TABLE 2. *Glucose-6-phosphate dehydrogenase and 6-phosphogluconate dehydrogenase activities of cell-free extracts of Pasteurella pestis and P. pseudotuberculosis**

Organism tested	Activity (units per mg of protein)			
	Incubation at 26 C Substrate		Incubation at 37 C Substrate	
	G6P†	6PG‡	G6P	6PG
P. pestis				
AVO_2	0.0	2.85	0.0	2.47
OX/AVO_2....	0.0	1.91	0.0	1.98
Siam.....	0.0	2.14	0.0	—
OX/Siam.....	0.0	2.44	0.0	—
M23.....	0.0	1.90	0.0	3.43
OX/M23.....	0.0	3.50	0.0	2.50
Yokohama....	0.0	4.22	0.0	—
A12.....	0.0	1.78	0.0	—
Java.....	0.0	1.65	0.0	2.51
TRU	0.0	1.78	0.0	2.29
P. pseudo-tuberculosis				
PBI/+.....	0.76	0.50	2.91	2.90
PBI/−.....	2.96	2.12	1.95	2.72
PTB I (2)....	1.54	1.36	0.30	3.54
PTB II (16)..	1.82	1.58	2.29	2.37
PTB III (43)..	2.21	1.76	1.11	3.61
PTB V (25)...	3.15	1.69	2.50	3.15
2 C.....	3.27	2.65	3.70	2.70
3 C.....	3.55	2.37	—	—
3 E.....	3.95	3.73	—	—
4 C.....	4.73	4.47	—	—

*The assay system consisted of 100 μmoles of phosphate buffer (pH 7.0), 2 μmoles of $MgSO_4$, 0.3 μmole of triphosphopyridine nucleotide, 0.2 ml of cell-free extract, and water to make 2.9 ml; 0.1 ml of 1 M glucose-6-phosphate or 6-phosphogluconate was added to start the reaction. One unit of activity represents an optical density change of 0.01 per min; — = not determined.

† Glucose-6-phosphate.

‡ 6-Phosphogluconate.

tivity when cultured at 37 C. Possible explanations include heat lability of the enzyme, production of an inhibitor, or repression of enzyme formation at 37 C.

From a taxonomic viewpoint, glucose-6-phosphate dehydrogenase activity appears to be as significant in the differentiation between the two species as is the formation of urease.

INHIBITION BY CITRATE OF THE GROWTH OF COAGULASE-POSITIVE STAPHYLOCOCCI

C. G. RAMMELL

Wallaceville Dairy Laboratory, Department of Agriculture, Wellington, New Zealand

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While developing a rapid method for the detection of coagulase-positive staphylococci in cheese, it was found that comparatively low concentrations of sodium citrate inhibited the growth of coagulase-positive staphylococci. The extent and nature of this inhibition were then examined.

Experiments were performed using Oxoid nutrient broth (CM1, Oxo Ltd., London, England) of constant pH but varying citrate concentrations, and broth of constant citrate concentration but varying pH. Broth was inoculated with 0.1 ml of a broth culture (18 hr, 37 C) of *Staphylococcus aureus*, phage type 42D, followed by incubation without shaking, at 37 C. Bacterial growth was estimated, in "nephelos," with a Coleman model 7 photo-nephelometer (Coleman Instruments, Inc., Maywood, Ill.) at

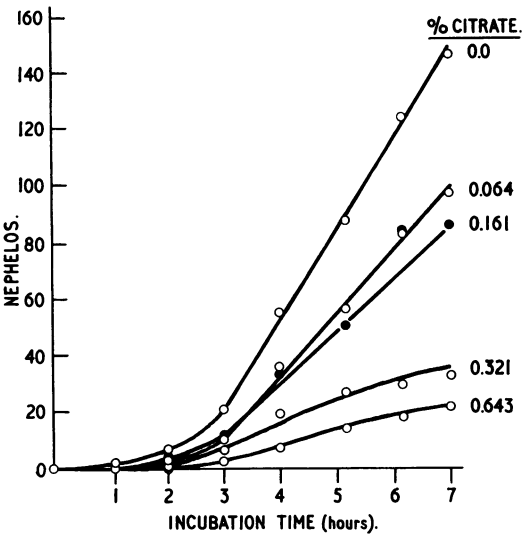

FIG. 1. *Effect of citrate ion concentration on the growth of Staphylococcus aureus, phage type 42D, at pH 7.2. Photo-nephelometer was nulled after the addition of 0.1 ml of an 18-hr (37 C) broth culture of staphylococci to 7.5 ml of broth. The left ordinate is a measure, expressed in arbitrary units of "nephelos," of the numbers of bacteria growing in the presence of five different concentrations of citrate.*

hourly intervals. The broth cultures were briefly agitated for 5 min before every reading, to insure even dispersion.

The effect of increasing concentrations of sodium citrate at pH 7.2 in checking the growth of coagulase-positive staphylococci is shown in Fig. 1. Ninety-five strains of coagulase-positive staphylococci, isolated from dairy products and humans, were tested for sensitivity to citrate. All strains were inhibited, although to varying extents. Any growth that did occur in the presence of citrate tended to be sedimentary. The inhibitory effect of sodium citrate increased with decreasing pH.

Since citrate is known to sequester certain ions shown by Shooter and Wyatt (Brit. J. Exptl. Pathol. **36**:341, 1955) to be essential for the growth of coagulase-positive staphylococci, it seemed likely that its inhibitory action could be overcome by the addition of these essential ions. Inhibition by citrate was, in fact, overcome completely by Ca^{++} and partially by Mg^{++}. Optimal growth could be achieved without precipitation of citrate by the combined addition of Ca^{++} and Mg^{++}. This finding has been used to devise a medium of high osmotic pressure and citrate concentration suitable for the direct solution and culture of cheese for coagulase-positive staphylococci. The composition of this medium is: Na citrate·$2H_2O$, 100 g; $CaCl_2$, 3.9 g; $MgCl_2$, 4.6 g; Oxoid CM 1 nutrient broth granules, 13 g; distilled water, 1 liter; pH adjusted to 7.5 with 1 N HCl.

No study has yet been made of the effect of citrate on coagulase-negative staphylococci, but the work of Richardson (Nature **189**:78, 1961) on the use of chelating agents for the selective culture of coagulase-positive staphylococci suggests that they may respond differently.

INABILITY OF THYMINE TO REPLACE URACIL FOR GROWTH OF *THERMOBACTERIUM ACIDOPHILUM* (*LACTOBACILLUS ACIDOPHILUS*)

C. BISWAS AND H. P. BROQUIST

Laboratory of Biochemistry, Department of Dairy Science, University of Illinois, Urbana, Illinois

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Recent studies with *Neurospora crassa* (Fink and Fink, Biochem. Biophys. Research Communs. **6**:7, 1961) and *Bacillus cereus* (Sells, Biochim. et Biophys. Acta **40**:548, 1960) have provided some evidence that thymidine may be converted to uracil derivatives. A further indication that this might be a more general phenomenon in microorganisms appeared likely from a report of Løvtrup and Shugar (J. Bacteriol. **82**:623, 1961), who claimed that either thymine or uracil could satisfy the pyrimidine requirement of *Thermobacterium acidophilum* (*Lactobacillus*