2. The toxicity of acetylsalicylic acid is not influenced by the simultaneous administration of magnesium oxide. 3. The toxicity of a mixture of 50% magnesium oxide and 50% of acetylsalicylic acid is half as great as the toxicity of the amount of acetylsalicylic acid which is required to produce the same therapeutic effect.

8804 C

Incidence of Hemolytic Streptococci and Pneumococci in the Pharyngeal Flora of Normal Rhesus Monkeys.

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The normal bacteriologic flora of the pharynx of chimpanzees has been described by Dochez, Shibley and Mills[1] in connection with their experiments on the transmission of the common cold. The bacteria occurring in the throats of apes were similar, for the most part, to those found in human throats, although the percentile incidence varied for some organisms. Apparently, no such complete study of the pharyngeal flora of lower primates has been reported. The few reports of throat-cultures in the monkey are primarily concerned with the presence of *C. diphtheriæ* in the nasopharynx. Dold and Weigmann,[2] and Ramon and Erber[3] found diphtheria-like organisms in some monkeys, but Jungeblut[4] in 50 normal rhesus monkeys found no *C. diphtheriæ*. Dold and Weigmann reported the presence of staphylococci and streptococci but did not specify the reaction of these organisms in blood-media.

During studies of experimentally induced hemolytic streptococcal infection of rhesus monkeys, a survey of the incidence of hemolytic streptococci in the normal throats of 49 animals* was undertaken. The cultures were taken during the 2nd week of January, 1936, and within 9 days of receiving the animals in the laboratory from the dealer. A swab was rubbed firmly over the faucial surface, streaked

1 Dochez, A. R., Shibley, G. S., and Mills, K. C., *J. Exp. Med.*, 1936, **52**, 701.

2 Dold, H., and Weigmann, F., *Zeitsch. f. Hyg.*, 1934, **116**, 154.

3 Ramon, G., and Erber, B., *Revue D'Immunologie*, 1935, Sept., 415.

4 Jungeblut, C. W., *J. Imm.*, 1934, **27**, 17.

* These monkeys were made available through the courtesy of Dr. Jungeblut.

on sheep-blood agar, and after 24 hours' incubation, the plates were searched for hemolytic colonies.

From 28 of the 49 cultures a β-streptococcus[5] was isolated. On one plate a minute streptococcus of the type described by Bliss and Long[6] was also isolated. These organisms were tested for production of a soluble hemolysin, lysis of human and rhesus plasma-clot (Tillett and Garner[7]) and fermentation of lactose, mannite, and salicin. The antigenic group to which the organisms belonged was determined by the method of Lancefield.[8] Six of the strains were tested for the production of a toxin reactive in the skin of silver-fox rabbits.

The results of these tests are summarized in Table I: 19 of the strains fell in Group A, 4 in Group C, 5 in Group G, and 1, the minute streptococcus in Group F.

All Group A organisms were *Streptococcus pyogenes* by Holman's classification.[9] The diameter of hemolysis in rabbit-blood agar† pour-plates was 2.5 mm. A soluble hemolysin active against rabbit red blood cells was produced and a fibrinolysin which dissolved human-plasma clot within 3½ hours. The lysis of monkey-plasma clot required a longer time, for it was complete within 6 hours of initial observation in the case of only 3 strains. Seven of the strains produced complete lysis within 22 hours, another 7 were partially lytic in 24 hours, and 2 produced no lysis in 24 hours. A toxin active in the skin of silver-fox rabbits was formed by all the 6 strains tested.

The 4 strains of hemolytic streptococci belonging to Group C resembled the organisms of Group A in sugar-fermentations and elaboration of a soluble hemolysin and fibrinolysin. The diameter of hemolysis in pour-plates was 4 mm.

One of the 5 strains of hemolytic streptococci belonging to Group G was *pyogenes* and the other 4 were fermentatively *equi*. The diameter of colonial hemolysis in a rabbit-blood pour-plate was 4 mm. Their fibrinolytic and hemolytic activity was similar to that of the members of Groups A and C.

The streptococcus of Group F, *equi*, produced the characteristically small area of hemolysis in pour-plates with an almost micro-

[5] Brown, J. H., *Mono. Rockefeller Inst. Med. Res.*, 1919, **9**.

[6] Bliss, E. A., and Long, P. H., *J. Bact.*, 1934, **27**, 105.

[7] Tillett, W. S., and Garner, R. L., *J. Exp. Med.*, 1933, **58**, 485.

[8] Lancefield, R., *J. Exp. Med.*, 1933, **57**, 571.

[9] Holman, W. L., *J. Med. Res.*, 1916, **34**, 377.

† This medium contained 1% neopeptone.

TABLE I.
Cultural and Immunological Characteristics of Hemolytic Streptococci Isolated from the Throats of Normal Rhesus Monkeys.

No. of Strains	Fermentation			Pour plate; diam. of hemolyzed zone	Soluble hemolysin	Group	Fibrinolysis; human clot; time for lysis	Fibrinolysis; rhesus clot; time for lysis	Toxin
	Lactose	Mannite	Salicin						
19	+	0	+	2.5 mm.	+	A	19 min. to 3½ hrs.	3 hrs. to 0 in 24 hrs.	All 6 tested positive
4	+	0	+	4 mm.	+	C	19 min. to 3¼ hrs.	2¼ to 24 hrs.	
1	+	0	+	4 mm.	+	G	1½ hrs.	25% in 6 hrs. 100% in 24 hrs.	
4	0	0	+	4 mm.	+	G	1¼ to 2 hrs.	10% to 100%, 24 hrs.	
1	0	0	+	1 mm.	0	F	None, 24 hrs.	None, 24 hrs.	

scopic rosette colony. No soluble hemolysin or fibrinolysin was formed.

Of the colonies on the original plates a large number resembled pneumococcal colonies. Typable pneumococci were isolated from 33 of the 49 cultures: 3 type III, 9 type IV, 1 type V, 15 type VII, 1 type VIII, 3 type XIX, and 1 type XXXII. A rough pneumococcus agglutinating in all antipneumococcal sera was obtained from 2 cultures and a third culture yielded a pneumococcus which failed to agglutinate in any of the 32 type-sera.

A single throat-culture from a group of 49 normal rhesus monkeys yielded hemolytic streptococci in 29 and type-specific pneumococci in 33 cases. Nineteen of the 29 hemolytic streptococci belonged to human Group A, 4 to Group C, 5 to Group G, and 1 to Group F. The pneumococci belonged to types III, IV, V, VII, VIII, XIX, and XXXII, with types IV and VII predominating. These organisms were present in considerable numbers. Some of the plates contained as many as 50 isolated hemolytic colonies and the pneumococci also were well represented.

It is noteworthy that hemolytic streptococci isolated from the throats of monkeys are better able to lyze human than monkey-plasma clot. In this respect they are similar to the human microorganisms studied by Van Deventer and Reich.[10] It is also to be noted that the representatives of Groups C and G were fibrinolytically similar to Group A. These findings diverge from those of Madison,[11] who found that hemolytic streptococci isolated from diseases of animals were usually specifically lytic for their clotted plasma and suggested that the fibrinolysis of human plasma was associated specifically with Group A streptococci. Tillett[12] also reported that of 21 Group C organisms tested, only one was fibrinolytic for human plasma. Hare,[13] on the other hand, found all of 15 Group C and 9 of 13 Group G cocci fibrinolytic for human plasma; this agrees with the results obtained with our strains from monkeys. Possibly the interval between isolation and testing for fibrinolysis accounts for these differences, and the host from which the Group C and G organisms were obtained may influence the results also.

The animals studied had been in captivity for some time and it is therefore possible that they had acquired the hemolytic streptococci

10 Van Deventer, J. K., and Reich, T., PROC. SOC. EXP. BIOL. AND MED., 1933, **31**, 821.

11 Madison, R. R., PROC. SOC. EXP. BIOL. AND MED., 1934, **32**, 444, 49.

12 Tillett, W. S., *J. Bact.*, 1935, **29**, 111.

13 Hare, R., *J. Path. and Bact.*, 1935, **41**, 499.

and pneumococci from their human contacts. Irrespective of the source of these organisms, it is apparent from their numbers that they found a favorable environment in the pharynx of the monkeys. Occasional hemolytic streptococcal infections have occurred in handlers of monkeys and the findings suggest that the rhesus monkey may transmit the Group A organism to man.

We wish to express our thanks to Miss M. Olmstead of the Department of Medicine who helped us with the Lancefield grouping.

8805 P

Action of the Lytic Principle of Pneumococcus on Certain Tissue Polysaccharides.

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Avery and Cullen[1] obtained from both rough and smooth pneumococci a ferment capable of lysing heat-killed pneumococci. In unpublished experiments (Dubos) it was found that the same ferment preparation renders ineffective in rabbits the capsular type specific antigen of killed cells of virulent pneumococci.

A hydrolytic action of the ferment preparation of pneumococcus on two tissue polysaccharides is described here. This action seems to be identical with that of the same ferment on the pneumococci.

From bovine vitreous humor and from human umbilical cord two polysaccharides have been obtained in pure form, seemingly identical in composition, rotation, and general physical behavior.[2] They are composed of acetylglucosamine and glucuronic acid, the components having been shown to be present in equimolar ratio by isolation or quantitative analysis.

The 2 polysaccharide acids yielded reducing sugar following hydrolysis by the ferment obtained from pneumococcus autolysates. The hydrolysis is optimal between pH 5 and 6, and does not take place above pH 8 nor below pH 4.5. The action of the ferment on killed pneumococci has about the same pH optimum. The action on both the pneumococci and the 2 polysaccharide acids is abolished

1 Avery, O. T., and Cullen, G. E., *J. Exp. Med.*, 1923, **38**, 199.

2 Meyer, Karl, and Palmer, John W., *J. Biol. Chem.* (in press).