ADDENDUM:

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1. Scanning electron microscopy of chlamydospores of corn smut (Ustilago maydis (DC.) Corda).

In an earlier report (MGCNL 44:42, 1970) we discussed a new double-stage heat-pressure carbon replica technique. This technique was used to study chlamydospores of corn smut at the ultrastructural level using TEM (transmission electron microscope). Despite this, the conventional light microscope (LM) still has great potential, especially for measurements of size and for other comparative morphological studies of spores. In recent years a new type of electron microscope has been developed, the scanning electron microscope (SEM). The instrument is fundamentally different from the conventional TEM, in which either very thin sections or carbon replicas are used to record an image on photographic plates. In TEM an electron beam is transmitted through the objects, whereas in the SEM, entire objects (spores) are placed and a beam of electrons is used to bombard the object being studied. The electrons scattered by impact with the specimen and those emitted by the specimen during bombardment are collected to form a cathode-tube image, characterized by great depth of focus and a three-dimensional appearance. Before the objects are placed inside the SEM specimen chamber, it is important that the specimen have an even metal coating devoid of discontinuities. About 200Å to 500Å either pure gold or gold-palladium alloy coating is adequate. Stability of the image is further assured by addition of a thin coating of carbon (about 50Å to 100Å) prior to the metal coating, hence insuring greater stability of the specimens and also preventing artifacts under the electron beam. The coating process requires a vacuum-evaporator to evenly distribute the coating material which reflects the electrons. Micrographs taken with SEM further support our observations on the morphology of corn smut spores as earlier discussed. The tips of the spines are ordinarily curved, but the direction of the curvature varies from tip to tip. We postulate that this structural feature in some ways is an advantage to facilitate a mechanism for clasping the spores to the host tissues.

The chlamydospores used in this investigation were kindly supplied by Dr. J. E. Puhalla, at the Connecticut Experimental Station, New Haven, Conn.

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## 2. Pollen sterility in maize caused by fungus attack (*Ustilago maydis* (DC.) Corda).

Almost no information is available on pollen sterility caused by fungus attack on standing crops. The present investigation was carried out with respect to the influence of smut on maize pollen. A synthesized maize variety (a dent corn with the 4th chromosome from Nobogame teosinte) was chosen for this study. This maize variety exhibits an anomalous ear formation; at first the ears appear normal, but later, after the original tassel completes pollen shedding and the ears complete their normal pollination cycle, the tips of the ears elongate and produce staminate spikelets. The male spikelets show normal anther development and shed viable pollen grains which are perhaps useful for the younger developing ears at the receptive stage. Such anomalous ears are often called "laughing-ears." However, these plants also produce a normal tassel at the top. In this maize variety, the first four leaf-sheaths below the tassel are always sterile, the first ear being formed in the 5th leaf-sheath, and the second ear in the sixth leaf-sheath.

The seedlings in this study were grown in the greenhouse, then transplanted to the field. After the seedlings had reached about 6 inches in height, they were dusted with viable corn-smut chlamydospores. The chlamydospores were obtained from kernel infections. A cloudy, calm, humid afternoon was chosen for infecting the seedlings and the plants were carefully watered. The chlamydospores germinated and produced sporidia which infected the maize seedlings during the following evening. The infections appeared on the plants almost at the time when "laughing-ears" started to appear. The effect of fungus attack was appraised on the basis of the smut-gall formation on the ears bearing staminate flowers. In the control plants, staminate flowers of the "laughing-ears" developed normally and shed viable pollen grains at maturity. The male