culture of a mouse-virulent anthrax strain. He thought at first that this might be an anthrax mutant, but later decided that it was a *Bacillus megaterium* intermediate (Guex-Holzer and Tomcsik, J. Gen. Microbiol., 14, 14–25, 1956). We examined the culture and are in full agreement with this view.

This leaves the paper of Brown et al., (J. Bacteriol., 69, 590–602, 1955) as the only recent and circumstantial description of motile anthrax mutants. These workers exposed anthrax cultures to a bacteriophage obtained from a virulent, lysogenic *B. anthracis* (strain Ohio). None of the bacteriophage treated strains—and only these nine—regularly produced motile variants which Brown et al. found biochemically and pathologically indistinguishable from *B. anthracis*. A careful study of the paper by Brown et al. revealed no obviously uncontrolled source of error. However, in their assessment of pathogenicity, the authors may have been unduly influenced by Nordberg (Nord. Vet. Med., 5, 915–924, 1953), who has apparently found difficulty in distinguishing experimental anthrax infections from deaths following the injection of broth cultures of *B. cereus* (Chu, J. Gen. Microbiol., 3, 255–273, 1949).

Dr. Cherry very kindly sent us 6 of the motile transformed strains; 4 proved to be *B. subtilis*, and 2 *B. cereus*. Our findings were reported to Dr. Cherry in September, 1955. He re-examined his stocks of motile *B. anthracis* and agreed that these were now contaminated and that no trace of motile anthrax organisms remained. The matter was left in abeyance while Dr. Cherry tried to repeat the earlier work. These attempts have failed (Cherry, personal communication), and we feel that it should be made clear that the existence of motile *B. anthracis* is debatable, and that the ingenious genetic mechanism postulated for the derivation of these strains may be insecurely based.

In both the introduction and discussion to their paper Brown et al. placed considerable weight on the induction of motility and capsulation in an avirulent anthrax strain by Manninger and Nógrádi. However, the motile capsulated strain obtained was shown to be *B. subtilis* (Tomesik, personal communication). Brown et al. also discussed at some length the derivation by Tomcsik (1949) of anthrax strains able to produce capsules under ordinary atmospheric conditions from strains which were uncapsulated under these conditions. Tomesik actually stated that on the basis of experimental evidence the question of a transformation reaction has to be left open, and emphasized that he was unable to induce capsulation in a completely avirulent—that is to say, genotypically uncapsulated—anthrax strain.

It must therefore be emphasized that no transformation of *B. anthracis* from a genotypically noncapsulated to a capsulated state has yet been effected. Arguments are therefore invalid if based on the impression that such transformations have been demonstrated.

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**ADAPTATION OF THE MEMBRANE FILTER TECHNIQUE TO THE RECOVERY OF COAGULASE POSITIVE *STAPHYLOCOCCUS AUREUS* FROM HUMAN SALIVA**

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The use of the membrane filter was initiated by Goetz and Tsuneisuki (J. Am. Water Works Assoc., 43, 943, 1951) in bacteriological water analysis. It is presently being applied principally for the detection of *Escherichia coli*. There has been no report on the application of the membrane filter technique in the isolation and diagnosis of coagulase positive strains of *Staphylococcus aureus*, particularly from samples of saliva.

This study was suggested in conjunction with a current investigation of the efficacy of direct inoculation of saliva onto glycine tellurite agar (Difco) as compared to Chapman Stone medium in the recovery of coagulase positive *S. aureus* strains. It was observed that the specificity of
glycine tellurite agar and Chapman Stone medium for coagulase positive strains of *S. aureus* was 98.5 per cent and 32 per cent, respectively.

“Millipore” filters (MF membranes, Millipore Filter Corp.) were used in conjunction with S & S membrane filter holders (Schleicher and Schuell). The S & S “Coli 5” apparatus was sterilized by autoclaving at 15 lb pressure, 121 C for 15 min. The MF filters were autoclaved for 10 min at (12 lb) 118 C. Samples of saliva were shaken mechanically for 10 min; 1 ml of saliva was added to 9 ml of beef heart infusion broth. The sample being tested was allowed to pass through the membrane, and the container rinsed twice with about 10 ml of sterile distilled water. After filtration, the membrane filter was placed on the surface of a freshly poured glycine tellurite agar (Difco) plate. After each saliva sample the apparatus was rinsed with sterile distilled water. The petri dishes were observed with the naked eye and under the dissecting microscope after incubation at 37 C for 24 and 48 hr. One to five colonies from each filter were chosen for subculture and tested for coagulase activity with human citrated plasma. Simultaneously, a sterile swab was inserted into the original saliva samples, drained, and streaked onto the surface of glycine tellurite agar plates. This inoculum represented 0.1 ml. Total count was made after 24 and 48 hr by multiplying the number of isolated colonies by 10.

The results are tabulated in table 1. The glycine tellurite agar was as selective for saliva as for other body fluids in detecting coagulase positive strains of *S. aureus*. The membrane filter technique proved superior to the direct inoculating technique from several standpoints: (1) a higher count was obtained, irrespective of the dilution factor; (2) counts were obtained when no organisms were recovered by the conventional method; (3) failures in isolating and detecting organisms present was minimal; and (4) time of incubation was reduced to 24 hr for ascertaining the presence of coagulase positive staphylococci.

**TABLE 1**

Coagulase positive *Staphylococcus aureus* on glycine tellurite agar

<table>
<thead>
<tr>
<th></th>
<th>Frequency by Membrane Filter</th>
<th>Frequency by Direct Swab</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total specimens tested</td>
<td>1065</td>
<td>1065</td>
</tr>
<tr>
<td>Zero count</td>
<td>425</td>
<td>425</td>
</tr>
<tr>
<td>Membrane filter count greater than direct swab</td>
<td>546</td>
<td>89</td>
</tr>
<tr>
<td>Direct swab count greater than membrane filter</td>
<td></td>
<td>5</td>
</tr>
<tr>
<td>Similar counts</td>
<td>5</td>
<td></td>
</tr>
<tr>
<td>Membrane filter count but zero count from direct swab</td>
<td>318</td>
<td>5</td>
</tr>
<tr>
<td>Increased sensitivity by membrane filter technique</td>
<td></td>
<td>30%</td>
</tr>
</tbody>
</table>

**PROTOPLAST FORMATION BY MASS ADSORPTION OF INACTIVE BACTERIOPHAGE**

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The phenomenon of “lysis from without” described by Delbruck (J. Gen. Physiol., 23, 643, 1940) takes place after the rapid absorption of a great number of either active or inactive bacteriophage particles on a bacterial cell. This lysis is presumed to result from damage which occurs at the cell surface, since no active phage is produced and the cell does not demonstrate the cytochemical differences evident after phage infection (Luria and Human, J. Bacteriol., 64, 557, 1952). The release mechanism for the transfer of phage DNA into susceptible bacteria remains obscure. However, the absorption of active or inactive phage on isolated cell walls of *Escherichia coli* strain B has been shown to be accompanied by the action of enzyme(s) from the phage (Brown and Kozloff, J. Biol. Chem., 225, 1, 1957).