

which consists in the intrabronchial injection of 0.05 cc. to 0.5 cc. of an 18 hour culture of *Pneumococcus* Types I or II, suspended in a viscous starch-broth mixture. For this purpose a radio-opaque catheter is inserted under the fluoroscope and 0.5 cc. to 1 cc. of this pneumococcus starch suspension instilled into a small bronchus as near the periphery of the lung as possible. As a rule, within 24 hours typical lobar consolidation of the injected lobe has occurred as evidenced by x-ray and physical findings. The disease runs a febrile course of 3 to 7 days; the pneumonic lesion either remains localized in one lobe or spreads from lobe to lobe. This experimental disease resembles the natural disease in humans in the manner of the spread of the lesion, the localization of the process (pneumococci do not usually invade the blood stream), the immune response, the abrupt termination of the disease by crisis, lysis, or death and the rapid regression of the process after recovery. Animals were killed at different stages during the disease from one hour after injection of the infecting dose to one or 2 days following recovery. The lesion was found to spread evenly and contiguously thru the lobe, the line of the advancing process usually being sharply demarcated from the normal tissue. With the evolution of the disease the lesion progressed through the different stages observed in the human pneumonic lung from an initial marked congestion to red hepatization and finally to a modified gray hepatization. The microscopic pathology was that of a lobar pneumonia from the beginning and in its general characteristics resembled the picture of the human disease. Certain differences from the human pneumonic lungs were observed. These were the greater degree of blood vessel engorgement seen all thru the disease, the smaller amount of fibrin, and the more rapid decrease in size of the resolving lung.

5069

**On the Production of Variant Colonies by Certain of the
Intestinal Bacteria.**

STEWART A. KOSER.

From the Department of Hygiene and Bacteriology, the University of Chicago.

Much of the literature on microbic dissociation has emphasized the occurrence of rough (R) colonies, in contrast to the smooth (S) form, with, on the whole, relatively little mention of other

colony types. In contrast to this, it has been the writer's experience that frequently many variant colonies of different character are encountered.

The results presented in the accompanying table were obtained in connection with a study of factors which might stimulate the process of dissociation. They are typical of many other similar experiments and illustrate the changes in colony form which were found in cultures of certain of the colon-paratyphoid-dysentery organisms. Smooth forms, previously carried through a series of colony isolations, were used for inoculation of the broth or peptone media. The appearance of variant colonies was followed by streaking agar plates of standard composition at intervals. The changes thus represent those occurring in "ageing" cultures.

The change from S to R appeared to be a gradual one and was not accomplished by distinct or abrupt changes from one colony type to another. All gradations in colony form from the typical white homogeneous colonies with even margins, characteristic of the S form, to the flat, dull, dry-looking, coarsely-granular and irregular colonies of the R form were found. Colonies appearing

TABLE I.
Different Colony Forms Obtained from Ageing Broth Cultures.

Time, Days	<i>Bact. coli</i> Broth 37°C.	<i>Bact. coli</i> 5% Pep- tone 37°C.	<i>Bact. aertrycke</i> Broth 25°C.	<i>Bact. aertrycke</i> Broth 37°C.	<i>Bact. aertrycke</i> 10% Pep- tone 25°C.	<i>Bact. dysente- riae</i> Sonne 10% Peptone 37°C.
1	100 S*	100 S	100 S	100 S	100 S	100 S
2			100 S	100 S	100 S	100 S
4	100 S	88 S 10 I 2 R	95 S 5 †	80 S 19 † 1 R	99 S 1 I	90 S 10 I
7	65 S 30 I 5 R	60 S 39 I 1 R	86 S 8 † 6 R	35 S 8 I 22 † 35 R	70 S 30 I	40 S 57 I 3 R
10			96 S 4 †	27 S 68 † 5 R	75 S 25 I	
14	85 S 15 I	70 S 15 I 15 R			70 S 30 I	35 S 62 I 3 R
28	80 S 20 I	95 S 5 I	73 S 27 †		70 S 30 I	10 S 40 I 50 R

* Results are expressed in percentages of the different colony types.

† Small adherent, raised, and slightly convoluted colonies. Impossible to classify as either smooth, intermediate or rough forms.

to fall between these two extremes are referred to as intermediate (I) forms in the table. It will be seen that in ageing cultures such colonies were usually present in large numbers and often far outnumbered the true R forms. At times the change went no farther than the intermediate stage and true rough colonies were never seen, though there was nevertheless a decided departure from the typical smooth form.

It should be pointed out that there may be some doubt as to whether all of the colonies referred to as intermediate really represent transition forms whose destination is the R stage, or whether they may not be simply variants from the S type whose ultimate goal, if any, is not the R form.

At times other variant colonies appeared whose characteristics were so obviously different from those of the S, I or R forms that they could not be classed with them. Thus, from older cultures of *Bact. aertrycke* very small, sticky, raised and slightly convoluted colonies were at times obtained. They are designated in the accompanying table by a question mark. When transferred to agar slants and carried through successive daily transfers on agar at 37° C for periods of 30 to 45 days, they gradually returned for the most part to the S type. Also, when similar well-isolated colonies were allowed to age on agar plates they frequently developed a marginal outcropping of S growth. The individual cells comprising these colonies possessed a capsule-like sheath which surrounded the cell and at times appeared to tie together bunches of cells. Otherwise, there was little difference between them and the cells from smooth colonies. On a few occasions large watery mucoid colonies were found.

In Arkwright's communication,¹ which may be said to be responsible in large measure for the recent renewed interest in the subject of variation, he stated "The variety of colonies met with is so great and the differences between them often so indefinite that attention has purposely been directed only to the more obvious characters . . .". Some of the more recent workers who have followed the interesting though elusive changes included under the term dissociation, have likewise mentioned the occurrence of intermediate or other colony forms, but the study of these forms has usually been subordinated to that of the more striking R type. The purpose of this report is to call attention especially to the large number of intermediate forms and other variant colonies which may be encountered under certain conditions.

¹ *J. Path. and Bact.*, 1921, xxiv, 36.