

TABLE I.
Action of Cholinesterase and an Intracellular Factor in Synthesis of AC.

Samples*	Total AC in γ †		% destruction of added AC‡ by cholinesterase in 10 min.	
	I	II	I	II
5 g washed tissue	216.0	152.0	5	15
5 g washed tissue + 150 γ Es	262.0	166.5	0	0
5 cc juice	1.7	1.6	82+	55
5 cc juice + 150 γ Es	1.7	1.7	0	0
5 g washed tissue + 5 cc juice	185.0	120.6	—	38
5 g washed tissue + 5 cc juice + 150 γ Es	559.8	361.7	—	0

*Three hours are allowed for the action to proceed in all the samples before the extraction.

†Assayed on the rectus as AC-chloride according to Chang and Gaddum.⁶

‡Assayed on the rectus according to Meng.⁷

when the juice is added to the washed tissue in the presence of Es, the AC-yield is very much increased, indicating clearly the collaboration of the two factors in the biological synthesis of AC.

Further experiments are in progress to determine the nature of the intracellular factor and to search for the source of the substrate concerned in this synthesis.

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Recovery of Antibody from Immune Precipitate of Type B Friedländer Bacillus.

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In previous reports¹ from this laboratory, it has been shown that free antibody can be recovered from the immune precipitate or agglutinate of Type I pneumococcus by treating with dilute acid which causes a shift of the antigen-antibody equilibrium. The present communication extends our observation to the immune precipitate of another organism, Type B Friedländer bacillus.

⁶ Chang, H. C., and Gaddum, J. H., *J. Physiol.*, 1933, **79**, 255.

⁷ Meng, C. W., *Chinese J. Physiol.*, 1940, **15**, 143.

¹ Liu, S. C., and Wu, H., *Chinese J. Physiol.*, 1938, **13**, 449; *ibid.*, 1940, **15**, 465; *Proc. Soc. Exp. Biol. and Med.*, 1939, **41**, 144; *ibid.*, 1940, **43**, 747; Lee, K. H., and Wu, H., *Proc. Soc. Exp. Biol. and Med.*, 1940, **43**, 65.

Antisera of horse and rabbit origin were prepared by immunizing animals with heat-killed Type B Friedländer bacilli. The specific polysaccharide of this organism was prepared according to the method of Heidelberger, Goebel, and Avery.² Polysaccharide was added to the antisera to form the specific precipitate. The precipitate was washed and suspended in 2% or 3% NaCl solution. The use of NaCl increases the percentage of recovery by decreasing the solubility of the precipitate.³ Four cc portions of the suspension, containing about 5 mg N, were treated with equal volumes of dilute HCl of different concentrations according to the procedure previously described.³ The percentage of recovery and the solubility of "acid-precipitates" (that is, the precipitate from which a part of the antibody has been set free by acid) at different pH's were calculated. Results are shown in Fig. 1. For the sake of comparison the curves

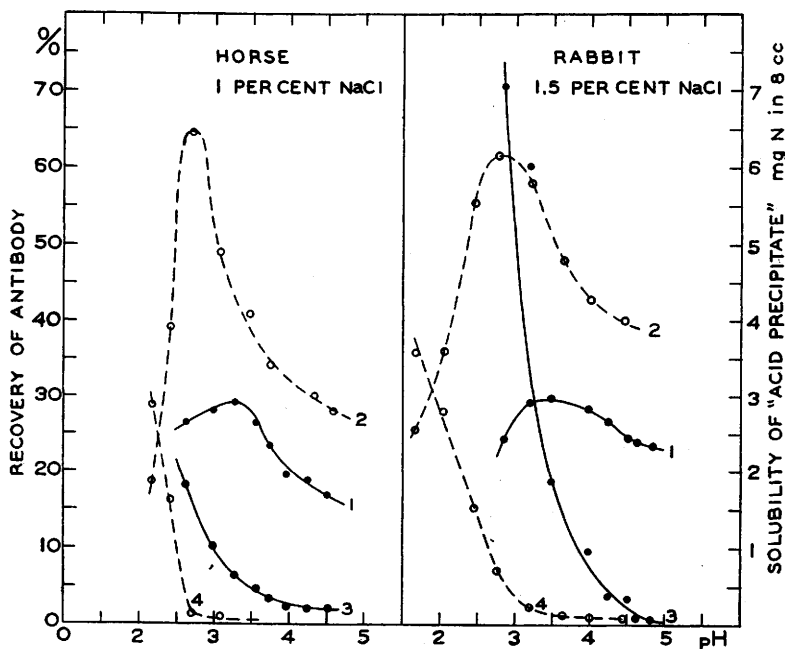


FIG. 1.

Recovery of antibody from immune precipitates of Type B Friedländer Bacillus at different pH's. $t = 25^{\circ}\text{C}$.

1. Percent recovery from Friedländer Bacillus immune precipitate.
2. Percent recovery from Pn I immune precipitate.
3. Solubility of Friedländer Bacillus acid precipitate.
4. Solubility of Pn I acid precipitate.

² Heidelberger, M., Goebel, W. F., and Avery, O. T., *J. Exp. Med.*, 1925, **42**, 709.

³ Liu, S. C., and Wu, H., *PROC. SOC. EXP. BIOL. AND MED.*, 1939, **41**, 144.

for Type I pneumococcus previously reported³ are also shown in the figure.

For the precipitate obtained with horse serum in 1% NaCl, the optimal pH for recovery was 3.25, while for that with rabbit serum in 1.5% NaCl, the optimal pH was 3.50. These pH values are distinctly different from those (2.75 and 2.70) found for Type I pneumococcus under similar conditions. The optimal pH for the recovery of the antibody thus depends more on the nature of the specific polysaccharide than on the species of animal that produces the antibody.

The solubility of the acid-precipitate from rabbit serum was higher than that from horse serum, even though the solubility of former had been reduced considerably by the use of a higher concentration of NaCl (1.5%) than that (1%) used in the latter.

The maximal recovery was about 30% in both cases. This recovery is much lower than that (60%) previously reported for Type I pneumococcus. This difference is due to the higher solubility of the acid-precipitate of Type B Friedländer bacillus. The dissolved acid-precipitate decreases the percentage of recovery by recombination with the liberated antibody.

The purity of the recovered products was tested by quantitative precipitation. To 4 cc portions of the recovered antibody-solution (0.25 mg N per cc) were added 1 cc portions of the specific polysaccharide solution of different concentrations. The precipitates were washed and their N contents determined. At the point of maximal precipitation, the products recovered from the immune precipitates of horse and rabbit origin were respectively 89% and 85% precipitable.