few weeks in the group with renal involvement. Whether kidney damage results in greater retention or chemical modification of administered cortisone, or whether the response of the diseased kidney to the steroid is altered, cannot be determined from the present study. It is of interest that hypertension associated with desoxycorticosterone can be achieved more readily in nephritic rats (5). It is recommended that cortisone be used with greater care in patients with renal disease, particularly if antecedent hypertension is evident.

Conclusions. Modification of arterial tension by cortisone, in the doses generally employed, has been noted but rarely in short- and long-term observations of subjects without renal disease. A rise in blood pressure has been the rule among patients with renal damage of varying types irrespective of the presence or absence of antecedent hypertension.

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Specificities of Hyaluronidases Formed by Several Groups of Streptococci.* (18567)

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In a previous study(1) enzyme-inhibitors appearing in human serums during streptococcal infections could not be accounted for on the assumption that Group A enzymes were type-specific. It appeared that heterotypic Group A strains of streptococci gave rise *de novo* to analogous enzyme-inhibitors. Since then, some experimental observations have been made in regard to the relative specificities of hyaluronidases produced by Groups A, B, C and G strains of streptococci. These studies form the basis for the present report.

Materials and methods. Streptococcal hyaluronidases. One hundred and fifteen strains of streptococci‡ were studied for hyaluronidase activity. Each strain was grown in buffered Todd-Hewitt broth for a period of 15 to 18 hours. The enzyme titer was determined by the mucin clot prevention test of McClean (1,3). Streptococcal hyaluronidases of satisfactory titer (*e.g.* >1:16) were stored in sealed glass ampoules at −70°C. The number of strains in each group studied and the number producing enough enzyme for working purposes appear in Table I.

Enzyme-inhibitor. Hyaluronidases of high activity (titer of 1:128 or more) were derived from 10 strains of streptococci.§ Bacteria-free filtrates were titered and aliquots of ac-

TABLE I. Hyaluronidase Activity of Streptococci.

Group	No. of strains	Enzyme activity			No. of enzymo producers	% producing enzyme
		<16*	16-128	128-1024		
A	35	27	7	1	8	23
B	7	5	2	0	2	29
C	28	15	10	3	13	46
D	16	16	0	0	0	0
G	28	10	13	5	18	64
K	1	1	0	0	0	0

* MCP test; reciprocal of enzyme dilution.

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1. Friou, G. J., and Wenner, H. A., *J. Infect. Dis.,* 1947, v80, 185.

‡ Acknowledgement is made to Drs. Rebecca Lancefield, Lowell Rantz, Nobel Sherwood, Elaine Updyke, and Julia Coffey for supplying many of the strains used in the study.

3. McClean, D., Rogers, H. J., and Williams, B. W., *Lancet,* 1943, v1, 355.

TABLE II. Hyaluronidase Inhibitor Titers of Immunized Rabbits.

Group	Strain	Enzyme-inhibitor titer*
A	Type 3 (Meeks)	128
A	Type 4 (T4/95/Rb5)	128
A	Type 4 (Bois)	256
A	Type 22 (T22/83/3)	64
B	V9	0
C	13	256
C	J148B/0/3	128
C	B3	64
G	K127	128
G	SS258	256

* Expressed as reciprocal of serum dilution; 16 antigenic units of homotypic enzyme preinoculation serums did not contain inhibitor.

tive enzymes were inoculated into 24 rabbits by intravenous and intramuscular routes. Enzyme-inhibitors in varying amounts, appeared within 2 to 6 weeks. The best enzyme-inhibitor titers obtained appear in Table II. *Enzyme-inhibition.* Quinn's modification of the method described by McClean(2,3) was used. The specificity of streptococcal enzyme-inhibitors was also studied using serial dilutions of serums and enzymes in box titrations. *Antibody absorption.* Cross absorption tests with enzymes and bacterial cells and immune serums derived from several strains of Groups C and G streptococci were made. Equal parts of 1:4 dilutions of an antiserum were separately mixed with enzyme and washed streptococcal cells. The mixtures were placed in a waterbath for one hour at 37°C, and then in the icebox (4°C) overnight. Enzyme-inhibition tests were made using serial dilutions of serums and enzymes in box titrations. Enzyme and serum controls were included in the tests.

Results. Type specificity of hyaluronidases from Group A streptococci. Among 35 strains of Group A streptococci, 8 produced sufficient hyaluronidase for inclusion in the tests.

The specificity of hyaluronidases obtained from 6 strains of known type was measured by reciprocal enzyme-inhibition reactions. The results of these tests, which appear in Table III, indicated that several heterotypic strains of Group A streptococci produced enzymes with similar characteristics.

Quantitative distinctions may be observed in the reciprocal neutralization of heterotypic Group A strains. These variations do not indicate that there are major differences in enzyme configuration as measured by enzyme-inhibitor, for among rabbits inoculated with identical enzymes and producing, as a rule, inhibitor with heterotypic overlap, several serums inhibited only the parent enzyme.

Specificity of hyaluronidases of Groups A, B, C and G streptococci. Twenty-five strains of streptococci were studied by reciprocal enzyme-inhibition reactions. These tests revealed that Groups A and B enzymes were serologically distinct. Enzymes obtained from Groups C and G strains were serologically related but were unrelated to the Group A or B enzymes.

The serologic distinction of these streptococcal enzymes was explored further in box titrations of enzymes and enzyme-inhibitors. The specificities of Groups A and B hyaluronidases and the serologic overlap of enzymes derived from Groups C and G strains were again observed. Additional tests, with Group specific enzyme-inhibitors and enzymes prepared from Groups A, C and G strains other than those tabulated here, reaffirmed these relationships. Results obtained in one of these tests appear in Table IV.

The immunologic relationships of Groups C and G hyaluronidases were further determined by reciprocal neutralization and absorption. These studies indicated that a single exposure of serums to (a) active enzymes or (b) washed bacterial cells of Group C or G reduced Group C and G enzyme-inhibitor titers, respectively.

Discussion. The specificity of hyaluronidases formed by streptococci has not been explored, except sketchily. McClean(4) prepared enzyme-inhibitors with three Group C strains and found that Group A enzymes were not inactivated by them. Friou(5) observed the failure of human Group A enzyme-

§ Dr. E. L. Updyke, Communicable Disease Center, United States Public Health Service, kindly rechecked the group and type of streptococci used in these tests.

2. Quinn, R. W., *J. Clin. Invest.*, 1948, v27, 471.

4. McClean, D., *Biochem, J.*, 1943, v37, 169.

5. Friou, G. J., *J. Infect. Dis.*, 1949, v84, 240.

TABLE III. Type Specificity of Group A Hyaluronidases.

Serum-inhibitor	Reciprocal of serum dilution inhibiting enzyme* action					
	Type 3 (Meeks)	Type 4 (Bois)	Type 4 T4/95/Rb5	Type 4	Type 22 T/22/83/3	Type 22
Type 3 (Meeks)	128	64	128	128	16	32
" 4 (Bois)	32	16	128	16	4	16
" 4 (T4/95/Rb5)	128	128	128	256	0	128
" 4 (101)	32	32	128	32	32	32
" 22 (T22/83/3)	0	0	0	8	64	0

* 16 units of enzyme.

TABLE IV. Group Specific Enzyme-Inhibitor Titers of Groups A, B, C, and G Streptococcal Hyaluronidases.

Antigenic units of enzyme	Final dilutions of group specific enzyme-inhibitors* inactivating indicated enzymes											
	A enzyme-inhibitor				C enzyme-inhibitor				G enzyme-inhibitor			
	A_e	B_e	C_e	G_e	A_e	B_e	C_e	G_e	A_e	B_e	C_e	G_e
2	128	2	0	0	0	0	128	96	128(?)	0	64	64
4	96	0	0	0	0	0	96	128	0	0	24	48
8	64	0	0	0	0	0	32	32	0	0	8	32
16	16	0	0	0	0	0	16	16	0	0	<8	16
32	16	0	0	0	0	0	<8	0	0	0	<8	8
64	0	0	0	0	0	0	0	0	0	0	0	0

A_e = Group A, type 4 enzyme. C_e = Group C, strain 13 enzyme.
B_e = Group B, strain V9 enzyme. G_e = Group G, strain SS258 enzyme.
(?) Insufficient enzyme, control at 1:2 dilution of enzyme gave partial hydrolysis.
* Enzyme-inhibitor specific for Group B enzymes could not be prepared; neither of two strains studied produced enough enzyme.

inhibitors to affect hyaluronidases produced by Group C and G streptococci. The latter observations have failed to define group specificity of hyaluronidases. They do point out, once again, that in the natural history of human streptococcal infections most are caused by Group A strains.

The finding that enzyme-inhibitors prepared from Groups C and G enzymes were serologically similar was of interest because of other relationships found among these groups. Hare(6) found some serums produced by the injection of Group C strains to give cross precipitation with Group G strains; this usually occurred, however, if the animals had been immunized for a longer period than usual to obtain a satisfactory titer. Lancefield(7) observed that Groups C and G streptococci possessed a common protein antigen which was responsible for the confusing cross reactions observed by Hare. It is not suggested here that this protein antigen is related to hyaluronidases produced by Groups C and G streptococci. It may or may not be. We wish only to add at this time that some strains in these Groups shared the distinction of forming hyaluronidases possessing similar antigenic components.

Summary. Hyaluronidases produced by strains of Groups A and B streptococci are serologically distinct. Hyaluronidases produced by strains of Groups C and G are serologically indistinct. Each of the latter is distinctly different from hyaluronidases formed by strains of streptococci, Groups A and B.

6. Hare, R., *J. Path. and Bact.*, 1935, v41, 499.

7. Lancefield, R., The Harvey Lectures, 1940-1941, series 36, 267.

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