MORPHOLOGICAL AND CYTOLOGICAL STUDIES OF *SCLEROSPORA* SPECIES ON CORN IN THAILAND

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Introduction

The downy mildew (DM) diseases reported thus far on corn *(Zea mays* L.*)* in Thailand are sorghum DM *(Sclerospora sorghi* Weston and Uppal*)* and brown stripe DM *(Sclerophthora raysseae* var. *zeae* Payak and Renfro*)*. The former is of greater economic importance. It occurs in 24 provinces and 50 to 100% loss in yield has been reported in the main corn-growing areas (2). Morphological studies of the corn DM pathogens have been made by many workers. They found that conidia were spherical, measured 16-29 × 11-21 μ (av. 22.3 × 14.3 μ) and germinated by forming a germ tube. No oospores have been observed in corn tissue. The conidia of *S. sorghi* reported by Weston and Uppal (7) were suborbicular, hyaline with thin walls, germinated by hyphae and measured 15-28.9 × 15-26.9 μ. Conidiophores were erect, had basal cells and a main axis and were dichotomously branched. The basal cell was approximately 100-150 μ in length, with 7-9 μ in width. The main axis was extended by 80-150 μ.

The cytological knowledge of the corn DMs is very limited. The nuclear number was reported to be 10-29, 10-26, 22-48 and 15-38 in *S. philippinensis*, *S. sacchari*, *S. miscanthi* and *S. spontanea*, respectively (6). Dogma *et al.* (3) showed that nuclear number was proportionate to conidial size. There is no report of nuclear number per conidium nor the basic chromosome number of *Sclerospora sorghi*. Studies were conducted to determine the morphology, cytology and germination of conidia of *S. sorghi*.

Materials and Methods

Infected corn leaves were incubated in moist petri dish for at least 8 hours at 24°C and examined morphologically (1). For sporulation and germination tests conidia were allowed to discharge on 2% thin plated water agar. For cytological studies, diseased leaves were incubated as described and the ones that produced conidia were sectioned; fixed in alcohol-glacial acetic acid solution for 24 hours and stained with iron-alum haematoxylin solution. The discharged or germinated conidia were fixed and stained in the same manner. Giemsa stain was also used for nuclear staining of conidia and germinated conidia on 2% thin plated water agar. For histopathological observation, sections of diseased leaves were fixed, embedded in paraffin and the ribbon tissues were stained with safranin and fast green (4). The fungal nuclei appeared dark red.

Results

Morphological studies of *Sclerospora sorghi*. — Conidia are nearly spherical, hyaline and thin walled. They measured 14.6-28.3 × 12.5-20.9 μ with an average of 23.5 × 17 μ. Conidiophores emerge singly or in groups from stomata and are erect, spreading, have a basal cell, a main axis and a branch system. They are usually 1, rarely 2 or 3, septate. The basal cell is knob-like at the bottom, then uniform in diameter of about 150 μ. The average length of the basal cells are about 164 microns and 5-8 μ in width. The length from the septum of the basal cell to the tip of the sterigmata is approximately 100 μ. The branch system is comprised of 3-4 primary branches which will produce the secondary dichotomously branch. Sterigmata are attached to this secondary branch and bear conidia which lie in a hemisphere. Conidia germinate by producing one germ tube with rarely two.

Cytological study of *S. sorghi.* — Conidia, conidiophores, germ tubes and mycelium were found to be multinucleate. The nuclei vary in size and shape. The number of nuclei in mature detached conidia was 6-12. The number of nuclei in the bulb-like conidiophore initial was 1-4. They then divided mitotically and at the same time increased in number, while the conidiophore enlarged. Nuclei divided mitotically and migrated into conidia through sterigmata. At an earlier stage, young conidia were formed at the tips of the sterigmata. One daughter nucleus migrated into the newly formed conidia, until each conidium contained an average of 6-12 nuclei before they were matured; i.e. until septa were formed. The number of nuclei in the detached matured conidia were eight (Fig. 1). These nuclei divided in the conidia as well as in the conidiophore.

Chromosomes within the nuclear envelope could be observed under an oil objective. The nucleus was rather spherical in shape and contained about 12 chromosomes which were linked together by a fine thread. The chromosomes were irregular in shape and the number varied from 12 to 24 during active mitotic division.

Microscopic examinations of diseased plant materials recently collected from Farm Suwan, Pakchong by Somkiat Titatarn revealed the presence of a different conidia kind, adjacent to and observed in the same microscopic field with *S. sorghi.* Subsequent study indicated mixtures of different species of *Sclerospora.* The newly found conidia were hyaline, cylindrical, measured 31-58 μ × 11-16 μ (Av. 14 × 44 μ). They had erect conidiophores 245-400 μ long comprising the basal cell, a main axis and were dichoto-

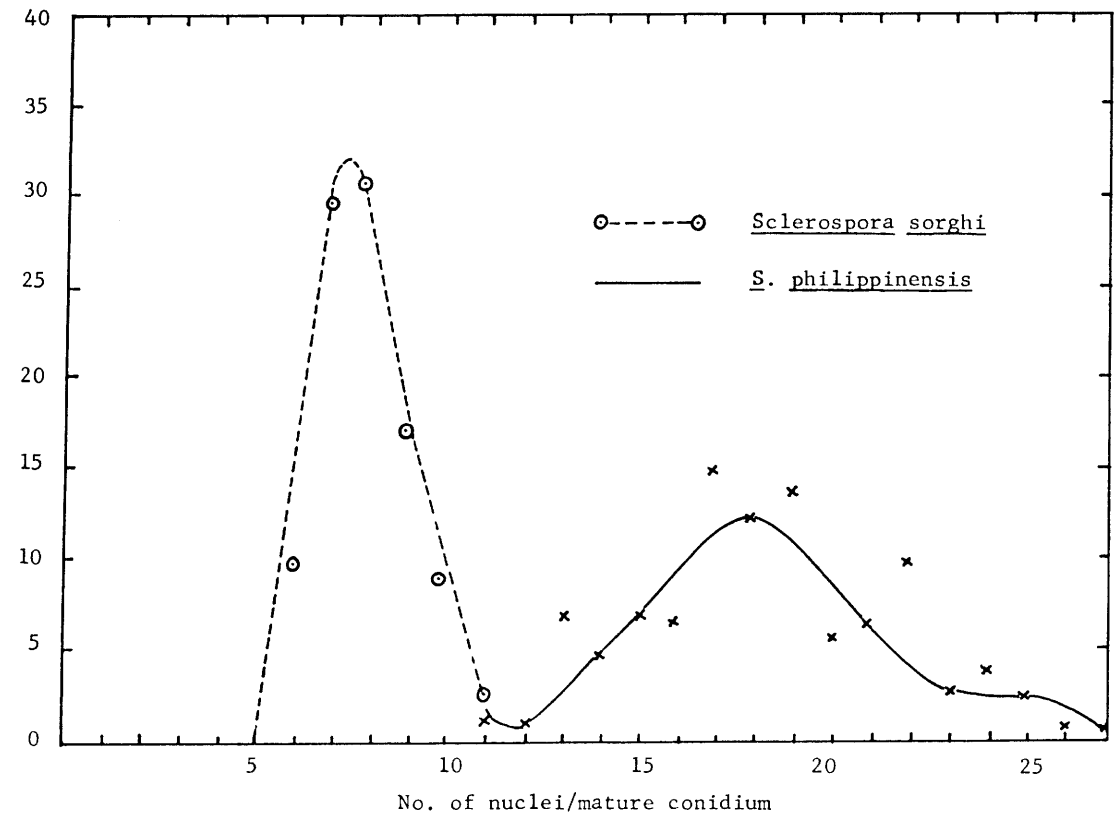

Figure 1. Number of the nuclei per mature conidium of *Sclerospora sorghi* compared to *S. philippinensis.*

mously branched. This description closely agrees with those of *S. philippinensis* Weston (7).

Growth and development of *S. sorghi* in corn leaves. — *S. sorghi* penetrated corn leaves directly through the stomata and, after colonization in the substomatal cavity, ramified intercellularly between mesophyll cells. The fungus formed a sheath partially surrounding the host cells and penetrated intercellularly. The size of the intercellular mycelium in the mesophyll was found to be variable and seemed to depend on the intercellular space. Several nuclei were observed within each thallus without the presence of a cross wall.

Discussion and Conclusion

The DM fungus reported here differs slightly in morphological characteristics from *Sclerospora maydis*, which infects maize in Indonesia (5). The conidia of *S. maydis* were reported to be spherical or sub-spherical and slightly larger in size. The morphological studies of the fungus causing DM of corn in Thailand are agreeable to the descriptions of Weston and Uppal (7) and Boon-Long (2). Hence, the nomenclature of this fungus should be *Sclerospora sorghi* (Weston and Uppal).

The newly found conidia which is first reported in Thailand as *S. philippinensis* Weston were also agreeable with those reported by Visarathanonth (6) in the Philippines. The mixing of these species of the same microscopic field may cause genetical variations of the pathogens and make it more difficult to control by using a resistant variety.

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