

the first step in the formation of a new generation. The information obtained from a study of the vertical sections of both human and bovine colonies of *M. tuberculosis* would seem to substantiate this claim.

It seems as though we must conceive of the colony as a half sphere and presumably some growth is taking place throughout the entire area as evidenced by the few non-acid-fast rods and granules which may be seen scattered even through the strongly acid-fast "medulla". The peripheral zone, however, reveals considerably larger numbers of these forms, and is therefore presumably the area in which most active growth is taking place.

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Precipitins Against Fractions of Streptococci in Hemolytic Streptococcus Disease, Glomerular Nephritis, Rheumatoid Arthritis, and *S. Viridans* Endocarditis.

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The sera of 310 patients have been studied for precipitins against 2 protein fractions of *S. hemolyticus*, the nucleoprotein of *S. viridans* and the group specific carbohydrate of *S. hemolyticus*. The *S. hemolyticus* fractions were freshly prepared. The *S. viridans* protein first used* was 5 years old. Later tests with a sample newly isolated from the same strain gave identical results. A description of the precipitin test and the antigens used has been presented in a previous communication.¹

Sera from the following groups of cases have been studied:

1. A control group of 39 healthy nurses during the fall season.†
2. A control group of 16 healthy medical students and nurses during the spring season.

* Kindly supplied by Dr. Rebecca C. Laneefield of the Hospital of the Rockefeller Institute for Medical Research.

¹ Seegal, D., Heidelberg, M., and Jost, E. L., *PROC. SOC. EXP. BIOL. AND MED.*, 1932, **29**, 939.

† These data were made available by Dr. A. F. Coburn.

3. A control disease group of 17 cases with temperatures of 102° or over. These patients were not affected with hemolytic streptococcus disease. All these sera were obtained during the spring season.

4. A control disease group of 100 cases during the spring season. None of these patients was known to have rheumatic fever, rheumatoid arthritis, sub-acute *S. viridans* endocarditis, proven hemolytic streptococcus disease, glomerular nephritis or peptic ulcer.

5. Twenty-three cases of proven sub-acute *S. viridans* endocarditis.¹

6. Thirty-six cases of rheumatoid arthritis.‡

7. Sixteen cases of proven hemolytic streptococcus disease.

8. Fourteen cases of acute glomerular nephritis.

9. Sixteen cases of sub-acute glomerular nephritis.

10. Seven cases of healed glomerular nephritis.

11. Five cases of healed glomerular nephritis during hemolytic streptococcus infection.

12. Twenty-one cases of peptic ulcer with symptoms.

A series of tests was made on the sera of each patient with hemolytic streptococcus infection and glomerular nephritis. The precipitin test shown in the table was the maximum value obtained during the 2 to 5 week period after the known onset of the patient's illness. This procedure depended upon our experience that the precipitin test against fractions of the hemolytic streptococcus in frank hemolytic streptococcus disease and glomerular nephritis is negative or very weak in the early days of the disease. Usually 2 to 5 weeks are required for the development of the antibody if it is to appear.

The accompanying table illustrates the variation in the maximum degree of precipitin formation in the groups studied thus far.

The following conclusions may be drawn: 1. Precipitins against the 3 fractions of the *S. hemolyticus* and the one *S. viridans* protein are absent or minimal in the 4 control groups. 2. Strong precipitin formation against the hemolytic streptococcus protein antigens is noted chiefly in the cases of subacute *S. viridans* endocarditis and rheumatoid arthritis. 3. Moderate precipitin formation against these same fractions is noted in many of the cases of proven hemolytic streptococcus disease, acute and subacute glomerular nephritis and peptic ulcer.

4. The strongest reactions against the group specific carbohydrate of *S. hemolyticus* occur in the sera of patients with either proven hemolytic streptococcus infection or rheumatoid arthritis.

‡ These sera were made available by Dr. M. H. Dawson.

TABLE I.
Precipitins in Human Sera Against Fractions of *S. hemolyticus* and *S. viridans*.

Precipitin titer [§]	Control Fall Group	Control Spring Group	Control Disease Fever Group 102° or over	Control Disease Group	<i>S. viridans</i> endocarditis	Rheumatoid Arthritis	<i>S. hemolyticus</i> Infection	Acute Glomerular Nephritis	Sub-acute or Chronic Glomerular Nephritis	Healed Glomerular Nephritis	Healed Glomerular Nephritis during <i>S. hemolyticus</i> infection	Peptic Ulcer with Symptoms
<i>S. Hemolyticus</i> Nucleoprotein D												
++++	0	0	0	0	2	3	0	0	0	0	0	0
+++	0	0	0	0	9	7	3	1	2	0	0	2
++	0	0	0	3	8	10	7	6	4	0	2	5
+	0	6	2	24	3	13	3	2	3	2	1	6
0	39	10	15	73	1	3	3	5	6	5	2	8
Total No. Cases	39	16	17	100	23	36	16	14	15	7	5	21
<i>S. Hemolyticus</i> Nucleoprotein K												
++++	0	0	0	0	1	6	1	1	0	0	0	0
+++	0	0	0	0	16	11	3	6	2	0	0	2
++	0	0	0	0	0	3	3	1	2	0	0	1
+	0	0	0	8	2	7	5	1	1	1	0	6
0	39	16	17	92	4	9	4	5	11	5	4	12
Total No. Cases	39	16	17	100	23	36	16	14	16	6	4	21
<i>S. Hemolyticus</i> Carbohydrate C.												
++++	0	0	0	0	1	2	0	0	0	0	0	0
+++	0	0	0	2	6	3	1	0	0	0	0	1
++	0	0	2	1	5	2	3	0	0	0	0	0
+	0	0	2	4	2	0	0	2	1	2	1	1
0	16	17	96	16	22	9	10	14	5	3	19	
Total No. Cases	16	17	100	23	36	16	14	16	6	5	21	
<i>S. Viridans</i> Nucleoprotein												
++++	0	0	0	1	0	0	0	0	0	0	0	0
+++	0	0	0	3	0	0	0	0	0	0	0	0
++	0	0	0	2	1	0	0	0	0	0	0	0
+	0	2	0	6	2	0	1	0	0	0	0	1
0	16	7	76	11	25	13	11	10	7	1	20	
Total No. Cases	16	9	76	23	28	13	12	10	7	1	21	

Nucleoprotein D: Acetic acid precipitable *S. hemolyticus* protein extractable at neutrality; concentration, 1:2000.

Nucleoprotein K: Acetic acid precipitable *S. hemolyticus* protein extractable (after removal of less alkaline extracts) between pH 11 and 13.3; concentration, 1:2000.

Carbohydrate C: Species specific polysaccharide; concentration, 1:200,000.

Concentration of *S. viridans* nucleoprotein: 1:2000.

§ The precipitin titer was determined as in (1). These readings represent the precipitates after standing over night in the cold and centrifuging.

Seven members of the group of 16 cases of proven hemolytic streptococcus infection showed a strong anti-carbohydrate reaction. Four of these 7 had positive blood cultures of hemolytic streptococcus.

Only one of the 9 remaining cases with negative anti-carbohydrate reactions had a positive blood culture. This fact is in agreement with our unpublished observations that rabbits injected subcutaneously with heat-killed *S. hemolyticus* only occasionally develop precipitins against the carbohydrate fraction, although the same rabbits generally develop this precipitin when the organism is injected intravenously.

5. The cases of *S. viridans* endocarditis were the only group in which precipitin formation against the *S. viridans* nucleoprotein was marked. It is of interest, in this respect, that the precipitin reactions in the cases of rheumatoid arthritis more closely correspond to those seen in hemolytic streptococcus disease than to those found in *S. viridans* endocarditis.

6. The *S. hemolyticus* protein precipitin tests in the cases of acute and subacute nephritis in this series closely parallel those found in proven hemolytic streptococcus disease. This observation is additional evidence in favor of the opinion that glomerular nephritis may be related to hemolytic streptococcus infection. The 7 cases of healed glomerular nephritis show no significant precipitin formation. In this series, in 5 cases of healed glomerular nephritis a hemolytic streptococcus infection occurred without an exacerbation of the nephritis. Precipitin formation in this group is present, but diminished.

7. The finding of precipitins in the sera of patients with active peptic ulcer is unexplained, and is of interest since Derick and Fulton² obtained a high percentage of positive skin reactions in this group with comparable streptococcus protein fractions.

8. Some sera from normal patients and those with streptococcus disease which show no precipitins when tested within a few days may give a moderately positive test after remaining in the ice-box one to 9 weeks. The development of this reaction is in many instances paralleled by a non-specific reaction against a typhoid nucleoprotein. With few exceptions, inactivation of these old sera by heating at 56°C. for one-half hour prevents this reaction.

² Derick, C. L., and Fulton, M. M., *J. Clin. Invest.*, 1931, **10**, 121.