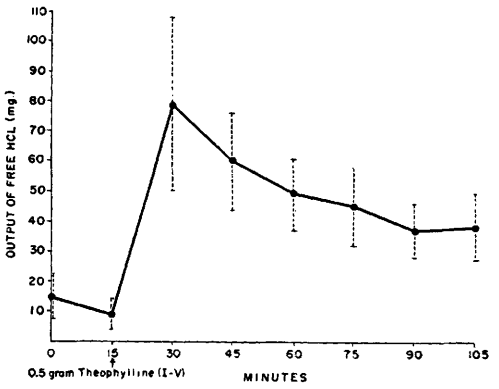

FIG. 1.
Stimulation of free HCl output by intravenous administration of theophylline ethylenediamine. Each dot represents the mean value of determinations on 10 subjects. The vertical bars represent the standard errors of the respective means.

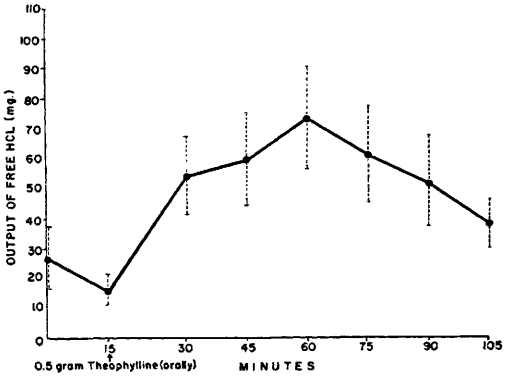

FIG. 2.
Stimulation of free HCl output by oral administration of theophylline ethylenediamine. See legend of Fig. 1.

similar to caffeine in its effect on gastric secretion in man.

In the course of the study 3 patients received a tablet form of theophylline with variable response, and 4 patients received theobromine orally with no gastric stimulation.

No decision can be made as to whether the stimulation produced by orally administered aminophylline is due to direct local action or to an action after absorption.

Summary. Theophylline stimulates the secretion of acid gastric juice in man on oral or intravenous administration.

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17183. Bacterial Conversion of Dextrin into a Polysaccharide with the Serological Properties of Dextran.

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The purpose of the present paper is to report the capacity of certain acetic acid bacteria, and of cell-free solutions of enzymes obtained from them, to synthesize from dextrin material with serological properties like those which we have earlier described for dextran.[1] Hitherto, the only enzymatic pathway known for the synthesis of dextran was that from sucrose.[2,3] We know of no report of interconversion of material of the starch-glycogen class and dextran, although both possess related chemical structures and can indeed be synthesized from the same substrate (sucrose) by appropriate bacterial enzyme systems.[2-5] Ultimate proof of the biological synthesis of dextran from dextrin must of course rest upon chemical studies, which are now in progress. However, the unequivocal nature of the present serological data leaves little doubt that such a conversion does in fact occur.

British brewing bacteriologists[6-11] have for

1 Sugg, J. Y., and Hehre, E. J., *J. Immunol.*, 1942, **43**, 119.

2 Hehre, E. J., and Sugg, J. Y., *J. Exp. Med.*, 1942, **75**, 339.

3 Hehre, E. J., *J. Biol. Chem.*, 1946, **163**, 221.

4 Hehre, E. J., and Hamilton, D. M., *J. Bact.*, 1948, **55**, 197.

5 Hehre, E. J., *J. Biol. Chem.*, 1949, **177**, 267.

some years recognized that certain varieties of acetic acid bacteria, now known as *Acetobacter viscosum* and *Acetobacter capsulatum*, often are associated with the type of spoilage of beer known as "ropiness." Recently Shimwell[12,13] has shown that the production of ropiness by these bacteria depends upon their capacity to form slime from dextrin, a natural constituent of beer. This author found that cultures of *A. viscosum* and of *A. capsulatum* became grossly viscous in dextrin-rich beer or in a medium of yeast extract containing 4 per cent dextrin, but not in beer devoid of dextrin or in yeast extract media in which the dextrin was omitted or replaced by glucose, fructose, maltose, or sucrose. It is of historical interest that Henneberg[14] fifty years previously had reported slime formation in dextrin-rich media (also often in beer) as a differential feature of the related *Bacterium* (*Acetobacter*) *industrium*.

Little has been recorded of the nature of the slimy material produced by the acetic acid bacteria associated with ropy beer. Hampshire[8] isolated a mucilaginous material from beer made ropy by inoculation with *Acetobacter* R, a variety now said to be closely related to *A. viscosum*.[13] This material was ash free, contained approximately 1% nitrogen, and on acid hydrolysis yielded 89.5% reducing sugar, calculated as glucose. Polarimetric readings were stated not to agree with this figure, but nevertheless the author concluded that the material was "of the nature of a dextran", *i.e.*, a polyglucoside. In the case of the slimy materials known to have been produced from dextrin by *A. viscosum* and *A. capsulatum* we were unable to find any published information apart from the statement of Shimwell[12] that these must be of carbohydrate nature because of their origin.

The present study was made with two strains of *Acetobacter*, procured from the National Collection of Type Cultures, capable of producing a gelatinous type of growth in media containing dextrin: *i.e.*, *A. viscosum* 7216, isolated from a ropy top fermentation of beer by Tosic, and *A. capsulatum* 4943, isolated from ropy beer by Shimwell.[9,12] The capacity of these strains to produce material serologically like dextran was established by experiments made in comparison with a strain (B) of *Leuconostoc mesenteroides* originally used in the establishment of the serological reactivity of sucrose-derived dextrans.[1]

The utilization of dextrin as the substrate for the formation of serologically reactive material by the *Acetobacter* cultures, in contrast to the utilization of sucrose for the synthesis of serologically similar material by the *Leuconostoc mesenteroides* can best be illustrated by the following experiment. The *Acetobacter* and *Leuconostoc* strains, after preliminary passage in glucose broth, were inoculated into a series of tubes of broth consisting of 0.5% Difco yeast extract plus 5% of one of a number of different carbohydrates, including dextrin and sucrose. The dextrin was of bacteriological grade, that is, a starch-free, alcohol-precipitated product; the sucrose was a sample of beet sugar known to be free of the traces of material precipitating with dextran-reactive antiserums which occur in most lots of reagent and commercial sucrose;[15,16] all the other sugars were of reagent grade. After incubation at 25°C for one week, the cultures which had grown were centrifuged, and their neutralized supernatant fluids tested at several dilutions for capacity to give precipitation with each of two different dextran-

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[7] Day, F. E., and Baker, J. L., *Cent. f. Bakt., Abt. 2*, 1913, **36**, 433.

[8] Hampshire, P., *Bull. Bureau of Bio-Technology*, 1922, **1**, 179, 199.

[9] Shimwell, J. L., *J. Inst. Brewing*, 1936, **42**, 585.

[10] Comrie, A. D., *J. Inst. Brewing*, 1939, **45**, 342.

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[13] Shimwell, J. L., *Wallerstein Labs. Commun.*, 1948, **11**, 27.

[14] Henneberg, W., *Cent. f. Bakt., Abt. 2*, 1898, **4**, 933 (Abstract).

[15] Neill, J. M., Hehre, E. J., Sugg, J. Y., and Jaffe, E., *J. Exp. Med.*, 1939, **70**, 427.

[16] Neill, J. M., Sugg, J. Y., Hehre, E. J., and Jaffe, E., *Am. J. Hyg.*, 1941, **34B**, 65.

TABLE I.
Capacities to Precipitate with Dextran-reactive Antiserums Shown by the Culture Fluids of *Acetobacter* and *Leuconostoc* Grown with Different Carbohydrates.

Strain	Highest dilution of the fluids from cultures with different carbohydrates that gave precipitation with type 2 and with type 20 pneumococcus antiserums*				
	Dextrin	Maltose	Sucrose	Raffinose	Other sugars†
Acetobacter viscosum, 7216	50,000	10‡	0	0	0
" *capsulatum*, 4943	50,000	0‡	0	0	0
Leuconostoc mesenteroides, B	0	0	100,000	20	0

* 0 = no precipitation with either antiserum in tests with 1:10 or 1:100 dilutions of the culture fluids.

† Glucose, fructose, mannose, galactose, sorbose, xylose, arabinose, mannitol, inositol, trehalose, α-methylglucoside, lactose, melibose, or inulin.

‡ Fluids from cultures grown with a different sample of commercial "C. P." maltose gave reactions at dilutions as high as 1:400 with the dextran-reactive antiserums.

reactive antiserums; these were a type 2 pneumococcus antiserum (1:50 dilution) and a type 20 pneumococcus antiserum (1:18 dilution) known to give precipitation with purified *Leuconostoc mesenteroides* strain B dextran in dilutions as high as 1:4 million. Mixtures of 0.2 ml of diluted culture fluid and 0.2 ml of diluted antiserum in clear 10 x 75 mm test tubes were incubated at 37°C for 1 hour and then observed for the development of cloudiness or precipitation.

As shown in Table I, large amounts of material reactive with the type 2 and the type 20 pneumococcus antiserums were present in the fluids of the *A. viscosum* and *A. capsulatum* cultures grown with dextrin, while with one exception, none was detected in the fluids of the cultures grown with other carbohydrates. Traces of reactive material were found in the fluids of the cultures with maltose, but we believe these can be accounted for by the presence of minute amounts of dextrin in the maltose. A large amount of material (dextran) reactive with the same antiserums was produced from sucrose by the *Leuconostoc* culture, and a trace also from raffinose, but none was produced from the dextrin or from any of the other sugars tested.

More complete information on the nature of the serologically reactive substance produced by *A. capsulatum* 4943 has been obtained by studies made with material isolated from a dextrin broth culture. Several liters of viscous culture fluid yielded an amount (approximately 15 g) of serologically reactive polysaccharide which corresponded to more than 20% of the weight of the dextrin used in the medium. An account of the separation, purification, and chemical properties of the *A. capsulatum* polysaccharide will be published in a subsequent report. Results of the serological precipitation tests which we have made with this material in comparison with purified dextran isolated from a sucrose broth culture of *Leuconostoc mesenteroides* B[1] are shown in Table II.

Both the *A. capsulatum* polysaccharide and the *L. mesenteroides* dextran gave visible precipitation with type 2 and type 20 pneumococcus antiserums in dilutions from 1:10,000 to 1:4 million; both gave precipitation also with type 12 pneumococcus antiserum though only at much lower dilutions; and both failed to react at 1:10,000 or higher dilutions with any of the control serums, which included high-titered antipneumococcal serums of several types, normal serums from rabbits which furnished type 2 and 20 serums, and an antiserum capable of precipitating with high dilutions of the polyfructoside levan. The mutual cross-reactions with the types 2, 20 and 12 pneumococcus antiserums, given by the *A. capsulatum* 4943 and *L. mesenteroides* B polysaccharides, and especially the comparably high degree of reactivity versus the types 2 and 20 in comparison to the lower degree of reactivity versus the type 12 are evidences of an extraordinarily close serological likeness between these two materials. Additional evidence for the close serological similarity, not presented in Table II, was that absorption of type 2 pneumococcus anti-

TABLE II.
Serological Similarity of the Dextrin-derived Polysaccharide of *Acetobacter capsulatum* to the Sucrose-derived Dextran of *Leuconostoc mesenteroides.*

Kind of serum	Dilution of serum	Highest dilution of antigen that gave precipitation*	
		Acetobacter capsulatum polysaccharide	*Leuconostoc mesenteroides* dextran
Antipneumococcus, type 2 (Led)†	1:50	4,000,000	4,000,000
" " 2 (rabbit S5)	1:25	2,000,000	2,000,000
" " 20 (" 02)	"	4,000,000	4,000,000
" " 12	"	50,000	10,000
" " 1, 3, 5, 8, 14 (Led)†	1:25, 1:50	0	0
Normal, pre-immunization (rabbits S5, 02)	1:12, 1:25	0	0
Levan-reactive, *Bacillus* N9	1:12	0	0

* 0 = no precipitation with 1:10,000 or any higher dilution of the polysaccharide.
† Refined, or concentrated, rabbit antiserums kindly supplied by Dr. H. D. Piersma, Lederle Laboratories, Pearl River, N.Y.

serum with either polysaccharide removed the antibodies reactive with both polysaccharides.

The enzyme system in *A. capsulatum* responsible for the conversion of dextrin into material serologically like dextran is readily obtainable in solution, free from bacterial cells. Using the fluids from glucose broth cultures as starting material, we have been able to prepare potent concentrates by a method previously described.[5] When "bacteriological" dextrin, Lintner's soluble starch, or certain hydrolysates made either from amylopectin *or from crystalline amylose* are incubated with such enzyme preparations, amylase-resistant polysaccharide material that gives serological precipitation with dextran-reactive antiserums is formed. These results suggest that the action of the *Acetobacter* enzyme is to transfer 1:4 linked glucose units to new polysaccharide material in which the units are linked in some other fashion, very likely 1:6. Although proof of the exact mechanism of the reaction must await the results of systematic chemical study, recognition of the enzymatic convertibility of amylaceous to non-amylaceous polysaccharide by itself is of biological significance.

Summary. Cultures of certain acetic acid bacteria from "ropy" beer, when grown with dextrin but not when grown with other common carbohydrates, produce abundant amounts of polysaccharide material which has serological properties like those of the dextran which is produced from sucrose by *Leuconostoc mesenteroides.* A cell-free enzyme system, capable of causing the conversion of dextrin to material serologically like dextran, has been obtained from one of the cultures.

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17184. Anoxic Diversion of the Renal Cortical Blood Flow.

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During 1947-8 Trueta, Franklin, and Amoroso laid plans for the joint solution of various renal problems, and the research to be described, though in fact of more fortuitous origin, is to be regarded as part of that cooperative effort. Briefly, it was thought probable that the renal cortical blood flow is diverted whenever more important, or temporarily more important, organs are rendered too anoxic. Experiments were, therefore, begun with a view to testing this hypothesis, and in those to be described the whole body was