20-day formolized filtrate was mixed with either its homologous toxic filtrate or with the 20-day filtrate of the atoxic variant. Likewise, the antiserum prepared against the 20-day filtrate of the atoxic variant produced a wide zone of precipitation in the presence of both the homologous filtrate and the 20-day filtrate of the toxic strain.

The results of these experiments indicate that (1) the filtrates of the young cultures of the toxic variant are relatively free from bacterial protein and hence their antisera contain antitoxin and no detectable antibacterial antibody; (2) that the filtrates of the old cultures of the toxic variant contain bacterial protein in addition to the toxin, thereby stimulating the production of both antibacterial antibodies and antitoxins; (3) that the usefulness of the Ramon test in the *in vitro* standardization of staphylococcal toxin and antitoxin is limited by the fact that the flocculating power is not strictly parallel to toxicity but depends upon the presence of bacterial proteins in the antigen and antibacterial antibodies in the antitoxic serum.

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Auto-Injection of the Biliary Passages from the Gall Bladder in Rabbit.

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Contraction of the rabbit gall bladder may drive its contents back into the biliary passages if the bile papilla is tonically contracted or if the choledochus is clamped, and dyes previously injected into the gall bladder may then be demonstrated in the liver and arterial blood.

Experimental Procedure. 300 mg sodium barbital subcutaneously per kg animal; clamp choledochus; expose gall bladder and withdraw 1 cc of bile; through the same hypo needle kept *in situ* inject 0.5 cc of 5% sodium fluoresceinate solution; clamp the puncture in the gall bladder by means of a small narrow-jawed bulldog clamp; to contract the gall bladder, 0.5 cc per kg of a crude secretin preparation containing histamin was injected intravenously.

Within one or 2 minutes after the secretin injection the gall bladder usually becomes tense, decreases in volume and the choledochus and hepatic ducts appear as bulging, rounded, green cords; the choledochus may be so distended that it is palpable. In 15 minutes ap-

proximately some liver lobes may show an irregular yellowish-green mottling on their surfaces. This mottling is generally most marked in the left anterior and posterior lobes and is least in the caudate lobe. At no time was any dye visible in the lymphatics of the gall bladder or the liver hilus. If the animal is now bled to death from the abdominal aorta, the dye may be demonstrated in the serum and the relatively bloodless liver now reveals the dye more clearly than before. If the liver substance is scraped away, scraping from the free border of a lobe toward the hilus, yellow hepatic ducts can be easily demonstrated. These yellowish ducts may often extend practically to the border of the lobe; not all hepatic ducts in any one lobe exhibit the dye, however, and their distribution is irregular.

On the basis of these results it seems probable to us that the chain of conditions described above plays a rôle in producing or maintaining a cholangitis or hepatitis when it happens to a subject with an infected gall bladder. This supposition is strengthened by several observations in rabbit where contraction of the gall bladder occurred at a time when the bile papilla was tonically contracted, so that clamping of the choledochus was not necessary; in these instances the choledochus and bile ducts became markedly distended with the dye-bile solution.

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Bacterial Synthesis of Cocarboxylase.*

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The authors[1] have submitted evidence indicating that *Propionibacterium pentosaceum* can phosphorylate vitamin B_1 to its pyrophosphoric acid ester (cocarboxylase). This synthesis was carried out in the presence of liver-extract and hexosediphosphate. More recently, Ochoa and Peters[2] have shown that vitamin B_1, or suitable pyrimidine fractions, can in the presence of cocarboxylase, catalyze

* Appreciation is expressed to Merck and Company for synthetic crystalline cocarboxylase, to Winthrop Chemical Company for synthetic vitamin B_1, and to Anheuser-Busch for the bottom-yeast.

1 Silverman, M., and Werkman, C. H., *Enzymologia*, 1939, **5**, 385.

2 Ochoa, S., and Peters, R. A., *Biochem. J.*, 1938, **32**, 1501.