

## NOTES

### THE USE OF POTASSIUM TELLURITE, SODIUM AZIDE, AND ACETIC ACID IN A SELECTIVE MEDIUM FOR THE ISOLATION OF *LISTERIA MONOCYTOGENES*<sup>1</sup>

M. L. GRAY,<sup>2</sup> H. J. STAFSETH,<sup>3</sup> AND FRANK THORP, JR.<sup>2</sup>

*Michigan Agricultural Experiment Station, East Lansing, Michigan*

Received for publication December 12, 1949

In attempting to isolate a specific microorganism from contaminated material, the advantages of a satisfactory differential or enrichment medium are obvious. Numerous investigations have established that potassium tellurite, sodium azide, and acetic acid are effective for depressing gram-negative bacterial flora. These substances were incorporated with various media for isolation studies of *Listeria monocytogenes*.

Pure cultures of *L. monocytogenes* either alone or in mixture with both gram-positive and gram-negative bacteria were streaked on tryptose agar, pH 7.4, containing sodium azide in concentrations ranging from 0.2 to 0.03 per cent and potassium tellurite in concentrations from 0.1 to 0.01 per cent; tryptose agar was used as a control. It was observed that sodium azide would have no application in the isolation of strains of *Listeria*, as these organisms are completely suppressed in concentrations as low as 0.03 per cent. Potassium tellurite, however, showed considerable selectivity. Growth of *Listeria* was not greatly suppressed in 0.1 per cent concentrations, and growth was uninhibited at a concentration of 0.05 per cent. These concentrations were sufficiently high to suppress most gram-negative bacteria. However, micrococci and streptococci also grew freely at the higher concentrations. A dissecting microscope (Gray *et al.*: *J. Bact.*, **55**, 471, 1948) was employed at this point. The *Listeria* colonies appeared black, as do all colonies on this medium, but the characteristic green color was evident at the periphery of the colony. The micrococci were pinkish yellow at the periphery, intensely black at the center, and extremely glossy in appearance. The streptococci were smaller, pinkish gray in color, with a dull surface.

A less artificial method of determining the effectiveness of this medium for the isolation of *Listeria* consisted of swabbing the nasal passages of supposedly normal sheep. The swabs were placed in nutrient broth, and three drops of a 24-hour broth culture of *Listeria* were added. The tubes were incubated from 4 to 8 hours at 37 C and plated on potassium tellurite plates and tryptose agar plates. Identification of colonies of *Listeria* was invariably impossible on the tryptose agar, but they were found without difficulty on the potassium tellurite agar.

<sup>1</sup> Approved by the Director of the Michigan Agricultural Experiment Station as Journal Article 1091.

<sup>2</sup> Department of Animal Pathology.

<sup>3</sup> Department of Bacteriology and Public Health.

Excellent results were obtained when 0.05 per cent potassium tellurite was added to plain nutrient broth and used as an enrichment medium. The materials to be cultured, nasal swabs, straw, feces, etc., were added directly to the broth and incubated 6 to 24 hours at 37 C. A loopful of this material was then plated on plain tryptose agar and incubated for 20 to 24 hours at 37 C. Examination of the plates with the aid of the dissecting microscope revealed the absence or presence of *Listeria*. With this method nearly pure cultures of *Listeria* could be obtained providing the number of micrococci in the material was not too great. With this technique it was possible to isolate *Listeria* in nearly pure culture from the feces of a rabbit that for 21 days received a broth culture of *Listeria* as the sole source of fluid intake.

Acetic acid added to deep brain medium to make concentrations ranging from 0.1 to 1 per cent also proved effective in the higher levels when exposed to conditions similar to those previously described.

## A MOTILE LACTOBACILLUS FROM THE CECAL FECES OF TURKEYS

A. P. HARRISON, JR., AND P. ARNE HANSEN

*Department of Bacteriology and the Livestock Sanitary Service Laboratory, University of Maryland, College Park, Maryland*

Received for publication December 22, 1949

Although motility has been reported in a few strains of *Streptococcus*, the family *Lactobacteriaceae* as a whole is considered as consisting of nonmotile bacteria. The genus *Lactobacillus*, particularly, was always thought to contain nonmotile species only.

During a study of the bacterial flora of the cecal feces of turkeys, a gram-positive rod was isolated which shows the typical behavior of a *Lactobacillus* in all respects except that it possesses active motility. The presence of peritrichous flagella is demonstrated in the accompanying photomicrograph (figure 1). Furthermore, an electron micrograph<sup>1</sup> (figure 2) has been prepared that seems to exclude the contention that the flagella in figure 1 could possibly be staining artifacts. Numerous parallel isolations have given strains that in cultural characters agree with the motile strain but are typical nonmotile lactobacilli (of the species *L. plantarum*).

The motile strain has never been observed to form spores; it does not reduce nitrate or produce catalase. It is a facultative anaerobe, showing more surface growth when incubated under anaerobic than under aerobic conditions. In tomato juice glucose broth the rod averages 0.9 by 4  $\mu$ , although some very long cells may be encountered. It occurs singly or in pairs; long chains are never ob-

<sup>1</sup> We are indebted to Major R. L. Reagan, Department of Veterinary Science, University of Maryland, for the preparation of the electron micrograph.