Infection of *Aedia leucomelas* L. (Lepidoptera : Noctuidae) by an *Entomophthora* Species of the “*Grylli*” Type in Japan

Tamotsu KUSHIDA, Kazumasa KATAGIRI

Asakawa Forest, Government Forest Experiment Station, Hachioji, Tokyo 193, Japan

and

Joji AOKI

Faculty of Agriculture, Tokyo University of Agriculture and Technology, Fuchu, Tokyo 183, Japan

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Field observations of an epizootic in *Aedia leucomelas* L. by an *Entomophthora* sp. of the “*grylli*” type were conducted during the autumn of 1974 in many fields of sweet potato in the Kanto District of Japan. Greater than 99% of the larval population was killed by the fungus. Most of the dead larvae were found at elevated parts of the plants. The infected larvae died within 9 days. The morphological characters of the fungus were similar to those of *Entomophthora grylli* FRES.

INTRODUCTION

There are several reports that *Entomophthora aulicae* (REICH.) SOROK. attacks some lepidopterous insects in Japan (HASEGAWA and KOYAMA, 1941; KOYAMA, 1954; AOKI, 1974). Recently, an entomophthoraceous fungus has been observed attacking *Aedia leucomelas* L. (Lepidoptera : Noctuidae) in the suburbs of Tokyo. In morphological characters, the fungus is more similar to *Entomophthora grylli* FRES. than to *E. aulicae*, which is the more common pathogen of lepidopterous insects. Some investigators (THAXTER, 1888; GUSTAFSSON, 1965) have placed *E. aulicae* in synonymy with *E. grylli*, but others (TUBEUF, 1893; LAKON, 1919; MACLEOD and MÜLLER-KÖGLER, 1973) consider them as distinct species. In this case, the infection of *E. grylli* on a lepidopterous insect is rather unusual. Accordingly, we have studied the fungus and its infection in the insect host.

OBSERVATIONS

Appearance of field infection

An extensive epizootic caused by an *Entomophthora* sp. occurred in populations of *A. leucomelas* during the rainy season in October, 1974. The observation was made in many fields of sweet potato, *Ipomoea batatas* (L.) POIR, var. β , *edulis* MAKINO, grown at Kamioshima, Sagami, Kanagawa Prefecture, Kanto District of Japan. The sweet potato leaves were severely attacked by a large number of *A. leucomelas* larvae, and there were very few intact leaves in the fields (Fig. 1A). At the end of the season, the fungus killed more than 99% of the larvae (Fig. 1B). A large number

Fig. 1. Field infection of *Aedia leucomelas* and *Herse convolvuli* by an *Entomophthora* species of the "grylli" type. A: A field of sweet potato infected by *A. leucomelas*; B: Dead larvae of *A. leucomelas* infected with the "grylli" type fungus; C: Dead larva of *H. convolvuli* infected with the "grylli" type fungus; D: Dead larva of *A. leucomelas* hanging from a sweet potato leaf.

of larvae of *Herse convolvuli* L. (Lepidoptera : Sphingidae) and *Agrotis fucosa* BUTLER (Lepidoptera : Noctuidae) were also attacked by the fungus in the same fields (Fig. 1C).

Symptoms and signs of infection

During the early stage of infection, the larvae showed no significant external symptoms. As the disease advanced, the larvae became restless, and prior to death, most of the infected larvae migrated to elevated parts of the plants. No color change and no patch formation were observed on their body surfaces until they died. Numerous dead larvae were attached with their abdominal legs to the leaves and petioles (Fig. 1D). Shortly after death, the dead larvae lost turgidity; the liquid contents of their bodies decreased; the larvae became flaccid and laterally compressed. Some of larvae fell on to leaves on the lower parts of the plants or to the ground. An hour or so after death, a greyish growth appeared on the dead bodies. The discharge of the conidia from the cadavers were initiated at the 5th hour after death and ceased at the 14th hour. The conidia were discharged to a distance of about 5 to 30 mm (the body length of the larva was about 30 mm). The cast-off conidia formed a white aureole around the dead larva. After the discharge of the conidia, the larval body became black and mummified.

When a conidial suspension ($6 \times 10^6/\text{ml}$), which was prepared from the cadavers, was sprayed on the larvae at laboratory, the larvae were killed within 9 days.

Morphological characters of the fungus

The hyphal bodies are unicellular, and variable in form. The conidiophores which develop from the hyphal bodies are unbranched and cohered. The conidiogenous cells are club-shaped (Figs. 2A, B). The conidia are hyaline, mostly pyriform

Fig. 2. Morphology of an *Entomophthora* species of the "grylli" type. A: Conidiogenous cells ($\times 75$); B: Scanning electron micrograph of the conidiogenous cells. ($\times 500$); C: Scanning electron micrograph of conidia ($\times 500$); D: Conidia containing a globule ($\times 300$); E: Chlamydospores produced in the host cavities ($\times 600$); F: Chlamydospore formation from the tips of hyphal bodies. ($\times 300$).

with a broad papillate base and evenly rounded apex, and $20\text{--}44 \times 18\text{--}23 \mu$ (av. $31 \times 21 \mu$) in size. The basal papillae are not extended (Fig. 2C). The contents of the conidium are granular with one large globule that ranged in diameter with an average of $11 \times 13 \mu$ (Fig. 2D). Sometimes two or more globules are found in a conidium. The resting spores have not been observed, but some colorless chlamydospores, with thin walls, are produced in the host cavities (Fig. 2E). The chlamydospores are spherical or near spherical, $14.5\text{--}17.0 \mu$ in diameter, and are produced interstitially or from the tips of the hyphal bodies (Fig. 2F). The cystidia and rhizoids have not been observed.

DISCUSSION

THAXTER (1888) and GUSTAFSSON (1956) placed *E. aulicae*, a major pathogen of lepidopterous insect, in synonymy with *E. grylli*. On the other hand, TUBEUF (1893) and LEKON (1919) regarded them as distinct species. Recently, MACLEOD and MÜLLER-KÖGLER (1973) also recommended that *E. aulicae* and *E. grylli* be retained as distinct species. They separated *E. aulicae* and *E. grylli* on the basis of differences

in the shapes of the conidia, the former with mostly obovate and the latter with mostly pyriform conidia. AOKI (1974) observed that *E. aulicae*, found attacking *Lymantria dispar japonica* Motsch. (Lepidoptera : Lymantridae), had mostly obovate conidia, which differed distinctly in shape from those of the present fungus. Our observations suggest that, on the morphological characters, the present fungus is similar to *E. grylli* rather than to *E. aulicae*.

MACLEOD (1956) isolated an entomophthoraceous fungus from many lepidopterous insect species. According to this report, the conidia of the fungus are not separable from those of *E. grylli*. He treated this fungus as an *Empusa* (*Entomophthora*) *grylli* type, and reported that the resting spores (zygospores or azygospores) were not produced by any isolate, except one. This situation resembles that of our present fungus. Furthermore, the size of the chlamydospores of our fungus closely matches the description of the "*E. grylli*" type. Accordingly, the present fungus may be the same species as the "*E. grylli*" type described by MACLEOD. He also reported that the "*E. grylli*" type is one of the more widely distributed and generally abundant of the *Empusa* (*Entomophthora*) species. Nevertheless, MACLEOD and MÜLLER-KÖGLER (1973) did not establish a taxonomic position for the "*E. grylli*" type in their recent revision of the classification of *Entomophthora* species. Several investigators have cited *E. grylli* as a pathogen on species of Orthoptera, Lepidoptera, Diptera and Coleoptera (NOWAKOWSKI, 1883; THAXTER, 1888; SHELDON, 1903; RIDDLE, 1906; ROZSYPAL, 1958; GUSTAFSSON, 1965). However, these observations need confirmation (MACLEOD and MÜLLER-KÖGLER, 1973). We believe that our present fungus, "*E. grylli*" type, and *E. grylli* may be identical. We question whether *E. grylli* normally infects only Orthoptera, and this should be determined by cross infection tests with the present fungus and *E. grylli* on different orders of insects. It has also been left unsolved whether or not our fungus, "*E. grylli*" type, can grow on an artificial culture medium.

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