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**Scientific Notes**

# Viability of conidia of *Peronospora tabacina* on Tomato Fruit

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**Abstract** : *Peronospora tabacina*, pathogen of tobacco blue mold, is not occurred in Japan and one of the important quarantine pests. The viability of conidia of *P. tabacina* was evaluated on tomato fruit in Canada. Conidial suspension of *P. tabacina* ($10^{4-5}$ conidia/m*l*) was inoculated to the surface of tomato fruit and stored with shade at 16-36°C, RH 25-80% for 2 to 8 days in a glasshouse. The present experiment showed that the viability of conidia of *P. tabacina* on tomato fruit was less than 2 days.
**Key words** : viability, *Peronospora tabacina*, tomato

## Introduction

*Peronospora tabacina* D.B. Adam (Syn. *P. hyoscyami* de Bary) is a causal agent of tobacco blue mold which has great economic impact on tobacco production in Europe, North America and Australia. During the early 1960s, *P. tabacina* has spread from north-western Europe east to the U.S.S.R. and Turkey and south to Algeria and Tunisia. Carried by the predominant west wind, the pathogen spread quickly and within four years had invaded the tobacco-growing areas of Europe and the Mediterranean countries (DELON and SCHILTS, 1989). In 1979, a severe epidemic of blue mold occurred throughout the tobacco production areas of the United States and Canada. Estimates of crop losses caused by the disease were US $250 million in 1979 and more than US $84 million in 1980 (DAVIS *et al.*, 1981). Tobacco blue mold is a high-risk disease.

The asexual stage of the pathogen is very important in spreading the disease (MAIN and DAVIS, 1989). Many studies (HILL, 1962 ; KRÖBER, 1965 ; COHEN and EYAL, 1984 ; ZHANG *et al.*, 1994) have indicated that conidia of *P. tabacina* have long viability under the dry and cool conditions. Japan is a country with tobacco cultivation, and provides a range of climatic conditions suitable for tobacco blue mold. *P. tabacina* has been shown to have a high potential to establish at tobacco production areas around the world. This fungus is thus an important quarantine pest for Japan.

In recent years, ban of importation of fresh fruit of several tomato varieties from Canada and the United States has been lifted. As tomato plant was reported as a host of *P. tabacina* in the USA by ARMSTRONG and ALBERT (1933), whether fresh tomato fruit can be a carrier and a source of primary infection by *P. tabacina* during transportation and processing of imported fruit is of quarantine concern. Therefor, prior to the lifting ban, the viability of conidia of *P. tabacina* was accordingly investigated on tomato fruit. This experiment was carried out at the Centre for Plant Health, British Columbia, Canada in

1995.

## Materials and Methods

Inoculum

*P. tabacina* KY-78 introduced from the University of Kentucky was maintained on tobacco seedlings (cv. White Burley) in a growth chamber set at 20/18°C day/night, 16 hour photo period. The atmosphere of the growth chamber was water saturated for 24 hours immediately following inoculation and 48 hours prior to expected sporulation. *P. tabacina* conidia were collected in distilled water and their concentration was adjusted to $10^{4-5}$ conidia/m*l* before use. Germination of conidia was confirmed on an agar plate before experiments (50-60%).

Inoculation

Conidial suspension of *P. tabacina* was inoculated to the surface of tomato fruit (cv. trust) and then pieces of filter paper were placed onto the inoculated place. Inoculated fruit were placed in a plastic box with shade at 16-36°C, RH 25-80% for 2 to 8 days in a glasshouse.

Examination of conidia

Two, four, six and eight days after inoculation, pieces of filter paper were removed from the inoculated fruit and cut in two with a scissors. One piece was infused with 0.1% acridine orange (Sigma) and examined microscopically with epi-fluorescence (495 nm). At least 100 conidia in 10 microscope fields per sample were observed and presence of germinating conidia was recorded. The other piece was placed onto the healthy leaves of tobacco and maintained in a growth chamber for 2 to 6 days as described above in Inoculum. After maintenance, tobacco leaves and the piece of filter paper were examined microscopically as described above, or the tobacco leaf tissue was cleared by heating in lactophenol and stained with cotton blue. Upper and/or bottom of the leaf surface were examined microscopically and presence of germinating conidia or *P. tabacina* hyphae was recorded as described above.

## Results and Discussion

There were no germinating conidia on the pieces of filter paper nor tobacco leaves (Table 1). Hyphae of *P. tabacina* were not observed in the leaf tissues treated with lactophenol or acridine orange.

These results show that conidia of *P. tabacina* on the tomato fruit have been died within 2 days after inoculation in the glasshouse. In general, conidia of *P. tabacina* have a remarkable capacity for survival under the dry conditions characteristic of the regions in which *Nicotiana* spp., the natural host plants, occur. However, the survival times were very short at high relative humidity (80-100%) (HILL, 1962). In this experiment, viability of conidia was examined on the tomato fruit stored in the glasshouse. Those conditions

**Table 1.** Number of germinating conidia of *P. tabacina* on tomato fruit 2-8 days after inoculation.

| Days after inoculation | Sample | Piece of filter paper on | | Tobacco leaf observed with | |
|---|---|---|---|---|---|
| | | tomato fruit | Tobacco leaves | lactophenol | acridine orange |
| 2 | 1 | 0/100[a)] | 0/100 | ND[b)] | ND |
| | 2 | 0/100 | 0/100 | ND | ND |
| | 3 | 0/100 | 0/100 | ND | ND |
| 4 | 1 | 0/100 | 0/100 | ND | ND |
| | 2 | 0/100 | 0/100 | ND | ND |
| | 3 | 0/100 | 0/100 | ND | ND |
| 6 | 1 | 0/100 | 0/100 | ND | ND |
| | 2 | 0/100 | 0/100 | ND | ND |
| | 3 | 0/100 | 0/100 | ND | ND |
| 8 | 1 | 0/100 | 0/100 | ND | ND |
| | 2 | 0/100 | 0/100 | ND | ND |
| | 3 | 0/100 | 0/100 | ND | ND |

a) No. of germinating conidia/no. of conidia observed.
b) ND: not detected.

were similar to them (e.g. 16-22°C, RH 71-88% in Jan. 1993) of the glasshouses for commercial tomato production in British Columbia, Canada (Mauza, personal communication).

For plant quarantine purposes, it is reasonable to test the viability of a harmful pathogen on the plant first of all. If the quarantine pest does not survive on the imported materials, they can more safely be imported and the risk of introduction of *P. tabacina* into a new area could be managed to a negligibly low level (ZHANG *et al.*, 1994). The present experiment showed that the viability of conidia of *P. tabacina* on tomato fruit was less than 2 days.

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## 和 文 摘 要

# トマト果実上におけるタバコベと病菌（*Peronospora tabacina*）分生胞子の生存期間

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タバコベと病菌（*Peronospora tabacina*）は我が国未発生の重要な検疫病害虫の一つである。カナダにおいて，トマト果実上における本菌の分生胞子の生存期間を調べるため，胞子懸濁液($10^{4-5}$個/m*l*)を果実表面に接種し，ガラス室内（16-36°C, RH 25-80%）の日陰に2から8日間保管した。試験の結果,供試菌の分生胞子の生存期間は2日間以内であった。