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Effect of Bacteriophage on Strepto-Fibrinolysin Production.*

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To test the effects of bacteriophage on the production of fibrinolysin by hemolytic streptococci, 100 cc. flasks of 0.5% glucose veal-infusion broth (pH 7.4) were inoculated with 0.1 cc. of a 24-hour broth culture of *Streptococcus hemolyticus* and an amount of bac-

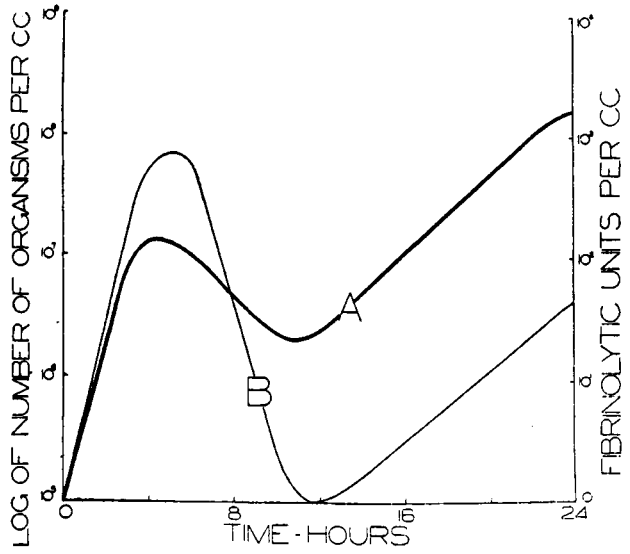

FIG. 1.

Phage-acceleration of Fibrinolysin Production.

100 cc. veal infusion broth, plus 1 cc. bacteriophage (homologous 'phage-lysed filtrate), inoculated with 0.1 cc. 24-hour broth culture of *S. hemolyticus*, incubated at 37°C with constant stirring.

A, fibrinolysin production. Composite data from 6 titrations with 2 different bacterium-phage combinations. For titration technic and definition of fibrinolytic unit see previous paper.[1]

B, parallel changes in total bacterial population as determined by the Petroff-Hauser counting chamber.

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1 Madison, R. R., and Taranik, Jeannette D., PROC. SOC. EXP. BIOL. AND MED., 1937, **36**, 1.

teriophage ('phage-lysed homologous streptococcus filtrate) demonstrably sufficient to produce lysis was added to each flask. The flasks were incubated at 37°C. with constant stirring and samples were withdrawn at regular intervals for total population counts and fibrinolysin titrations. Composite data from 6 such counts and titrations with 2 different streptococcus-bacteriophage combinations are recorded in Fig. 1.

From this figure it is seen that the initial increases in fibrinolytic titer are roughly parallel with the logarithmic increase in bacterial population. Bacterial lysis is accompanied by a partial (and in some cases nearly complete) destruction (or binding) of the fibrinolytic enzyme. The beginning of a terminal overgrowth with phage-resistant variants, however, is accompanied by a renewed and greatly accentuated formation of fibrinolysin. The fibrinolytic titer at the 24-hour stage of the terminal overgrowth is at least 10 times that of the maximum initial titer with the parent phage-susceptible strain.

In order to confirm these results, phage-resistant variants were isolated in pure culture from such over-growths, and 0.1 cc. of a 24-hour sub-culture of each phage-resistant variant was inoculated into 100 cc. flasks of veal-infusion broth, control flasks being inoculated with an equal volume of a 24-hour broth culture of the homo-

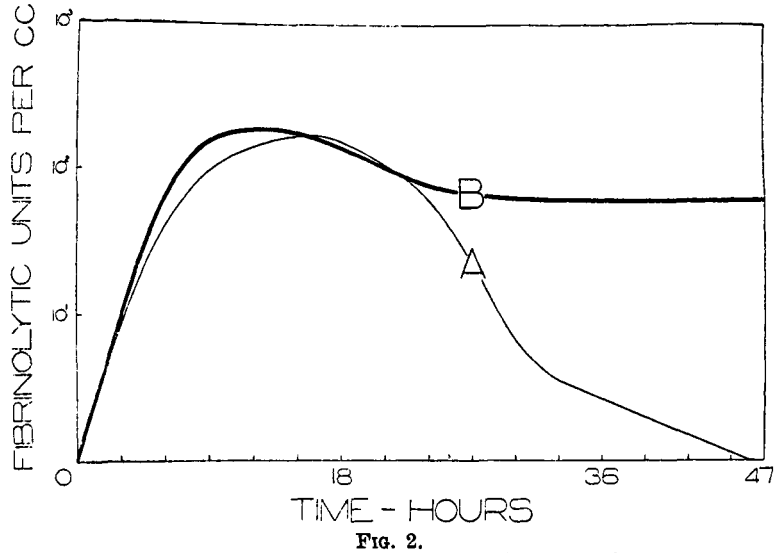

FIG. 2.

Fibrinolysin Production by Phage-resistant Variants.

100 cc. veal infusion broth inoculated with 0.1 cc. 24-hour broth culture of *S. hemolyticus*, incubated 37°C with constant stirring.

A, fibrinolysin production and subsequent destruction by control homologous phage-susceptible parent cultures. Composite data from 6 titrations.

B, fibrinolysin production by homologous phage-resistant variants. Composite data from 6 titrations.

logous, phage-susceptible parent strain. A typical comparison of fibrinolysin production by parent strain and phage-resistant variant is recorded in Fig. 2. Within the limits of the experimental error the population changes in both flasks were identical throughout the tests so are not recorded in the figure.

From Fig. 2 it is seen that while both flasks produced fibrinolysins at the same rate during the initial stages of population increase, the phage-susceptible parent strains rapidly destroyed or otherwise inactivated the fibrinolytic enzyme after reaching maximum titer. This destruction, however, did not take place (or was counterbalanced by continued fibrinolysin production) with the homologous 'phage-resistant variants.

If as a result of local phage injections all local streptococci could be destroyed, bacteriophage therapy would presumably aid in preventing the spread of streptococcus infections. The danger of the local production of phage-resistant variants with heightened fibrinolytic function and presumably heightened virulence,[2] however, would seem to contraindicate phage therapy in local streptococcus infections. This conclusion is in line with the experimental evidence reported by Bronfenbrenner and Sulkin[3] in their study of the local therapy in experimental staphylococcus infections in rabbits.

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Influence of Adrenalectomy and Choline on the Fat Content of Regenerating Liver During Fasting.

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Removal of the adrenal glands prevents the deposition of fat in the liver due to phosphorus poisoning,[1] that following the administration of anterior pituitary extracts,[2] that which generally accompanies total pancreatectomy,[3] and also interferes in other cases in which fat may wander from the fat depots to the liver.[4, 5] Choline prevents the fatty liver which occurs when a diet low in choline or

2 Fraser, F. H., and Madison, R. R., *Ibid.*, 1935, **33**, 307.

3 Bronfenbrenner, J., and Sulkin, S. E., *Ibid.*, 1935, **32**, 1418.

1 Verzár, F., and Laszt, L., *Biochem. Z.*, 1936, **288**, 356.

2 MacKay, E. M., and Barnes, R. H., *Am. J. Physiol.*, 1937, **118**, 525.

3 Long, C. N. H., and Lukens, F. D. W., *Am. J. Physiol.*, 1936, **116**, 96.

4 Laszt, L., and Verzár, F., *Biochem. Z.*, 1936, **285**, 356.

5 MacKay, E. M., *Am. J. Physiol.*, 1937, **120**, 361.