man and the dog, a series of studies on the interrelationships between experimental alcoholism and adrenal function was begun in dogs. The present communication is concerned with the effect of acute ethanol intoxication on steroid secretion as indicated by 17-hydroxycorticosteroid levels in the peripheral plasma of these animals.

Methods. Mongrel dogs of both sexes were used repeatedly and under a variety of conditions so that control and experimental data could be obtained from the same animals. Blood samples were obtained by venipuncture of the intact jugular vein before and 1, 2, 4, and 6 hours after one of the following intravenous infusions or injections: saline, 10 ml/kg; corticotropin (Wilson's ACTH solution), 1.2 units/kg; ethanol (redistilled absolute), 1.0, 2.0 or 3.0 g/kg (diluted to 20%, w/v, with saline); or hydrocortisone (free alcohol), 1 mg/kg. Saline and ethanol solutions were infused into a leg vein during the course of 10 to 20 minutes. ACTH solution was diluted with saline to a volume of 5 ml and administered by rapid injection. Hydrocortisone was dissolved in 5 ml of 5% ethanol in saline and injected in a similar manner.

Plasma was analyzed for 17-hydroxycorticoid content by the water-benzene partition

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procedure of Eik-Nes(4). Hydrocortisone, which is considered to be the principal adrenal steroid in the dog(5,6,7), was employed as the reference standard and plasma values are expressed as free 17-hydroxycorticosteroid concentration. Ethanol determinations were done by the alcohol dehydrogenase method of Brink *et al*(8).

Statistical significance of the data was determined by analysis of variance. Variability of the results is expressed as standard errors of the means. Regression lines for hydrocortisone disposal rates were calculated by the method of least squares.

Results. 17-Hydroxycorticosteroid (17-OHCS) concentration in control plasma samples. Control blood samples obtained on various occasions from each dog were drawn under similar conditions during the period of 8:30 to 10:00 A.M. Mean steroid values, comprised of 5 to 8 separate determinations, of the individual dogs ranged from 4.1 ± 0.7 to 5.7 ± 1.3 μg/100 ml. The differences observed among these mean values are not significant. The composite mean value for 57 determinations in the entire group of dogs was 4.6 ± 0.86 μg/100 ml, which is comparable to 17-OHCS control concentrations reported for dogs by other workers(9,10,11).

Changes in 17-OHCS plasma levels induced by infusion and injection procedures. The effects on plasma 17-OHCS concentration of saline, ACTH and ethanol administration are shown in Fig. 1. Calculation of percentage change in steroid content is based on collective differences between control and experimental values of individual experiments rather than on differences between group means.

Results of the saline experiments indicate that handling of the animals, infusion of a volume of saline equivalent to that needed to provide 2 g/kg of ethanol, and serial blood sampling (5 samples of about 30 ml each) during a period of 6½ hours did not induce significant changes in the plasma steroid level.

Typical sensitivity to ACTH was demonstrated by the marked elevation in 17-OHCS plasma concentration which characteristically reached a peak at the 2-hour interval. The maximum change at this time corresponds to an increase of 435%. The differential percentage increase between saline and ACTH is highly significant ($P<.001$) at every time

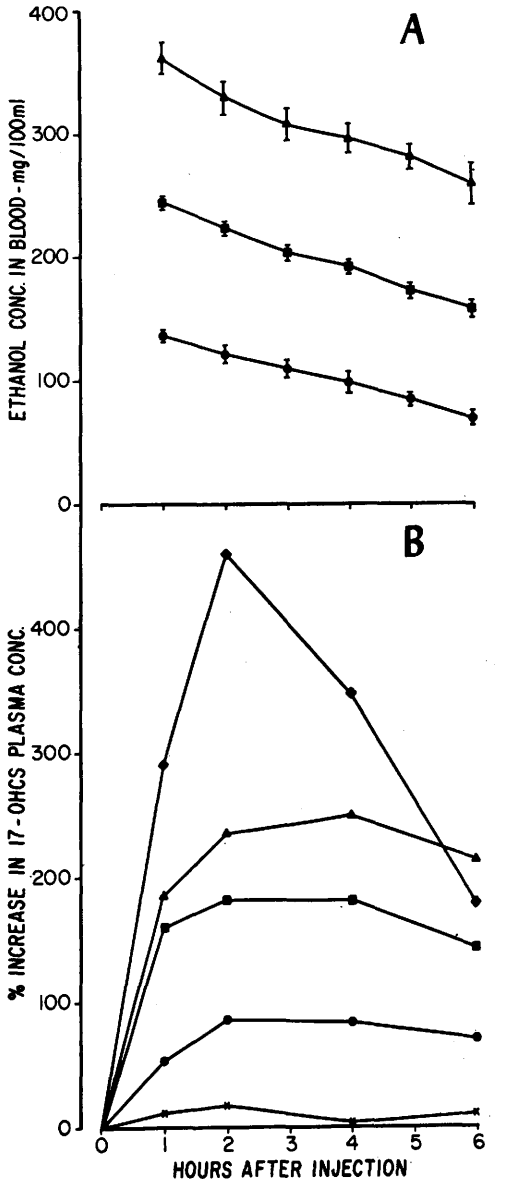

FIG. 1. Blood-alcohol curves and changes in plasma concentration of 17-OHCS following the injection of ethanol. ×—×, 10 ml/kg saline; ●—●, 1 g/kg ethanol; ■—■, 2 g/kg ethanol; ▲—▲, 3 g/kg ethanol; ◆—◆, 1.2 units/kg ACTH. All injections were intravenous. Part A: mean values ± standard errors of means. Part B: by analysis of variance, all experimental values are significantly different ($P<.02$) from the corresponding saline means except the 1 g ethanol value at 1 hr and 6 hr ($P>.05$). See text for other P values. Each curve represents 5 to 8 animals.

TABLE I. Mean Plasma Levels of 17-OHCS in Dogs After Injection of 1 mg/kg of Hydrocortisone.

Treatment	No. of exp	Control value	μg/100 ml—means ± S.E.* Hr after hydrocortisone 1	2	4	6
Saline 10 ml/kg	9	3.5 ± .8	35.2 ± 4.4	17.3 ± 1.5	6.4 ± 1.7	3.9 ± 1.0
Ethanol 2 g/kg	12	4.4 ± .6	42.1 ± 3.9	23.2 ± 2.0	9.2 ± 1.5	6.6 ± 1.2

* Standard error of mean.

interval throughout the period of observation.

Ethanol administration, at each dosage level, was followed by a rise in plasma steroid concentration which generally persisted throughout the experiment. The effect of 1 g/kg evoked an incipient increase which became significant at 2 ($P<.025$) and 4 ($P<.05$) hours but this change had fallen below the significant level at 6 hours. There was a much more pronounced and uniform response to the 2 g/kg dose which induced highly significant ($P<.001$) rises above the saline levels at every time interval. Even though the mean responses to 3 g/kg were the highest changes produced by ethanol, variations in these effects were also the largest. Because of these wide variations the mean differential values caused by this dose are significant at lower levels ($P<.005$-$.01$) than those after 2 g/kg. Thus, while a distinct difference in steroid levels was apparent between the 1 and 2 g/kg doses, the significance of relative changes produced by 2 and 3 g/kg doses was above the 5% statistical level at every point of comparison. Although the mean results appear to reflect some type of dose-response effect, this relationship was distorted by the largest dose of ethanol.

Following the injection of 1 g/kg of ethanol depression of the central nervous system was irregularly evident primarily in terms of slight ataxia for 10 to 30 minutes. The effects of 2 g/kg were characterized by marked ataxia or inability to stand and pronounced reduction in response to ordinary stimuli for 30 minutes or more after ethanol administration. The largest dose of ethanol rendered the animals unable to walk or stand for 30 to 60 minutes and produced considerable ataxia for 2 hours or more. Progressive changes in the blood-alcohol concentrations during these experiments are shown in Fig. 1.

Plasma levels of 17-OHCS after injection of hydrocortisone into normal and ethanol-intoxicated animals. Since elevated plasma steroid levels in the peripheral circulation may reflect not only adrenocortical secretion but also the physiological efficiency with which free steroids are metabolized, conjugated and excreted, it was desirable to determine the influence of ethanol on the steroid disposal rate. Table I and Fig. 2 show results of experiments in which the plasma level of injected hydrocortisone was determined after the infusion of saline or

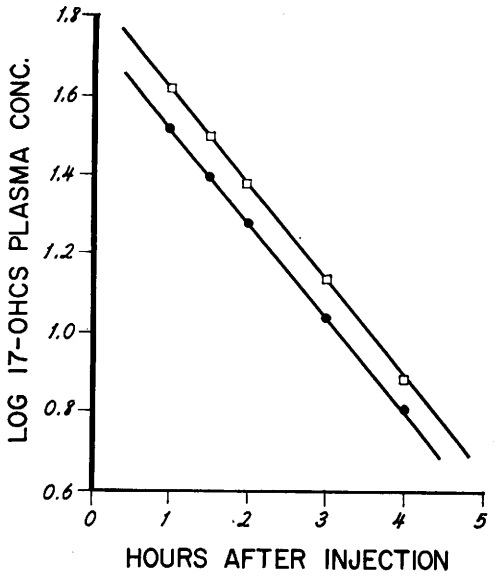

FIG. 2. Disposal rates of exogenous hydrocortisone after intravenous injection of 1 mg/kg. Regression lines calculated by the method of least squares. ●—●, normal dogs (9 experiments but only 2 determinations at 1½ and 3 hr); □—□, ethanol-treated dogs, 2 g/kg of ethanol (12 experiments but only 3 determinations at 1½ and 3 hr).

ethanol in the same animals referred to above. Presumably the higher mean values observed in the ethanol experiments are due to secretion of endogenous steroid induced by alcohol. Regression lines were calculated by the method of least squares from steroid values obtained as hydrocortisone progressively disappeared from the blood stream. As indicated by the slopes of the curves in Fig. 2, there is no difference between the disposal rates in normal and ethanol-treated animals.

Discussion. Since ethanol intoxication did not alter the normal disposal rate of exogenous hydrocortisone, the elevated plasma levels of endogenous 17-OHCS observed in the ethanol experiments indicate that alcohol evoked a corticosteroid response in the dog. This observation is consistent with previous findings of ethanol-induced pituitary-adrenal stimulation in rats(1,3,12). The high degree of variability observed after 3 g/kg of ethanol suggests that a considerable difference existed among the animals in their sensitivity to various doses of ethanol or even that the large dose may have partially suppressed rather than stimulated corticosteroid secretion. Despite this variability, comparison of the saline and ethanol effects shows that alcohol clearly produced adrenocortical stimulation at lower doses. Fluctuation in steroid levels could not be correlated with the behavioral reaction (*e.g.*, degree of activity in the cage, barking, or sleep pattern between blood samplings) of the animals to alcohol nor explained by diurnal variation since very little change occurred in saline experiments conducted over essentially the same portion of day (8:30 a.m.-4:30 p.m.) as the other experiments. Other workers also have observed little diurnal fluctuation in 17-OHCS levels in the dog(11,13,14).

The observations of this study suggest and are consistent with the hypothesis that the adrenocortical response evoked in dogs by ethanol is mediated by secretion of ACTH. These results do not permit, of course, the exclusion of a possible direct action on the adrenal cortex. However, this mechanism is unlikely in view of conclusive evidence that ethanol does not lower adrenal ascorbic acid concentration(1) nor elicit corticosterone secretion (Ellis, in press) in hypophysectomized rats. It also seems improbable that vasopressin, shown by Andersen and Egdahl (15) to produce an ACTH-like activity in dogs, could have been responsible for elevating 17-OHCS concentrations since available evidence indicates that ethanol suppresses secretion of the antidiuretic hormone(16).

Data presented in this paper appear to demonstrate that ethanol, like reserpine(17), morphine(18), and chlorpromazine(19), is capable of stimulating adrenal cortical secretion after acute administration to dogs. These results do not necessarily mean that the effect recurs indefinitely upon repeated exposure to ethanol over an extended period of time. Studies in rats have shown that frequent administration of some drugs, which initially stimulate adrenal cortical secretion, leads to an apparent adaptation in the secretory mechanism with the consequent loss of the corticosteroid response(20). It is anticipated that studies in progress will provide additional information about the nature of this response to ethanol in dogs maintained under experimental conditions of prolonged chronic alcohol intoxication.

Summary. The effect of acute ethanol administration on corticosteroid secretion has been studied in dogs by the direct measurement of 17-hydroxycorticosteroid concentration in peripheral plasma. Procedures associated with the basic experimental design such as repeated handling of the animals, intravenous infusion of a sizeable volume of fluid and serial blood sampling over a 6½-hour span of time had no appreciable effect on steroid plasma levels. Intravenous infusion of ethanol in different doses resulted in a significant elevation of plasma 17-hydroxycorticosteroid concentration but because of a high degree of variability in the results, no definite dose-response relationship could be established. Ethanol intoxication did not alter the normal disposal rate of exogenous hydrocortisone after the intravenous administration of this steroid. These observations suggest that the ethanol-induced corticosteroid response in the dog may be mediated by ACTH secretion.

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Lowering of Human Serum Cholesterol by an Oral Hydrophilic Colloid.* (30643)

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The influence of the various forms of dietary bulk on serum cholesterol levels has not been extensively studied. Keys(1) found that addition of pectin but not cellulose to the diets of middle-aged male patients produced a small (5%) decrease in serum cholesterol values. Studies on pigs by Cooper(2) showed that addition of fibrous but not powdered cellulose to the diet lowered serum cholesterol and suggested that the ability of the fibrous cellulose to hold water may have been responsible for the difference. The present paper describes experiments on normal young male medical students which show that on mixed or egg-supplemented diets the addition of a hydrophilic colloid to the diet lowers the serum cholesterol to a statistically significant extent.

Materials and methods. A preparation of hydrophilic colloid derived from blond psyllium seed (*Plantaga ovata*-Forsk) available commercially as Metamucil,† was used as the bulk-increasing supplement and as supplied was mixed with an equal weight of dextrose. During the periods of mucilloid supplementation in all the experiments, 6.4 g of Metamucil were taken 3 times daily with meals, making a total of 9.6 g of hydrophilic colloid ingested each day. All serum cholesterol values were obtained by the method of Zlatkis (3) and carried out in duplicate. The experimental subjects were 14 normal male medical students, from 21-25 years of age, and one male faculty member 33 years of age. All subjects maintained their customary activi-

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