

due to one specific type of Group B. This conclusion coincides well with the observation of Stableforth.<sup>2</sup>

The total lack of reaction in the case of Herd II is very interesting, and does indicate that there is some correlation between the occurrence of a given type of *Streptococcus mastitidis* in the herd and the presence of antibodies in the sera of that herd.

It is not the purpose of this work to advocate the routine use of serological methods in the diagnosis of mastitis. Considerable work would be necessary before one would be justified in drawing conclusions as to the correlation between mastitis and the presence of agglutinating antibodies. Such data would necessitate a survey such as that conducted by Stableforth in order that we may attain some knowledge of the number of types of *Streptococcus mastitidis* encountered in the United States. Furthermore complete studies should be made on herds in order that we may draw conclusions regarding the correlation between mastitis as shown by our more common tests and the agglutinative response. Until such data have been collected the serological reactions of cow serum can only be of theoretical interest.

*Conclusions.* Cow sera may contain streptococcal antibodies which appear to be type-specific. There is evidence of a correlation between the presence of *Streptococcus mastitidis* in the herd and the occurrence of agglutinating antibodies in the serum of the individual cows. It appears probable that infection in an individual herd may be due to a specific type under Group B.

### 10303 P

#### **Sputum Studies in Lobar Pneumonia. Extra-Cellular Encapsulated Pneumococci.**

ARTHUR W. FRISCH. (Introduced by W. O. Nelson.)

*From Wayne University College of Medicine, Detroit, Michigan.*

In a previous report concerning rusty sputum from cases of lobar pneumonia, 3 types of phagocytosis and the effect of serum therapy were described.<sup>1</sup> The present communication deals with the number of extracellular capsulated pneumococci in rusty sputum as an index

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<sup>1</sup> Frisch, A. W., PROC. SOC. EXP. BIOL. AND MED., 1938, **39**, 473.

of the severity of the pneumonia. No attempt will be made at present to relate the counts to therapy.

Seventy-eight roentgenographically proved cases of pneumonia with the following type distribution have been studied: 20 cases of type I, 30 type II, 3 type III, 2 type V, 1 type VI, 11 type VII, 3 type VIII, 1 type IX, 3 type XII, 1 type XVI, and 2 mixed. Thin smears were made directly from representative rusty portions of the sputum and treated with Wright's blood stain for 3 to 7 minutes. Without the addition of buffer the stain was flooded off with water and the slides were air dried. The capsules of the pneumococci usually did not stain but were readily visualized as clear or occasionally as pink zones around the dark blue diplococci. The mucin stained pale pink and the leukocytes were differentiated as on a blood smear. At first, samples of rusty sputum were examined at 4- to 6-hour intervals throughout the acute stages of the disease. However, specimens taken every 12 hours were found to be equally satisfactory and most of the cases were followed in this manner.

During the study it was found that rusty sputum, relatively free of organisms other than pneumococci, gave the most consistent results. Frankly purulent specimens contained large numbers of capsulated pneumococci which had no relation to the clinical course or outcome of the pneumonia. Mucoïd sputa early in the disease are now under investigation.

The number of extracellular capsulated diplococci per oil immersion field varied considerably on different parts of the same slide depending upon the thickness of the smear. Approximately 50 representative fields were counted at first and the average number per field was estimated considering each pair of cocci as one organism. Counts of 2 slides made from different portions of each specimen of sputum and determined separately by 2 individuals usually agreed within 10%. At present, fairly accurate checks are obtained by counting only 10 fields. Because one is dealing with individual samples of sputum which probably originate from various portions of the infected lung, one might expect that the number of free pneumococci in specimens obtained at frequent intervals would fluctuate sufficiently to invalidate the results. Actually the counts from sputum to sputum in the same patient were surprisingly constant and the increase or decrease in the number of pneumococci per oil immersion field was progressive rather than sudden.

In Table I under 4 subdivisions (A, B, C, D) are listed the highest number of extracellular capsulated pneumococci per oil immersion field obtained in the sputum of the patients at any time during the

TABLE I.

	Group A 0 to 10	Group B 11 to 30	Group C 31 to 65	Group D Over 65
No. of cases	31	22	10	15
Positive blood culture	2	9	7	11
Leukopenia (less than 10,000)	1	3	5	7
More than one lobe involved	9	5	4	11
Number of deaths	1	2	2	13

acute stages of the disease. These counts were compared with data\* of known prognostic significance from the same cases.

From the table it may be noted that the incidence of unfavorable prognostic signs such as positive blood culture and leukopenia was roughly proportional to the number of extracellular capsulated pneumococci per oil immersion field. The high mortality in those cases with more than 65 free pneumococci per field (Group D) coincided with the greater incidence of positive blood cultures and leukopenias. The single death in Group A was, in the author's opinion, due largely to asphyxia from thick purulent sputum and occurred in a case of type II pneumonia with a negative blood culture and a leukocytosis. The sputum at the time of death showed an occasional type II pneumococcus and myriads of influenza bacilli. The two deaths in Group B and one in Group C occurred in patients with type III infections and were associated with leukopenia and positive blood cultures. In these and other fatal cases of type III pneumonia the count of extracellular pneumococci in the sputum and in the lung at autopsy has been consistently low. The other death in Group C occurred during aspiration of an interlobar empyema in a patient convalescing from a severe type II pneumonia. The deaths in group D were, for the most part, fulminating pneumonias in patients who either came to the hospital moribund or in whom the therapy seemed to have no effect. Of the 2 type I cases who survived in Group D, one showed a high sputum count in only one specimen and the second recovered after a difficult infection which was complicated by empyema, gluteal abscess, and a panophthalmitis which necessitated the removal of one eye. Type I pneumococci were recovered from all the lesions.

If it can be shown that the extracellular capsulated pneumococci in rusty sputum reflects the number present in those areas of the lung where the pneumonic process is advancing, then the method and the results presented in this preliminary report are encour-

\* Data obtained through the courtesy of Doctors A. E. Price and G. B. Myers in charge of the Commonwealth Fund pneumonia study at Receiving Hospital.

aging as a means of evaluating the severity of a given case of pneumonia on admission of the patient to the hospital and during the course of the disease. Further study of a large series of cases will be necessary to establish this point.

## 10304

## Serial Production of Phage from Intracellular Phage Precursor.\*

A. P. KRUEGER AND E. J. SCRIBNER.

*From the Department of Bacteriology, University of California.*

Recently experimental evidence was advanced indicating that phage production occurs according to the reaction: inactive phage precursor + phage  $\rightarrow$  phage.<sup>1</sup> The particular point of interest in this concept is the dissociation of phage production from what was previously held to be an essential conditioning factor, namely, bacterial growth. Furthermore, there exist authentic models for such a reaction in the autocatalytic transformation of inactive enzyme precursors into the active enzymes when the precursors are brought in contact with small amounts of active enzyme.

To summarize the experimental data it was found by Krueger and Baldwin<sup>1</sup> that under certain conditions cell-free ultrafiltrates of staphylococci added to solutions of bacteriophage resulted in 100% increases in phage activity. In studying the effect of salts on the phage-bacterium reaction it was found that the presence of sodium chloride<sup>2</sup> or sodium sulphate<sup>3</sup> during the phage-bacterium reaction produced a prelytic plateau in the bacterial growth curve for as much as 0.8 hour during which time phage production continued at a normal rate despite the absence of cellular reproduction. Later<sup>4</sup> experiments were reported in which phage production was dissociated from bacterial growth by means of changes in temperature and pH of the medium. Unfortunately these data do not offer conclusive proof that the phage precursor-phage reaction is the normal mode of phage production largely because the yields of pre-

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<sup>1</sup> Krueger, A. P., and Baldwin, D. M., *Proc. Soc. Exp. Biol. and Med.*, 1937, **37**, 393.

<sup>2</sup> Krueger, A. P., and Scribner, E. J., *J. Gen. Physiol.*, 1937, **21**, 1.

<sup>3</sup> Krueger, A. P., and Strietmann, W. L., *J. Gen. Physiol.*, 1938.

<sup>4</sup> Krueger, A. P., and Fong, J., *J. Gen. Physiol.*, 1937, **21**, 137.