한국식물보호학회지 제16권 제2호
Kor. J. Pl. Prot. Vol.16, No.2, p. 87~92(1977)

Occurrence of Northern Cereal Mosaic Virus in Korea

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우리나라 麥類 北地 모자익 바이러스病의 發生에 關하여

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ABSTRACT

A barley virus disease has been severe in central Korea since 1963.

To investigate the causal virus, examination of host ranges, transmission by insect vectors and electron microscopy were conducted.

In electron microscopy, particles identical with northern cereal mosaic virus were observed. The size of baciliform particles ranged from 300nm to 370nm in length and 57~60nm in diameter.

The virus was transmitted by the small brown planthopper *Laodelphax striatellus* (Fallen). The latent period in the vector was seven to nineteen days, with 10 days the most prevalent.

Barley, corn, wheat, rye, and oats were susceptible to the virus when inoculated by the insect vectors. It was concluded that the disease agent of the barley disease in Korea is northern cereal mosaic virus.

This is the first known report of this disease in Korea.

INTRODUCTION

A barley disease was observed in central Korea in 53. The disease became a serious problem in barley ds throughout the country in Chungjoo, Jinju and rangju districts in 1967.

Symptoms on barley plants were very similar to se of northern cereal mosaic virus (NCMV) in an. Preliminary studies by electron microscopy ealed the size and shape of NCMV dip preparations. Northern cereal mosaic virus was first reported by and Fukushi (1943) in Japan (4) They established host ranges and found inclusion bodies in the cells NCMV infected oats and the insect vector *Laodelphax atellus* Fallen (5, 6).

Later 3 species of insects, *Delphacodes albifascia* Matsumura, *Unkanodes sapporonus* Matsumura, and *Meulerianella fairmairei* Perris were also determined to be insect vector (1, 2, 3, 11).

Murayama and Lu (1967) studies the properties of NCMV by injecting virus extracts into vectors (9). Shikata and Lu (1967) demonstrated the presence of bacilliform particles in dip preparations from infected leaves, partially purified preparations, and in cells of diseased plants and insects by means of electron microscopy (7, 8, 9, 10). Yamada and Shikata (1969) were able to demonstrate multiplication of the virus in insect organs by organ culture methods (12).

This paper will describe studies on host ranges, insect transmission and electron microscopy of this barley

disease in Korea. The results indicated the causal agent of this disease is identical with NCMV. Symptoms and epidemiology of this disease also noted.

This is the first report of NCMV occurring in Korea.

MATERIALS AND METHODS

Diseased barley plants were collected from the fields of Institute of Agricultural Sciences in Suweon and maintained in the greenhouse.

Host range

Viruliferous insects *Laodelphax striatellus* Fallen fed on diseased barley for more than 6 days were transferred to barley, wheat, rye, oat, and corn plants in test tubes for an inoculation period of 3 days. Individual plants were inoculated with 2 viruliferous vectors in test tubes. After 6 days plants were removed and planted in large flat where development of disease symptoms was recorded.

Transmission by insects

Non-viruliferous *L. striatellus* were confined on healthy rice seedlings in the insectarium. Two to three instar nymphs had on diseased barley for 6 days were transferred individually to singly barley plants in a small test tube for one day feeding. The daily transfer of each insect to barley was conducted until the vector died. The inoculated plants were removed and planted in a large flat for development of the disease symptoms.

Electron microscopy

A dip method in a 2% phosphotungstic acid (PTA) was employed. The diseased leaves were cut with a razor blade into 1×5mm sections and the cut edge was dipped into 2% PTA on carbon coated formvar grids, and them dried. A Hitachi model HU-11E electron microscope was used for observation.

Ultrathin sections were made to obseve the virus particles in the plant cells. Diseased leaf, sections 1×5 mm, were fixed in 2.5% glutaraldehyde in 0.1M phosphate buffer at pH 7.0 for 90 minutes and post-fixed in 2% Osmium tetroxide. The fixed materials were dehydrated in 75, 90, 95, 100% of alcohol for 60 minutes. The materials were embedded in Epon and cut with glass knives with a Sorvall MT-1 Ultramicrotome.

The Ultrathin sections were doubly stained with uranyl acetate and lead citrate and them examined by a JEM 5-Y on electron microscope.

SYMPTOMS AND EPIDEMIOLOGY OF THE DISEASE

In barley field, natural infection rates were dependent on cultivars. Winter barley showed about 12% infection, and spring barley was about 11.3%. However, spring barley showed more severe infection, with 100% infection occurring in some areas.

The first symptoms of this disease were yellowish-white or whitishgreen specks on the young and newly-emerged leaves of barley, These symptoms also appeared on the leaf sheath. As the disease progressed, the infected plants became stunted and tillering increased. Fully developed symptoms on the diseased leaves with shortened narrow leaf blades often appeared as stripe patterns along the leaf veins.

The diseased plants produced either no or only few panicles. When infection occurred at early stages of plant growth, small panicles bored mostly unfilled grains.

RESULTS

Host range

Table 1. Susceptible hosts of Northern Cereal Mosai Virus

Host	No. of plants		% infection
	inoculated	infected	
Barley (*Hordeum vulgare L.*)	30	19	63
Rye (*Secale cereale L.*)	30	9	33
Oats (*Avena sativa L.*)	15	5	33
Corn (*Zea mays L.*)	12	2	16
Wheat (*Triticum aestivum L.*)	15	4	21

Five species of plants including barley, wheat, cc rye and oats, were inoculated with viruliferous inse All species inoculated produced disease symptoms.

As shown in Table 1, infection rates of plants NCMV were about 63, 33, 33, 16, and 21% in bar rye, oats, corn and wheat, respectively. The hig rate of infection was 63% in barley, and the lo in corn, 16% natural infections of NCMV to t

ɔst plants, except for corn, were found also in the ɔld.

The symptom on artificially inoculated barley seedlings pared as yellowish-white stripes parallel to the midrib, d infected leaves became short and narrow. Rye, oats, wheat, and *Alopeculus aeqalis* showed similar symptoms to those of barley. On corn the symptoms appeared in a stripe pattern on only one side of the leaf blade.

Transmission by insects

Fig. 1 Insect Transmission of NCMV by *L. striatellus* after feeding on diseased plants for 6 days.

s shown in Fig. 1 latent periods of the virus in *striatellus* were 7 to 19 days, however, the most alent period was 10 days.

onsistent serial transmission was not observed in transfers. This may be due to plant growth under vorable conditions in the green house.

Electron microscopy

The virus particles observed in dip preparations from the diseased barley leaves were baciliform, bullet shaped, as shown in Fig. 2. The size of the particles ranged from 300 to 370 nm long in lenght, with a width of 57–60 nm, Most particles showed cross-striations across

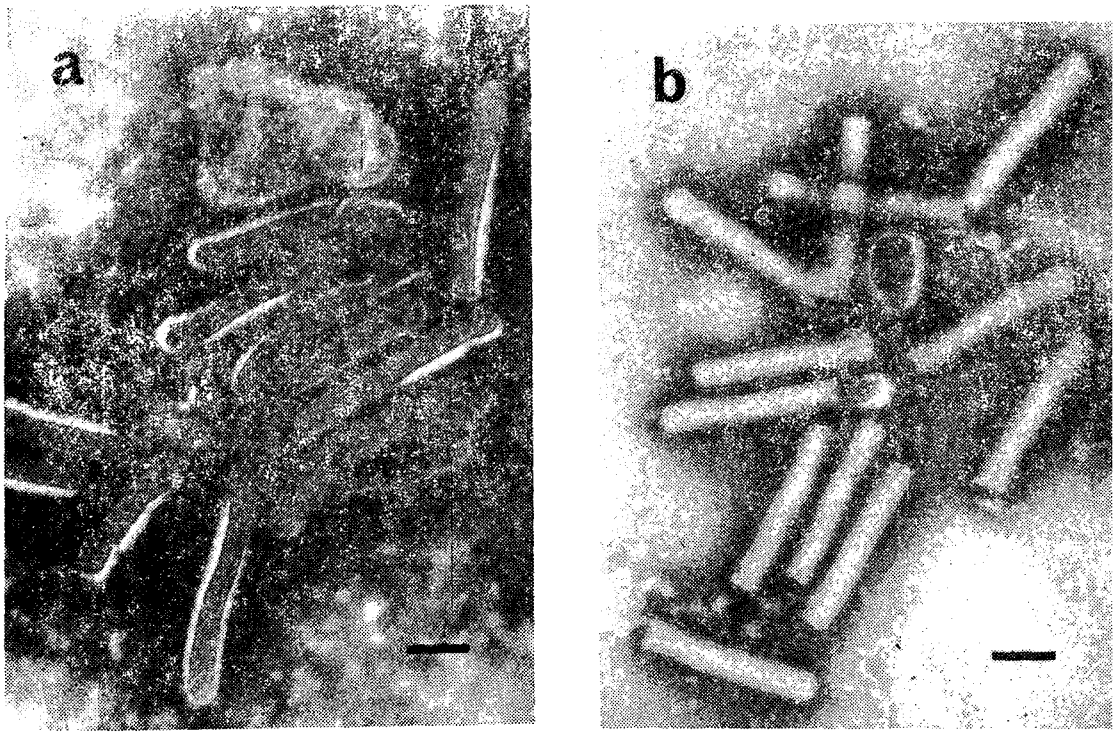

Fig. 2 An electron micrograph ofnorthern cereal mosaic virus by negative staining in dip preparation shov (a) empty particles with a central canal and (b) thick particles without canal. Bar length 100nm.

Fig. 3 Virus particles in a barley cell prepared by ultrathin section and stained with uranyl acetate and lead citrate. Bar shows 300nm.

Fig. 4 Symptoms of northern cereal mosaic viru the weed, *Alopeculus aequalis* Sosbol, colle from the field in Suweon.

the particles, but some particles showed no cross-banding (Fig. 2-b). Abundant baciliform particles were distributed in cytoplasm of infected cells of barley leaves.

The cross-sections of the particles showed two-layered outer membranes with a round central core (Fig. 3). The results obtained appeared identical to those of the previous study (10).

DISCUSSION AND CONCLUSION

The symotoms of this virus disease on barley in Korea varied little different with the cultivars. Yellowish-white specks were the most common symptoms on young leaves in early stages of infection. This was followed by formation of yellowish-white stripes parallel with the leaf veins in more ad anced stages. These symptoms were identical to those reported by Ito and Fukushi (1944).

In insect inoculation tests, oat barley, rye, corn and wheat were confirmed as host plants of the virus. In addition to these hosts natually infected populations of the weed species (*Alopeculcs aequalis*) were found n the fields.

As Ito and Fukushi reported that 23 species of plants ncluding the above 6 species were host plants of NCMV, and the symptoms on *A. aequalis* were dentical this weed plant was considered an important econdary host plant, responsible for the carry voer of irus sources in fields.

Planthoppers such as, *L. striatellus* (Fallén), *D. albi-ıscia* (MASTUMURA), *U. sapporonus* (MASTUMU A), and *M. faimairei* are known as vectors of NCMV Japan. In this study, *L. Striatellus* (Fallén), commonly und in Korea, was used in transmission tests with od results.

The incubation periods of the virus in vectors were 19 days, which was also similar to development of CMV in host plants.

The shape, size and structure of the virus particles served in leaf dip methods were identical wite the ults reported by Shinkata and Lu (1967). The virus rticles observed in Ultrathin sections were also ntical to those obtained the leaf dip method.

According to these results, it can now be concluded established that the virus disease of barley, widely ributed throughout the country, particulary prevalent in kyunggi, Chungbuk and Chungnam provinces, is caused by NCMV. This disease has been found in central and northern parts in Japan, and is now known to distribute throughout Korea.

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摘 要

1963年부터 中部地方에 큰 被害를 주고 있는 麥類의 바이러스病을 同定하기 위하여 電子顯微鏡에 依한 바이러스粒子 觀察과 媒介虫을 利用한 傳染試驗 및 寄主範圍調査를 한 結果 正常粒子의 平均 크기는 길이가 300～370nm였고 폭이 57～60nm였다.

媒介虫은 애멸구 *Laodelphax striatellus* (Fallén)로서 虫體內 潛伏期間은 7～19日이였고 大部分 10日間이였다.

Host range는 보리, 옥수수, 밀, Rye맥, 귀리 等으로 옥수수 외의 寄主植物들은 圃場에서도 發病이 되고 있다. 그러므로 中部地方에서 發生되고있는 麥類 바이러스病은 大部分이 北地모자익바이러스病으로 매년 10% 內外의 感染率을 보여주고 있으며 春播麥에서는 100% 感染 될때도 있다.