Studies on the Smut of Johnson Grass. 
II. Further Studies on Sporidial Cultures

WAYNE H. SILVER, Southeastern State College, Durant

In the first paper of this series (1) it appeared that variation in the smut of Johnson grass did occur, and that such variation could be detected by culture of sporidia. To further test this, it was thought necessary to collect over a wider area. Five collections were chosen as coming from the periphery of the geographical area covered. S-1 was obtained from the corner of 7th Street and Willow in Durant, the same location from which came most of the collections previously described; S-5, from about five miles west of the center of Durant along highway 70; S-6, from the back yard of Oklahoma Presbyterian College; S-7, from about two miles east of Durant along old highway 70; and S-8, from the roadside in front of the Woodlawn Country Club south of Denison, Texas.

The same general technics of culture and transfer were followed as reported in the previous paper (1). Modified Koser's citrate broth was again used, with KNO₃ substituted for Na(NH₄)₂HPO₄ and with various carbon sources in place of the citrate. These carbon sources included the monosaccharides, glucose, mannose, fructose, and sorbose; the disaccharides, sucrose, maltose, lactose, and cellobiose; the polyhydric alcohols, glycerol, mannitol, dulcitol, and sorbitol; and the following acids or their sodium salts: formic, acetic, propionic, butyric, succinic, citric, pyruvic, lactic, and tartaric. Previous results made it appear that the nitrate medium might be a better differential medium than the sodium ammonium phosphate in the original Koser's medium.

Sporidia of each of the five accessions were inoculated individually into solutions of the various carbon sources with the minerals, minerals alone, and plain water. All five accessions behaved similarly on all the carbon sources. Fructose, glucose, sucrose, and maltose all allowed good growth, with sorbose and cellobiose slightly less. Lactose, citrate, acetate, and butyrate supported still less growth but more than did the mineral and water controls. All of the others allowed little if any more growth than occurred in the controls. Thus under the conditions of this experiment, no variations could be detected among the five accessions.

A second series of cultures was prepared using the eight carbohydrates and sodium citrate and sodium acetate as carbon sources. In addition, the four enzyme inhibitors sodium azide, sodium iodoacetate, sodium malonate, and sodium fluoride were added individually to the carbon sources for a final concentration of 0.001 M. The control for the carbon sources was the basic mineral solution, and the control for the inhibitors were the complete media without the inhibitor. Only accessions S-1 and S-8 were used.

The results for both accessions were similar. After three days incubation, azide inhibited the growth in all carbon sources, but did not necessarily stop it. None of the other inhibitors were effective in decreasing the growth appreciably below that of the control with no inhibitor. The data, of course, yield no indication of the site of activity of the azide. Nor do they indicate whether the other inhibitors failed because of impermeable membranes, or the absence of susceptible enzymes in the sporidia.

LITERATURE CITED