Characteristice of the Resistance of Pinus armandii var. amamiana, an Endangered Pine Species in Japan, to Pine Wilt Disease after a Pin Sawfly Outbreak

Characteristics of the Resistance of *Pinus armandii* var. *amamiana*, an Endangered Pine Species in Japan, to Pine Wilt Disease

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Abstract - Pine wilt disease (PWD) was suggested as a mortality factor of *Pinus armandii* var. *amamiana* (PAA), an endangered pine species in Japan, from field surveys of the pathogenic nematode, *Bursaphelenchus xylophilus*, in dying and dead PAA trees and from a field inoculation test with the nematode. Disease development in nematode-inoculated PAA trees was slow and progressed gradually from the inoculated branch to the trunk, thereby demonstrating partial resistance of PAA to PWD.

I. Introduction

Pinus armandii var. *amamiana* (PAA) is an endangered pine species whose natural habitat is restricted to Yaku-shima and Tanega-shima Islands, southwestern Japan [1, 2, 3]. Pine wilt disease (PWD) might become a contributory factor in extinction of PAA because mass mortality related to needle blight, one of the typical symptoms of PWD, has occurred in its natural habitats [4]. In contrast, some researchers have speculated that PAA may be less susceptible to PWD, based on observations that PAA trees survived after bad epidemics of PWD in co-occurring *P. thunbergii* trees. Thus it is important to clarify the extent of PAA resistance to PWD in order to prepare a plan for conservation of PAA.

II. Survey of Pine Wilt Disease Infection

PWD is caused by the pinewood nematode, *Bursaphelenchus xylophilus* [5], which is mainly vectored by the Japanese pine sawyer, *Monochamus alternatus* [6, 7]. We tried to collect evidence of PWD infection in dying and dead PAA trees in the field. Feeding marks of the insect vector, *M. alternatus*, were commonly found on the twigs of PAA trees, suggesting that the insect-borne nematodes were frequently provided opportunities to enter PAA trees. We detected *B. xylophilus* in 5 out of 7 weakened or newly-dead PAA trees in the field [8]. These results suggest that PWD can cause mortality in PAA trees under field conditions.

III. Field Inoculation Test

Field-grown trees of PAA and *P. thunbergii* planted in the Experimental Forest of Kyushu Research Center, FFPRI, were selected and then inoculated with *B. xylophilus*. In the susceptible *P. thunbergii* trees, disease symptoms occurred simultaneously and developed quickly, which is typical of inoculation tests with *B. xylophilus*. In PAA trees, however, disease development was considerably different from that in *P. thunbergii* [9]. In PAA symptoms first occurred at the inoculation points and then slowly spread to the whole tree over a relatively long time period. Cessation of oleoresin flow, that is commonly known to be the first symptom in the development of PWD, occurred in the course of spread of foliage discoloration in PAA trees. These results suggest that PWD development in PAA trees progresses not systemically but gradually; progressing from the infected branches to the trunk and other branches. The occurrence of disease symptoms was delayed when the number of inoculated nematodes was small, but all PAA trees inoculated with nematodes eventually died irrespective of the inoculation density.

IV. Progressive Partial Death in PAA Trees

Some field-grown PAA trees had branches that could be divided into two sections, based on the time since death; sections on the top side that had died some time ago, and newly dead sections on the trunk side. This evidence suggests that the area affected by PWD in PAA trees spreads from the infected site towards the trunk over a period of several years.

V. Conclusions

The level of mortality of PAA trees in the nematode inoculation test was not different from that of susceptible *P. thunbergii* trees. However, the delayed disease development in PAA trees demonstrated some resistance to PWD. The progressive partial death observed in large PAA trees growing in the field resulted from delayed disease development. Due to this kind of resistance, field-grown PAA trees should have a chance to recover from PWD infection. Consequently, we conclude that PAA is less susceptible to PWD than *P. thunbergii*. Nevertheless, appropriate protection measures against PWD are still

needed to avoid further loss of field populations of this endangered species.

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