

Rose, Helmer and Chanutin.<sup>2</sup> Because of the variable amount of edema produced during the perfusion, it was thought desirable to calculate the results in terms of the dry weight.

In the control hearts, the total creatinine content varied from 144 to 168, with an average of 153 mg. per 100 gm. of muscle. In terms of dry weight, there were from 753 to 868, with an average of 795 mg. per 100 gm. of dried muscle.

In the hearts perfused with Ringer-Locke solution alone, the total creatinine values ranged from 100 to 151, with an average of 123 mg. per 100 gm. of moist muscle; or, from 652 to 827, with an average of 742 mg. per 100 gm. of dried muscle. In the series perfused with Ringer-Locke to which glycooll had been added, the values ranged from 103 to 159, with an average of 126 mg. per 100 gm. of moist muscle; or, from 623 to 855, with an average of 739 mg. per 100 gm. of dried muscle.

Under our experimental conditions, the addition of glycooll to the perfusion fluid had no evident influence on the total creatinine metabolism of the heart muscle. The fact which we observed, that in most instances the hearts perfused with a fluid containing glycooll beat more vigorously, and maintained a spontaneous rhythm for a longer period than did those perfused with Ringer-Locke alone, may be explained by assuming a specific stimulating action of the amino-acid. This has been suggested to us by Prof. B. M. Hendrix as the possible explanation of the effect of glycooll in diseases of striated muscle.

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### Comparison of a Rapid (Folger-Solé) Method and the Routine Loeffler's Method for Diagnosis of Diphtheria.

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Recently a paper by Solé<sup>1</sup> describing his diagnostic results in 200 cases of diphtheria, by a rapid cultural method originally suggested by Folger some 35 years ago but never published, inspired us to

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<sup>2</sup> Rose, W. C., Helmer, O. M., and Chanutin, A., *J. Biol. Chem.*, 1927, **75**, 543.

<sup>1</sup> Solé, Alphons, *Wien. Klin. Wchscht.*, 1934, **47**, 713.

apply the procedure with certain modifications to the problem as it presented itself to us at the Willard Parker Hospital. For many years we have been confronted by the difficulty of getting a sufficiently prompt and definite bacteriologic diagnosis in clinical diphtheria, or in ruling out diphtheria in carrier suspects within a reasonable length of time, by the classical Loeffler's blood serum slant method. By contrast, with the herein described rapid method we have been able to secure 95% positive cultures by the end of 4 hours.

The technique employed is extremely simple. Sterile cotton swabs are impregnated thoroughly with undiluted, unheated, horse-serum to which no preservative has been added. The swabs are then squeezed lightly against the sides of the tube to remove any surplus serum. They are removed and lightly heated over a flame to secure at least surface coagulation, and possibly as Solé says to destroy any serum antibodies. These swabs are then utilized to take the ordinary routine nose and throat cultures of the suspected case. They are placed in dry sterile tubes in the incubator and examined at the end of 2 and 4 hours by smear preparations. The slides are stained routinely by Ponder's method. They are graded as negative, suspicious or positive. In the tabulation presented here only undoubted positives are included. Too much emphasis cannot be placed upon the care with which these cultures should be taken, in order to secure satisfactory specimens. At the end of the 4-hour period, Loeffler's slants were routinely inoculated from these swabs, for isolation, control and subsequent identification by fermentation.

For convenience in interpreting our results the cases were divided into 3 groups: (1) cases of clinical diphtheria, (2) diphtheria carriers, and (3) negative controls (including streptococcic laryngitis, observation scarlatina, etc.) In respect to the first of these groups—the cases of clinical diphtheria—the results have been incorporated in Table I.

Of the 72 cultures representing 38 cases of diphtheria, 66 were positive. As may be seen from the chart, 34 or 92% of the throat cultures were positive at the end of 4 hours as compared with 28 or 76% by the Loeffler method at the end of 18 hours. Of the nose cultures only one represented a simple nasal diphtheria which accounts in part for the lower figures. The 38th case of this group was a frank clinical case of toxic diphtheria with membrane in which neither the hospital laboratory nor the Department of Health diagnostic laboratories were able to secure positive cultures. The second part of the table analyzes these figures in more detail.

In respect to the second or carrier group. We had 13 such cases

TABLE I.  
Clinical Diphtheria—38 Cases. Comparative Results—Routine and Rapid Methods.

No. of cal- tures	No. of KL cul- tures	Rapid Method		Loeffler Slant Routine Method		Comparison of Results: Loeffler's 18-Hour Slant Method vs. Rapid Method													
		Positive in		Positive in		Negative		Positive		Negative									
		2 hrs. No. %	4 hrs. No. %	18 hrs. No. %	18 hrs. No. %	Loeffler Rapid 18 hrs. No. %	Loeffler Rapid 4 hrs. No. %	Loeffler Rapid 4 hrs. No. %	Loeffler Rapid 4 hrs. No. %	Loeffler Rapid 4 hrs. No. %	Loeffler Rapid 4 hrs. No. %								
	34	13	45	17	59	27	93	11	38	18	53	8	24	2	6	9	27	51	15
Throat	38	29	78	34	92	37	100	28	76	9	24	6	16	0	0	28	74	1*	3
Total	72	42	64	51	77	64	95	39	59	27	38	14	19	2	3	37	51	6	8
Total cases	38	30	81	35	95	37	100	31	84	6	16	4	11	0	0	31	82	1*	3

\*Case had uniformly negative cultures, but clinically was considered toxic diphtheria.

†Cases of tonsillar or pharyngeal diphtheria with negative nasal cultures.

with 34 separate cultures in this series. Of the 34 cultures 29 were considered positive in the final analysis. Of these, 24 or 83% were isolated by the 4-hour method, whereas only 19 or 65% were identified as positive by the usual Loeffler's slant method at the end of 18 hours.

The particular point of interest in this group is the further fact that by this rapid method with a subplant to Loeffler's from the swab at the end of the 4-hour period, we can secure within 18 hours a practically pure culture suitable for the toxicity test.

In the third or control group there were 8 cases with 14 separate nose and throat cultures. These were uniformly negative both by the rapid method and the Loeffler method in our own hands and in the control reports from the Department of Health Laboratories, with the exception of a single nose culture reported positive by the Department of Health, but subsequently shown to be *B. hoffmanni* by fermentation reactions. This group needs no further comment.

*Summary and Conclusions.* A modified rapid cultural method for the diagnosis of clinical diphtheria is described whereby accurate reports may be rendered in over 80% of cases within 2 hours, and in 95% within 4 hours, as compared to the average of 83% accuracy at the end of 18 hours by the usual Loeffler's slant method. The method seems to us to be of great value in establishing early diagnosis and treatment in the disease. It further provides for the securing of pure cultures within 18 hours for toxicity tests, thus reducing the necessary period of isolation and possible exposure to true diphtheria for non-toxic carriers, a matter of grave importance, to patient and hospital alike. We feel that the method should supplant the older, more cumbersome procedure in all contagious hospitals, and that it should be utilized very largely by health departments both in their diagnostic and release cultures as an adjunct to their present methods. We are convinced that by earlier diagnosis and treatment the mortality of the disease should be further reduced. We believe that the carrier problem is materially improved with appreciable effective saving of hospital days to both the individual and the institution.