

# Angular Leaf Spot of Cucumber in Japan

YASUMASA WATANABE\* and AKIRA OHUCHI\*\*

\* National Institute of Agricultural Sciences

(Yatabe, Ibaraki, 305 Japan)

\*\* Hokuriku National Agricultural Experiment Station

(Inada, Joetsu, Niigata, 943-01 Japan)

Cucumber angular leaf spot is one of the most prevalent diseases of cucumber (*cucurbit*) in Japan. The affected fields covered 6,240 hectares in 1980, accounting for about a half of the total cucumber-planted area including plastic house cultivation.

The first incidence of cucumber angular leaf spot in Japan was reported by Tominaga & Tsuchiya<sup>14)</sup> in 1957. Since Mukoo et al.<sup>6)</sup> recognized its severe incidence in Kanto district in 1971, it has spread throughout Japan. The causal bacterium not only produces spotted lesions on cucumber leaves (Plate 1) but also affects cucumber fruit (Plate 2), followed by entire decay of the fruit.

The disease was so severe in cucumber-growing districts that the Ministry of Agri-

culture, Forestry and Fisheries organized a co-operative research program "Studies on the integrated control of bacterial diseases in cucurbits"<sup>23)</sup> which lasted four years from 1976 to 1979, in order to establish a combined cultural and chemical control procedure. The authors will describe outline of the results obtained in this research program.

## Causal bacterium

Ohuchi et al.<sup>9)</sup> examined more than one hundred isolates collected from affected cucumber plants which were distributed in the various parts of Japan and identified 110 isolates as *Pseudomonas syringae* pv. *lachrymans* (Smith & Bryan 1915) Young, Dye &



Plate 1. Typical symptom of cucumber angular leaf spot on leaf



Plate 2. A fruit affected by cucumber angular leaf spot disease, showing a clear secretion of tear-like droplets

Wilkie 1978 on the basis of their bacteriological properties and pathogenicity to host plants. On the contrary, the isolates from the lesions characterized by blighting of leaf margin were identified as *Pseudomonas viridiflava* (Burkholder 1930) Dowson 1939 (Table 1).<sup>10)</sup> This bacterium was responsible for the soft rot of cucumber fruit which was often observed in the cucumber fields affected by angular leaf spot, and was advocated as an additional causal agent of marginal blight of cucumber.<sup>12)</sup>

Ohuchi et al.<sup>11)</sup> developed successfully a rapid method for distinguishing *P. syringae* pv. *lachrymans* from other bacteria. The method consists of inoculating with bacterial

isolates by needle pricking on cucumber fruit segments and incubating in moist chamber at 24°C. *P. syringae* pv. *lachrymans* was detected by conspicuous ooze exudation on the fruit segments (Plate 3). Although several other bacteria were capable of exudating bacterial ooze, they were not pathogenic to cucumber, except *P. syringae* pv. *mellea* and *P. syringae* pv. *tabaci*. These two bacteria were distinguished from *P. syringae* pv. *lachrymans* by yellow haloes surrounding lesions on cucumber leaves.

### Disease cycle

The disease has spread throughout Japan in these several years. Such rapid spread of the disease suggests that the causal bacterium should be carried by infested seeds, as was proved by Carsner in 1918.<sup>2)</sup> Exact seed transmission of the disease in Japan was confirmed through the experiment using the seeds obtained from naturally infected fruit of cucumber in the open field for seed production.<sup>7,16)</sup> The rate of seed transmission was as low as less than one per cent. Less than one per cent of seed transmission, however, was proved to be enough for overall incidence of angular leaf spot in a nursery bed, when the bed was kept under high moisture condition.<sup>16)</sup>

Transmission of the disease through infested soil has been suspected in the fields where angular leaf spot broke out every year. Soil transmission, however, has not been confirmed clearly yet. Ogawa et al.<sup>8)</sup> showed that the disease symptoms appeared on the cotyledons of cucumber seedlings when the causal bacteria were introduced in the soil above sowing level, suggesting direct contact of cotyledons with causal bacteria during germination of seeds. The longevity of causal bacteria in soil was examined at several laboratories in Japan using indicator plant method.<sup>23)</sup> The longevity was dependent on seasons when diseased cucumber vines were buried in soil. In summer the causal bacterium died within 10 to 20 days, whereas longevity of causal bacterium was 3 to 4

Table 1. Major bacteriological characteristics of two species of pathogenic bacteria

Property	<i>Pseudomonas syringae</i> pv. <i>lachrymans</i> (110 isolates from affected cucumber plants characterized by angular spotted lesion on leaf)	<i>Pseudomonas viridiflava</i> (45 isolates from affected cucumber plants characterized by blighting of leaf margin)
Gram stain	—	—
O-F test	0	0
Flagella (polar)	1-5	>5
Poly- $\beta$ -hydroxybutyrate accumulation	—	—
Fluorescent pigment	+	+
Growth at 37°C	—	—
Growth in Cohn's solution	+	—
Arginin dehydrolase activity	—	—
Oxidase activity	—	—
$\beta$ -glucosidase activity	+	+
Tyrosinase activity	—	—
Hydrolysis of casein	—	+
Hydrolysis of cotton oil	—	—
Production of levan	+	—
Nitrate reduction	—	—
Liquefaction of gelatin	+	+
Utilization of tartrate	+	—
Utilization of sucrose	+	—
Softrot activity to potato	—	+
Hypersensitivity to tobacco	+	+

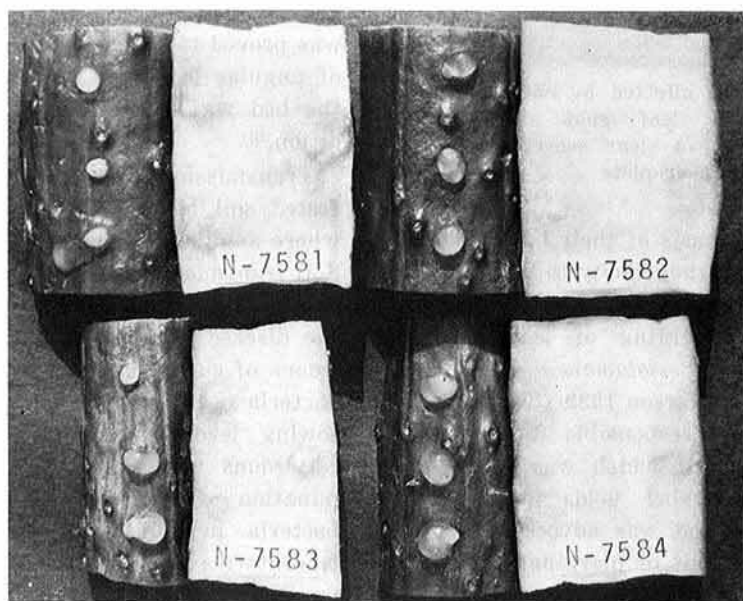


Plate 3. Conspicuous ooze exudation on the fruit segments inoculated with *P. syringae* pv. *lachrymans* by needle pricking

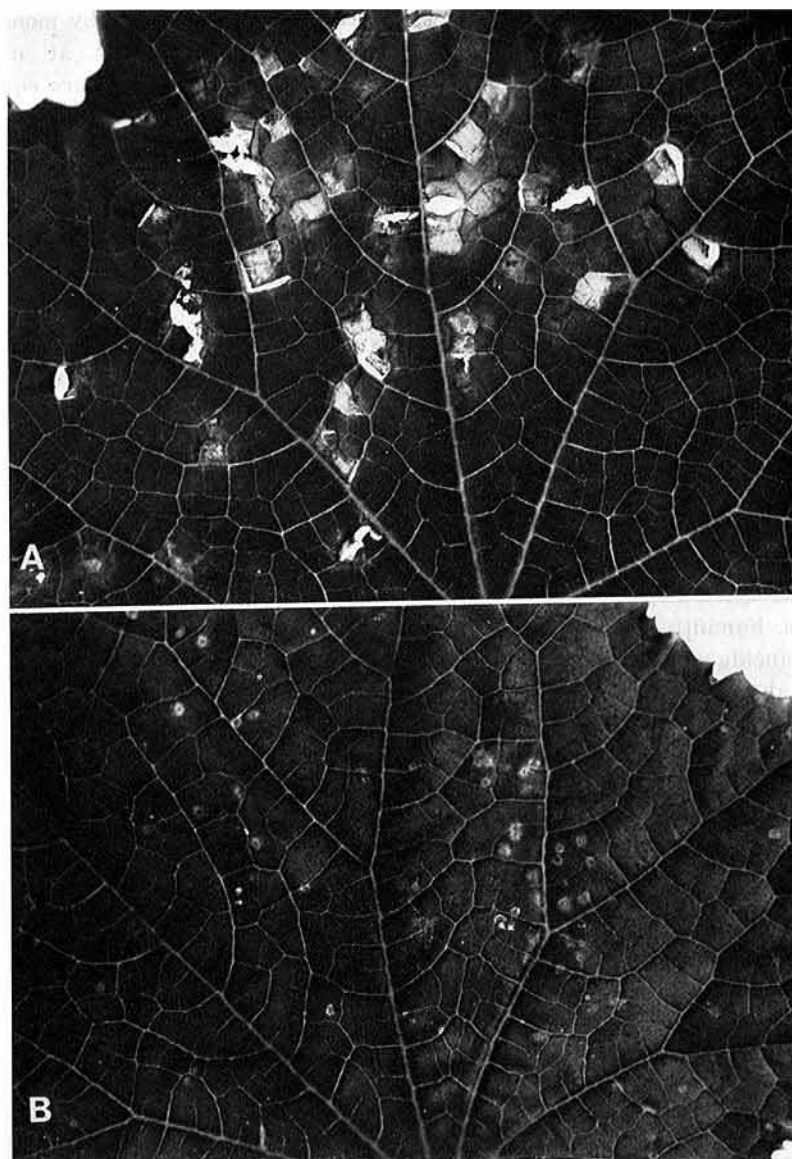


Plate 4. Effect of atmospheric humidity on the development of lesions of cucumber angular leaf spot

- A: characteristic angular spotted lesions developed under high moisture condition (relative humidity of 90–94% or above)  
 B: minute lesions developed at 85% relative humidity

months in winter.<sup>4)</sup> These results indicate that soil transmission of the disease is likely to occur in winter, if the period of fallowing in cucumber fields is within 3 to 4 months.<sup>5)</sup>

### Predisposing factors

Angular leaf spot of cucumber in Japan is severe in plastic house cultivation in winter as well as in open field cultivation in the northern part of Japan. Thus it seems that

a condition which combines low temperature with high humidity is favorable for the severe occurrence of the disease. Umekawa & Watanabe<sup>19)</sup> investigated the effect of temperature and atmospheric humidity on the occurrence of angular leaf spot of cucumber. The lesions developed severely at 15° to 25°C under a moisture saturated condition. Relative humidity of 90 to 94% or above was essential for the development of characteristic angular lesions, whereas only minute lesions developed at 85% relative humidity (Plate 4). The duration of misting after spray of bacterial suspension had a marked effect on lesion development. Twenty-four hours duration of misting after inoculation was necessary for the development of characteristic lesions, whereas minute lesions were produced when inoculated plants were kept under dry condition (relative humidity of 40 to 60%). These results coincide substantially with those of Williams et al.<sup>21)</sup>

Table 2. Effect of duration of misting at night on the development of lesions of cucumber angular leaf spot under a diurnal alternation of humid night and dry daytime

Diurnal alternation		Disease index	Type of lesion
Duration of misting (hr)	Duration of dry condition (hr)		
0	24	1.1	Minute lesion
1	23	1.1	Minute lesion
3	21	2.1	Minute lesion
6	18	3.1	Developed lesion
12	12	3.0	Developed lesion

Atmospheric humidity in fields or plastic houses, however, varies rhythmically from humid condition at night to dry condition in the daytime. Under such an alternating humidity, the duration of less than 3 hr of moist condition at night could not produce angular lesions but minute lesions on cucumber leaves (Table 2). These results suggest that it may be possible to reduce the incidence of angular

leaf spot of cucumber by means of controlling atmospheric humidity at night in plastic houses. The dehumidifiers equipped in plastic houses proved to be effective in controlling the incidence of angular leaf spot as well as for keeping atmospheric humidity in plastic houses below 90%.<sup>19)</sup> Only slight marginal blight lesions were developed on leaves of cucumber grown in the equipped houses (Plate 5).

Rainfall affects the incidence of cucumber angular leaf spot in open-field cultivation in northern part of Japan. Umekawa et al.<sup>18)</sup> revealed that the rain water dripping from the diseased leaves contained the causal bacteria at the concentration of  $10^5 \sim 10^6$  cell per ml of rain drop, and sheltering of cucumber vines by roof of plastic film was able to reduce the development of angular leaf spot lesions. They concluded that cultivation of cucumber avoiding rain under plastic roof was effective on the control of angular leaf spot.

Moisture content in soil also affects the development of lesions on leaves. Experiments conducted in a green house provided the results that the cucumber plants grown in the soil with higher moisture content were affected more severely by angular leaf spot.<sup>13)</sup>

## Varietal resistance

Most commercial varieties of cucumber now in use in Japan are susceptible to angular leaf spot, whereas 'Kurume-ochiai H and Maji-midori' were proved to be tolerant through the co-operative research program.<sup>23)</sup> Umekawa & Watanabe<sup>17)</sup> established the method of assaying the resistance to angular leaf spot during a seedling stage and evaluated that 'Saitama-ochiai No. 4, Shogoin-aonagafushinari, Kariha-fushinari and Sado-risshu' were as resistant as 'Poinsett' which was rated as highly resistant variety in USA.<sup>1)</sup> In order to develop the resistant varieties to angular leaf spot, Kawaide<sup>3)</sup> selected 'Saitama-ochiai No. 4, Sakata and Suyo for parents and established the populations carrying resistant genes.



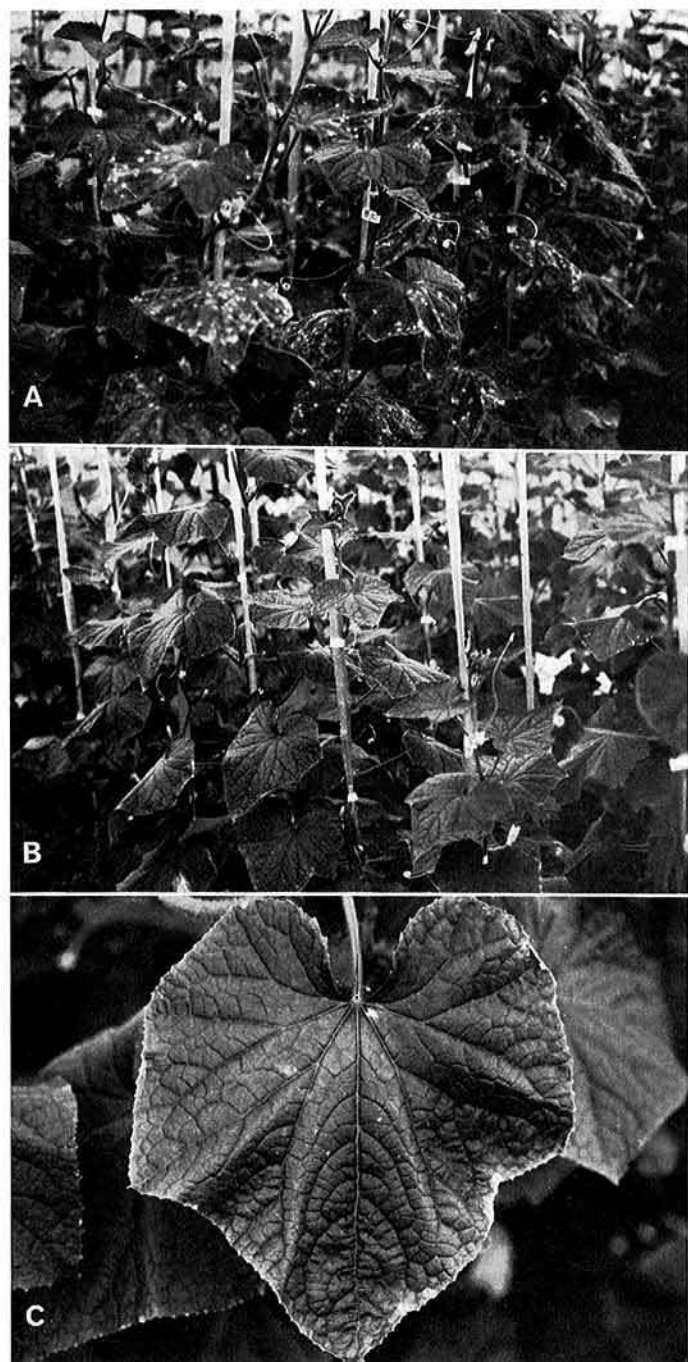


Plate 5. Effect of dehumidifiers equipped in a plastic house on the incidence of cucumber angular leaf spot in the plastic house.

- A. severe incidence of cucumber angular leaf spot in a standard plastic house
- B: very few lesions on the leaves of cucumber grown in a dehumidifier-equipped plastic house
- C: slight marginal lesions on the leaf of cucumber grown in the dehumidifier-equipped plastic house

## Seed disinfection and spray of chemicals

Various methods of seed disinfection against angular leaf spot were examined through the co-operative research program.<sup>23)</sup> Among the methods examined, soak treatment of infested seeds in 1:20 dilution of sodium hypochlorite solution (active Cl: 4%) for 20 min was proved to be effective against seed transmission of angular leaf spot from a lot of experimental results. Soak treatment in 0.25 to 0.5% solution of "Chemiclone G" (Calcium hypochlorite, active Cl: 70%) for 60 min was also effective. They were recommended for practical use because of their little phytotoxicity to seed germination. Soak treatment in streptomycin solution was not practical because of wide distribution of streptomycin-resistant strain of *Pseudomonas syringae* pv. *lachrymans* in Japan.<sup>22)</sup>

Hot water treatment at 54°C for 20 min or dry heat treatment at 70°C for 3 days gave promising results<sup>15)</sup> without causing a decrease in germination capacity of seeds during 6 months' storage after the hot water treatment or during 11 months' storage after the dry heat treatment.

Spray of Bordeaux mixture and inorganic copper compounds was practical<sup>20)</sup> for protecting the vines and fruit of cucumber from the disease, though inflicting slight phytotoxic lesions on vines.

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