pletely, resistant to the action of the toxin and in its presence showed little or no difference in permeability to fluids from normal intestine in the absence of toxin. Data from typical experiments are given in graphical form in Fig. 1.

These phenomena have been studied in detail and the investigation extended considerably further than indicated here; these studies will be reported fully elsewhere. It may be pointed out, however, that these results are consistent with and complementary to those reported in a preceding paper[8] and it is apparent that active immunity to Asiatic cholera in the experimental animal, and presumably also in man, includes antitoxic as well as antibacterial immunity. The significance of the former in enteric infection with the vibrios is clearly indicated though not as yet quantitatively evaluated. If it be assumed that the resistance of the immune intestine to the action of the endotoxin has an antigen-antibody basis, and if this is not assumed an entirely new type of immunity must be postulated, it is clear that the functional antibody cannot be circulating antibody in the isolated strip of intestine and may, therefore, be intracellular antibody, presumably in the macrophages of the tissues of the intestine.

[8] Burrows, W., Mather, A. N., Wagner, S. M., and McGann, V. G., PROC. SOC. EXP. BIOL. AND MED., 1944, **57**, 308.

14794

Influence of Citric Acid on Blood Pyruvate Levels in Man.

AUSTIN HENSCHEL, OLAF MICKELSEN, AND ANCEL KEYS.
From the Laboratory of Physiological Hygiene, University of Minnesota, Minneapolis.

Von Euler and Högberg[1] reported that the administration of citric acid produces a delayed decrease in the concentration of pyruvic acid in the blood. The principal evidence was from a small series of experiments with rats receiving oral or subcutaneous dosages of sodium citrate. Emphasis, however, was placed on a single experiment with a normal man whose blood pyruvate was lowered to 22% of the original level 12 to 15 hours after the oral ingestion of 2 g of citric acid as the sodium salt. The blood pyruvate change in this man was stated to be accompanied by indications of a profound general disturbance including sweating, shivering, and fever.

Apart from the suggestion of important theoretical relations this report is of interest because of the growing use of blood pyruvate measurements in the study of thiamine deficiency and the fact that the citric acid dosage employed is scarcely heroic. Two grams of citric acid would be supplied by one large lemon or by 200 cc of orange juice.[2]

In this Laboratory this question was studied in 5 experiments on 3 normal young men who had been maintained on a carefully standardized dietary for several months. In each experiment a blood sample was drawn before breakfast at 8:00 a.m. and 3 g of citric acid as Na citrate were administered orally in 200 cc of orange juice on the same day at 6.00 p.m. The final blood sample was taken before breakfast on the following morning at 8:00 a.m., that is, 14 hours after the citric acid administration. The blood samples were drawn from veins in the antecubital fossa. Pyruvate was estimated in duplicate by the method of Friedemann and Haugen.[3]

The results are summarized in Table I. It is clear that the citric acid had no effect of any kind on the pyruvate level at the time—14 hours—when the maximum change was

[1] Von Euler, H., and Högberg, B., *Z. Physiol. Chem.*, 1940, **265**, 244.

[2] Chatfield, C., and Adams, G., *U. S. Dept. Agric. Circular* No. 549, 1940.

[3] Friedemann, T. E., and Haugen, G. E., *J. Biol. Chem.*, 1943, **147**, 415.

TABLE I.
Blood Pyruvate Values in mg per 100 cc of Whole Blood Before and 14 Hours After the Ingestion of 3 g of Citric Acid.

Time	Subject 1			Subject 2	Subject 3
1. Before citric acid	0.56	0.96	1.06	0.58	0.73
2. After " "	0.58	0.70	1.03	0.72	0.67
Δ, 1-2	—0.02	+0.26	+0.03	—0.14	+0.06

expected from the report of von Euler and Högberg. Clinical effects and subjective complaints were likewise entirely absent in all of the present experiments. Blood lactate, which was also measured in 2 experiments, did not change. We are unable to offer any explanation for the divergence of these results from those previously reported but we may note that the details in the von Euler and Högberg report are scanty.

14795 P

Evacuation of Gall Bladder in Female Patients with Pernicious Anemia.

EDWARD A. BOYDEN AND JOHN A. LAYNE.
From the Departments of Anatomy and Internal Medicine, University of Minnesota.

In an earlier study[1] it was observed that in male patients with pernicious anemia the rate of evacuation of the gall bladder was not significantly retarded by the disease, but that in a surprisingly large proportion of the cases studied roentgenologically, the gall bladder could not be visualized, notwithstanding the use of the intravenous method and the absence of any gall bladder history. Accordingly it seemed desirable to examine a comparable number of female patients.

The present report is based upon cholecystographic studies of 48 consecutive, unselected patients with pernicious anemia (23 males and 25 females). In this group, 42% could not be visualized (35% of the males and 48% of the females). Also, a review of the 31,311 necropsies recorded by the Department of Pathology for the 15-year period between 1926 and 1940, showed that of the 105 individuals having pernicious anemia, 32.4% had had either cholecystitis or cholelithiasis (or both) or had had the gall bladder removed. Furthermore the incidence of gall bladder disease increased progressively with age to the 8th decade—whereas Blalock[2] has shown that the greatest percentage of cases of biliary disease occurs in the fifth decade. These figures suggest that pernicious anemia increases the incidence of gall bladder disease and may have an etiological relation to it.

Evacuation of the gall bladder in 12 female patients (average age, 58 years) showed marked retardation over the group of female controls[3] (average age, 65 years); for in the first 40 minutes after the standard meal, the pernicious anemia group had evacuated only 71.5% of the contents of the gall bladder as against 84% by the controls. The difference is statistically significant, being nearly 3 times the standard error.

It would thus appear that patients with pernicious anemia are more likely to have developed cholecystic disease than other individuals of comparable age. When one asks what changes occur in this disease which might affect the gall bladder, the factor of increased excretion of fecal urobilinogen imme-

[1] Layne, J. A., and Boyden, E. A., PROC. SOC. EXP. BIOL. AND MED., 1940, **43**, 534.

[2] Blalock, Alfred, *Bull. Johns Hopkins Hosp.*, 1924, **35**, 391.

[3] Boyden, E. A., and Grantham, S. A., Jr., *Surg., Gyn., and Obstet.*, 1936, **62**, 34.