

*S. viridans*), which are apparently equally specific for the fibrins of certain domestic animals. A summary of our streptococcus strains which dissolve fibrins from lower animals is given in Table I.

## 7717 C

### Enzyme-Concentration Method of Titrating Bacterial Fibrinolysins.\*

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Since but 17% of all local strains of *Streptococcus hemolyticus* originally isolated from superficial infections of man are demonstrably thrombolytic<sup>1</sup> by the Tillett-Garner plasma-clot technic,<sup>2</sup> we have retitrated these strains by their more delicate serum-free fibrin-clot method. This retitration has raised the percentage of demonstrably fibrinolytic strains to 25%.

It is of basic interest to determine whether or not the remaining 75% have or have not a demonstrable antihuman fibrinolytic capacity. To determine this we have again retitrated all local strains, using a modification of the Tillett-Garner enzyme-concentration method.

Since at least 75% of the fibrinolysin in a 24-hour broth culture of *S. hemolyticus* is usually lost as a result of filtration, enzyme-concentrates were prepared from unfiltered broth cultures. To prepare such a concentrate, 20 cc. unfiltered 24-hour veal-infusion broth culture is added to 60 cc. 96% alcohol, both culture and alcohol being ice-cold at the time of mixing. The resulting mixture is allowed to stand at refrigerator temperature for at least an hour. The precipitate is then collected by centrifugation, and rapidly dried in a vacuum desiccator.

To make the fibrinolytic test, the crude precipitate is suspended in 1 cc. buffered salt-solution and freed from undissolved residue by centrifugation. The resulting centrifugate usually has a fibrinolytic titer at least 20 times that of the original broth-culture, which is generally equivalent to at least 80 times that of the broth-filtrate.

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<sup>1</sup> Madison, R. R., PROC. SOC. EXP. BIOL. AND MED., 1934, **31**, 1018.

<sup>2</sup> Tillett, W. S., and Garner, R. L., J. EXP. MED., 1933, **58**, 485.

Retitration of 123 local strains of *S. hemolyticus* originally isolated from superficial infections of human beings has raised the percentage of recognizable fibrinolytic strains to 35%. About one-eighth of all apparently non-fibrinolytic strains of *S. hemolyticus* from infections in man, therefore, have a demonstrable fibrinolytic potential, recognizable by the 20-fold enzyme-concentration technic.

By the same enzyme-concentration technique the percentage of strains of *S. hemolyticus* from infections of lower animals positively lytic for human fibrin has been raised from 7% to 22%. None of the 40 local strains of *S. viridans*, however, has shown a demonstrable anti-human fibrinolytic capacity.

## 7718 C

### A Modified Method for the Estimation of Tryptophane.\*

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This paper deals with a modification of May and Rose's<sup>1</sup> method for the determination of tryptophane. In their method the protein is dissolved in HCl which contains Ehrlich's reagent, incubated at 35° for 24 hours, and then allowed to stand for an additional 40 hours at room temperature. The blue color which is thus produced is matched against a similar color obtained by treating casein in an identical manner.

In this procedure such factors as the particular preparation of casein used for the standard and the room temperature may be uncontrolled variables. May and Rose assumed the tryptophane content of casein to be 1.5%. It is based on the data of Hopkins and Cole.<sup>2</sup> Other values have been reported.<sup>3</sup> The uncertainty as to the tryptophane content of casein makes this protein of questionable value as a standard.

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<sup>1</sup> May, C. E., and Rose, E. R., *J. Biol. Chem.*, 1922, **54**, 213.

<sup>2</sup> Hopkins, F. G., and Cole, S. W., *J. Physiol.*, 1902, **27**, 418.

<sup>3</sup> Herzfeld, E., *Biochem. Z.*, 1913, **56**, 258, 0.51%; Thomas, P., *Ann. Inst. Pasteur*, 1920, **34**, 701, 1.7-1.8%; Fürth, O., and Nobel, E., *Biochem. Z.*, 1920, **109**, 103, 2.02%; Folin, O., and Ciocalteau, V., *J. Biol. Chem.*, 1927, **73**, 627, 1.4%; Holm, G. E., and Greenbank, G. R., *J. Am. Chem. Soc.*, 1923, **45**, 1788, 2.24%.