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Application of the Technique of Slide Agglutination of Hemolytic Streptococci to Human Sera.*

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This communication is to report that positive results can be obtained by applying the method of slide agglutination to human serum and hemolytic streptococci.

The materials used in the study consisted of strains of hemolytic streptococci and human blood sera. The streptococci were cultured from the nose, throat or suppurative foci of 22 patients ill with streptococcal disease. The blood sera were collected from these same patients for the most part at the end of the third week of their illness.

The cultures used originated from a single colony fished from the original blood agar plate of each patient. The colony was inoculated into rabbit blood broth, and 1 cc of the 18 hour culture was transferred to 5 cc of beef heart infusion broth containing 0.05% dextrose. The resulting 18 hour culture was centrifuged at 2000 rpm for one half hour and the supernatant fluid decanted. If the resulting thick suspension was granular, the culture was transferred to 9 cc of the dextrose broth to which had been added 1 cc of bacto-trypsin. The 18 hour growth was likewise treated by centrifuging and decanting and the remaining thick suspension was usually smooth. Typing was carried out by using one loopful of this suspension and one loopful of the rabbit typing serum, diluted to the optimal strength. The 2 were mixed on a slide by tapping the drop 5 to 6 times with the loop and rotating the slide rapidly 5 to 6 times while tilted at an angle of approximately 45 degrees. The organism was considered to be satisfactorily typed only when gross agglutination took place within 5 seconds.

Tests with the patient's sera were done in a manner similar to that used with the rabbit typing sera. Each human sample was tested with the strain of hemolytic streptococci from that patient and with a number of homologous and heterologous strains from other patients. The sera were used both undiluted and serially diluted with

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TABLE I.
Results of Slide Agglutination of Serum of Patient P.B. (Type 1) and Patient A.P. (Type 3) and Homologous and Heterologous Streptococci.

Patient	Strain Type	Serum PB		Patient	Strain Type	Serum AP	
		0	1-10			0	1-10
PB	1	+	0	AP	3	+	0
FM	1	+	0	SB	3	+	0
AC	1	+	0	RB	3	+	0
JG	NT*	0	0	JD	3	+	0
BB	NT	0	0	LB	3	+	0
FR	1	+	0	PB	1	0	0
AR	1	+	0	FM	1	0	0
AP	3	0	0	JG	NT	0	0
SB	3	0	0	BB	NT	0	0
RB	3	0	0	PS	1	0	0
PS	1	+	0	PA	1	0	0
PA	1	+	0	FR	1	0	0
JD	3	0	0	AR	1	0	0
LB	3	0	0	AC	1	0	0

*NT = No type obtained with available rabbit sera.

normal saline 1-10, 1-20, 1-40 and in some instances 1-80. No agglutination was found beyond 1-80.

The streptococci were typed, when possible, by the method of slide agglutination as described by Griffith¹ and modified by Boisvert.² Type 1 was recovered from 8 patients and Type 3 from 7 patients. Strains from 7 patients could not be typed with the rabbit sera available in our laboratory. Sera from 6 of the 22 patients were found to contain agglutinins for their own strain of streptococci and for homologous strains from other patients, but not for heterologous strains from other patients. Results from 2 of these patients (a Type 1 and a Type 3) are shown in Table I.

Summary. Twenty-two samples of human sera were tested and 6 were positive. They gave agglutination with their own and all homologous strains used. Crossed agglutinations were seen with none, but the element of time in the method employed should be reiterated. The agglutinations were read immediately and by 30 seconds the slides were discarded. The 6 positive sera were from 3 patients with purulent otitis media, 2 of these scarlatinal, 2 patients with empyema, and one patient with paronychia and axillary adenitis.

Conclusions. In 6 of 22 samples of convalescent human sera slide agglutination with the homologous hemolytic streptococci was detectable.

¹ Griffith, F. J., *J. Hyg.*, Cambridge, England, 1934, **34**, 542.

² Boisvert, P. L., *Science*, 1941, **94**, 193.