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Current Status of Researches on Mycoplasmal Diseases of Plants in Korea

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韓國의 植物마이코플라즈마病에 대한 研究動向

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INTRODUCTION

Diseases caused by, or associated with mycoplasma-like organisms (MLO), are most prevalent and of greater economic importance in tropical and subtropical regions where environmental conditions favor multiplication of the disease and vector activity. Korea, because of its long winter, which is unfavorable for both host plants and insect vector activity, may not be an ideal place for plant mycoplasmas to proliferate. Thus, in Korea, economically inportant plant mycoplasma diseases are more or less confined to perennial plants and vegetatively propagated plants, although mycoplasma disease has occasionally been found in annual crops grown from true seeds (2,4,11) (Table 1.)

Of a number of plant mycoplasma diseases occurring in Korea, jujube (*Zizyphus jujuba*) witches'-broom, paulownia (*Paulownia tomentosa*) witches'-broom and mulberry (*Morus* spp.) dwarf are among the most important, causing heavy losses to the host trees throughout the country and often devastating large tracts. Today, mycoplasma diseases have become the major factor limiting the successful cultivation of jujube, paulownia and mulberry in Korea. Our research effort has, therefore, been centered on the transmission and control of these three diseases. This paper outlines the disease situation, current status of research and control efforts directed toward these mycoplasma diseases in Korea.

1. Jujube witches'-broom

Jujube is an economically important fruit crop of Korea and has long been cultivated throughout the country. The outbreak of jujube witches'-broom (JWB) was first noted in Korea around 1950. The disease has since spread progressively throughout the country, causing extensive mortality in both isolated areas and large tracts. It is estimated that approximately 70-80% of the jujube trees in Korea have been killed or are infected with witches'-broom.

Jujube witches'-broom has long been assumed to be a virus disease (5, 10). In 1973, however, mycoplasma-like bodies were found to be associated with the disease (18, 30). Observed with the electron microscope, these mycoplasmal bodies are pleomorphic, and delimited by a single unit membrane. Rounded forms predominate but they range in shape from small round bodies to elongated oval forms. They range from 125 to 970 nm in length and occur abundantly in the sieve elements of

Table 1. Plant mycoplasma diseases reported in Korea

Disease	Insect vector	Host plant
Jujube witches' broom	*Hishimonus sellatus*	Jujube, mulberry, periwinkle, *Ligustrum obtusifolium*
Mulberry dwarf	*Hishimonus sellatus*	mulberry, jujube, periwinkle, white clover, red clover, Ladino clover, Chinese milk vetch
Paulownia witches' broom	*Cyrtopeltis tenuis*	*Paulownia tomentosa*
Ligustrum witches' broom	*Hishimonus sellatus*	*Ligustrum obtusifolium*
Sumach witches' broom	Not determined	sumach
Bush clover witches' broom	"	bush clover
Sweet potato witches' broom	"	sweet potato
Onion yellow dwarf	"	onion
Bupleurum witches' broom	"	*Bupleurum falcatum*
Cnidium witches' broom	"	*Cnidium officinale*
Plantago witches' broom	"	*Plantago asiatica*

'fected trees. No mycoplasma-like bodies have ever een observed in the phloem of healthy trees. [ycoplasma etiology of this disease has been ırther supported by therapeutic treatments with xytetracycline (17).

Fluorescence microscopy using flourochromes ·API (4'-6-diamidino-2-phenylindole 2HCl) and erberine sulfate has been very effective for rapid etection of mycoplasmal infection in jujube. Leaf etiole or young stem sections from infected jujube roduced distinct fluorescence in the phloem when :ained with either DAPI or berberine sulfate (1,6), /hile such fluorescence was absent in the phloem f healthy plants. Mycoplasmas were most abundant ı petioles of highly symptomatic twigs in brooms nd rarely found in nonsymptomatic portions of iseased trees (24). Fluorescence microscopy has lso shown that the quantity of mycoplasma apprently corresponds to the severity of symptoms 24).

Transmission studies of jujube witches' broom ıycoplasma (JWBM) have shown that the disease gent is transmitted by rhombic marked leafhopper *Hishimonus sellatus*) in Korea (20). Jujube seedlngs inoculated with infective rhombic marked eafhopper developed smaller, chlorotic leaves 40-60 days after inoculation. Electron microscopy of midveins and petioles of the infected jujube seedlings revealed the presence of numerous mycoplasma-like bodies in phloem tissues. In China, the disease was found to be transmitted by *Hishimonus chinensis* (8).

JWBM has also been transmitted to periwinkle (*Catharanthus roseus*) by rhombic marked leafhopper. Infected periwinkle developed yellows symptoms 25-38 days after inoculation with infective rhombic marked leafhoppers and was colonized with mycoplasma-like bodies (20). Rhombic marked leafhopper has long been known as the insect vector of mulberry dwarf in Korea and Japan (3,7). The fact that both judube witches'-broom and mulberry dwarf (MD) are transmitted by the same insect vector led to the investigation of possible interrelatedness between the two diseases. Mulberry seedlings inoculated with JWBM by using rhombie marked leafhopper developed typical mulberry dwarf symptoms. In the same way, mulberry dwarf mycoplasma (MDM) has been transmitted to jujube seedlings by rhombic marked leafhopper as was evidenced by the presence of the mycoplasma-like bodies in the infected jujube seedlings. Thus, it is assumed that both jujube witches'-broom and mulberry dwarf are caused by the same species of mycoplasma (22).

One of the common means of propagating the jujube is by vegetative root sprouts or "suckers". Therefore suckers from the infected trees are important means of spreading the disease. JWM is also readily transmitted by scion grafting and budding. The percentage of successful transmission is markedly reduced when diseased scions are cut and grafted in the spring as opposed to using similar scion wood collected the previous fall and kept in cold storage until grafting the following spring. Apparently the mycoplasma in above-ground portions of diseased trees are inactivated or killed by low winter temperatures (9). There is no evidence to indicate its transmission through seed or soil (15).

Complete remission of symptoms of witches'-broom was attained by trunk or root injection with oxytetracycline (OTC). For trees with 10-15 cm in DBH, a single injection with 1-2 g of OTC dissolved in 500 ml of water was effective in suppressing symptom development for 3 years. The time required for injecting 500 ml of OTC solution into diseased trees varied somewhat according to individual trees and the time of the year. In May, it required 1-2 days to inject 500 ml of the solution by gravity flow method (17). More recently, post harvest (Sept. - Oct.) treatments with OTC has been shown to be more effective than spring treatments (unpublished data). Currently, trunk injection of OTC is extensively practiced for the control of jujube witches'-broom in Korea and saves many thousands of valuable jujube trees every year. Along with trunk injection of OTC, preventive measures such as (1) propagation of suckers from disease-free areas, (2) use of both mycoplasma-free root stocks (obtainable by planting true seeds) and scions for producing disease free grafted seedlings, and (3) control of insect vectors etc. are strongly recommended to prevent the spread of the disease. Breeding of jujube trees resistant to witches'-broom is underway.

2. Paulownia witches'-broom

Paulownia witches'-broom (PWB) is undoubtedly the most important disease of paulownia in Korea causing severe damage in plantations throughout the country. It is common to find plantations with several thousand paulownia trees completely devastated by the disease. The disease poses a serious threat to the successful cultivation of this profitable tree species in Korea. There is no record as to exactly when the disease first occurred in Korea, but it may have existed for a considerably long time. The disease is also known to be wide spread in Japan, continental China, and Taiwan causing heavy losses to paulownias.

Most mycoplasma diseases of plants are transmitted by leafhoppers. Paulownia witches'-broom, however, was found to be transmitted by tobacco leaf bug (*Cyrtopeltis tenuis*) in Korea (13, 14) and China (8). Recently, brown-marmorated stink bug (*Halyomorpha mista*) was also shown to transmit the disease in Japan (26, 27).

The possibility of transmission of the disease by other than tobacco leaf bug and brown-marmorated stink bug also needs to be investigated since several species of sucking mouth type insects are commonly found to feed on diseased paulownias. Paulownias are propagated by both seed and vegetatively by root cuttings, and the disease is commonly spread by root cuttings from infected trees. There is no evidence to indicate its transmission through seed or soil.

Histochemical studies with fluorochromes DAPI, berberine sulfate and Dienes' stains have shown that the first two were effective for detecting mycoplasmal infections in Paulownia (25).

La and Shin (19) have shown that basal trunk or root collar injection with 0.2% OTC solution (2 liters per tree with 20 cm in DBH) is effective for suppressing the symptoms of paulownia witches' broom. Most of the paulownia trees failed to show symptoms for at least 2 years after treatments. The following integrated measures are currently being practiced and/or are recommended for the control of this disease. (1) Individual diseased trees the field should be injected with a solution of :ytetracycline at the initial stage of symptom pearance. Otherwise, the diseased trees should rogued from the field as soon as they are detec- d since infected trees serve as important sources infection. (2) Propagation of seedlings by true eds rather than root-cuttings is strongly recom- ended. Vegetative propagation by root cuttings en from healthy appearing trees is not recom- ended because of the potential danger of latent fection. (3) Seedlings should be raised in disease- e areas. Those raised in or near by infected ares, rticularly those propagated by root cuttings, unsafe and should be avoided for planting. (4) adication of insect vectors in the nursery by use insecticides is recommended to prevent early ge infection. In field plantings, however, control the insect vector by insecticides is considered be impractical for preventing natural spread of e disease.

Intercontinental spread of the disease is possible rough export of diseased root cuttings. Therefore, ecaution should be taken in the shipment or change of living plant material other than seed. ıfortunately, species of paulownia exhibiting ımunity or resistance to the disease have not been und. However, continuing efforts are being ade in the search for disease resistant clones.

3. Mulberry dwarf

Mulberry dwarf (MD) is the most important sease of mulberry in Korea and is of considerable onomic importance. More than 1.5 million fected mulberry trees were rogued out in Sangju unty alone in 1978. The first record of mulberry varf in Korea appeared in 1916 (3), although it presumed that the disease may have existed long fore this.

The disease is transmitted by rhombic marked ıfhopper in Korea. In Japan, the disease is known be transmitted by both rhombic marked leafho- er and false rhombic marked leafhopper (*Hishim- oides sellatiformis*), the former being the princi- l vector (7). Transmission of the MD by false rhombic marked leafhopper has not been deter- mined in Korea. As stated earlier, jujube witches'- broom is also transmitted by *H. sellatus*, suggesting the possible relationship between mulberry dwarf and jujube witches'-broom. Cross inoculation experiments confirmed that both MD and JWB are caused by the same mycoplasma.

In host range studies, the disease has been tran- smitted experimentally to periwinkle, red clover, white clover, Ladino clover and milk vetch by *H. sellatus* (12). Mulberry is propagated primarily by grafting, and scions from infected mulberry are an important reservoir for spread of the disease. The disease is not transmitted through seed or soil.

Both fluorochromes DAPI and berberine sulfate were effective for histochemical detection of mycoplasmal infections in mulberry and periwinkle. Dienes' stains was also effective for detecting MDM infections in mulberry and periwinkle (1).

Complete remission of dwarf symptoms was obtained when 100 ml of 0.5% OTC solution was injected into the root crown of mulberry tree. Mullberry varieties currently being cultivated have a bush type appearance with no main stem or trunk. Therefore, it is necessary to make the injection into the root crown. Foliar sprays and soil drenching with solutions of OTC were not effective for con- trolling the disease. More recently, post harvest treatment (Sept.-Oct.) of OTC solutions was found to be more effective than spring or summer treat- ment (unpublished data). Planting of disease-free seedlings, eradication of dwarf diseased mulberry, and vector control with insecticides are the principal measures currently practiced to prevent spread of the disease. It appears that most of the mulberry varieties now under cultivation are susceptible to the disease.

Conclusion

Although progress has been made in our under- standing of plant mycoplasma disease in Korea, it is still limited in scope and increased research is needed in our efforts to control these important diseases. In view of the economic importance of the mycoplasmal diseases of jujube, paulownia, and mulberry, research in the following areas are urgently needed:

1. Studies on the interrelationship of various mycoplasmas occurring in Korea and other areas and their host ranges. This information would provide a sound ecological basis for control.
2. Breeding for disease resistant or tolerant varieties of jujube, paulownias and mulberry. High priority should be given to this concept, which may offer the only method for practical control of mycoplasmal diseases of these trees.
3. The interaction of mycoplasmas with other disease causing agents should also be determined. There is the possibility that mycoplasmas may serve as antagonists to other pathogens. They also might predispose trees to other diseases.

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