

TABLE II.
Effect of Reticulo-Endothelial Blockade on Dye Test.

	% 5 min.	% 30 min.
Control (4 dogs)	5-10	0
30 min. after ink injection	70-100	25-50
24 hrs. " " " " (2 dogs)	65-100	35-50
48 " " " " (1 " ")	90	45
68 " " " "	35	10
92 " " " "	15	0
120 " " " "	20	0

ground graphite suspended in acacia did not impair the ability of the liver to excrete the dye. They thought that the effect of the ink injection was produced through a hindrance in the excretion of the dye into the bile. If this is correct, splenectomy then should have no effect on the dye excretion since the spleen does not have any known rôle in bile formation. Shellong and Eisler,¹ however, obtained almost as much retention of phenoltetrachlorophthalein after splenectomy as after ink injection. We also note a slight effect of splenectomy on bromsulphalein excretion.

Our results do not agree with Rosenthal and Lillie,⁹ who found no effect on the bromsulphalein excretion after splenectomy or colloidal quartz injection in rabbits. Our findings support the opinion of Herlitz that bromsulphalein is excreted through the reticulo-endothelial system, the Kupffer cell component of which plays a very important rôle.

7049 C

Specificity of Toxin of a Non-Hemolytic Variant of a Scarlet Fever Streptococcus.

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A strain of hemolytic streptococcus, No. 1452, isolated by Jones and Little¹ from the infected udder of a cow the milk of which had caused an epidemic of scarlet fever was sent to us about 3 years ago. On receipt, this streptococcus formed large mucoid colonies resembling *Str. epidemicus* on ascites blood agar and capsules were demonstrable in India ink preparations. The toxin of this strain

⁹ Rosenthal and Lillie, *Am. J. Physiol.*, 1931, **97**, 131.

corresponded to the specific toxin of the streptococcus of scarlet fever and not to the toxin of septic sore throat.²

The culture had been kept in the refrigerator on 0.5% blood infusion agar slants. At one transplanting it was noted that the strain had lost its hemolytic and mucoid characteristics; the colonies had become small and biconvex and produced green pigment on ascites blood agar. Stained organisms showed rather short chains of Gram positive cocci, typical of streptococci.

A single mouse passage was made and from the heart's blood 2 types of colony were grown: one, green pigment-producing and non-hemolytic like the parent variant and the other identical with the original hemolytic capsule-producing, mucoid colony-forming organism. In an hemolysin test in blood broth the green producing streptococcus completely failed to lyse the blood cells.

A toxin was prepared from bacteria from each type of colony in meat infusion broth (pH 7.4, 1% peptone, 1% NaCl) by incubating 7 days at 35°C. A 1:1000 dilution of the toxin of the non-hemolytic variant in physiologic saline and a 1:1500 dilution of the toxin of the hemolytic strain gave skin reactions practically equivalent in size and intensity to those produced by the Dick skin test toxin. Generally, however, the toxin of the hemolytic strain produced a slightly larger reaction. Intradermal skin tests with 0.1 cc. of each of the diluted toxins and 0.1 cc. of Dick skin test toxin as control were made on 10 previously determined Dick positive subjects, 6 adults and 4 children. All gave red areas from 1.5 to 2 cm. in diameter with both toxins.

These subjects were then immunized with 5 doses of the Dick toxin. Six weeks after the last dose identical skin tests were made. Nine out of 10 were negative to all 3 toxins. One child remained positive to the Dick skin test toxin and the toxin of the hemolytic strain in a dilution of 1:1000. There was no skin reaction to the toxin of the non-hemolytic variant in a dilution of 1:1000. Five additional Dick positive subjects who had given positive reactions to our toxins both in a dilution of 1:1000, after similar immunization gave negative skin tests to the toxin of the non-hemolytic variant 1:1000 and the toxin of the hemolytic strain 1:1500.

Eight out of 10 Dick negative subjects gave no skin reactions to the same toxins in the same dilution. In a dilution of 1:2000 of both toxins the 9th gave negative results but the 10th continued to be faintly positive to both.

¹ Jones and Little, *J. Exp. Med.*, 1928, **47**, 957.

² Pilot, I., and Davis, D. J., *J. Inf. Dis.*, 1933, **53**, 29.

Specific scarlet fever horse antiserum, 1:50, neutralized both toxins, 1:1000, in intradermal tests made before immunization in 3 of the 4 children mentioned above. The 4th child was serum sensitive. This same horse antiserum (1:50) failed to neutralize the toxin of a septic sore throat strain of *Str. epidemicus* (1:500) in 5 children who had shown positive cutaneous reactions to sore throat toxin. Nor did it neutralize the toxin of an erysipelas strain of hemolytic streptococcus of the mucoid variety (1:500) in intradermal tests in one child who had responded positively to the erysipelas toxin.

In this work we add another variation in cultural characteristics of scarlet fever streptococcus. Previous reports indicate that scarlet fever streptococci may or may not ferment mannitol and still produce a specific toxin. Similarly scarlet fever streptococci may become mucoid and encapsulated and still produce specific toxin. In the present work specificity of toxin is retained although the strain had lost its hemolytic property.

7050 P

Loss of Pupillary Light Reflex Resulting from Lesions in the Region of the Posterior Commissure.

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Systematic exploration of the interior of the brain in the region of transition between midbrain and forebrain has enabled us to trace the central path for the pupillo-constrictor reflex in response to light. This was done with a fine needle electrode accurately placed with the aid of the Horsley-Clarke stereotaxic instrument and the points stimulated were checked by subsequent microscopic examination. These experiments, which have been fully described elsewhere,¹ showed that the impulses responsible for pupillary constriction travel from the optic tract through the superior quadrigeminal brachium and pretectal region but not through the superior colliculus. There appears to be a partial crossing in the posterior commissure and another ventral to the cerebral aqueduct.

Taking advantage of the precise information thus obtained we have been able to obliterate completely and permanently the pupillary light reflex in both eyes by placing small electrolytic lesions in the

¹ Ranson, S. W., and Magoun, H. W., *Arch. Neurol. and Psychiat.*, in press.