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# Enhancement of Growth and Survival of *Populus*alba × P. glandulosa Cuttings Inoculated with Ectomycorrhizal Fungus, *Pisolithus tinctorius*under Fumigated Nursery Condition

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# 모래밭버섯 萬根萬의 人工接種에 依한 포플러 揷木苗의 生長促進 및 活着率 増進'

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### ABSTRACT

Populus alba  $\times P$ . glandulosa cuttings in nursery bed were inoculated with mycelium of ectomycorrhizal fungus, Pisolithus tinctorius (Pt) to evaluate effectiveness of the fungus in growth stimulation of poplar. Pt was cultured in 1l glass bottles with vermiculite-peatmoss mixture moistened with modified Melin-Norkrans' solution. The nursery bed was arranged for microplots of  $l \times 2m$  in size and fumigated with methyl bromide before inoculation and cutting placement. Fifty cuttings were placed in each microplot and two treatments (fumigation only and fumigation plus Pt inoculation) were replicated three times.

At the end of the first growing season, inoculated plants grew 19% faster in height and produced 49% more dry weight (above-ground portion) than uninoculated plants. Survival rate of inoculated cuttings was also improved by 20% over that of uninoculated cuttings. Inoculated cuttings developed abundant fine root system with golden brown zigzag tips. In the middle of September a sporocarp of Pt was produced from an inoculated plot, suggesting successful establishment of mycorrhiza between poplar and Pt fungus.

Key words: ectomycorrhizae; Pisolithus tinctorius; Populus alba x P. glandulosa; inoculation.

### 要 約

外生菌根을 形成하는 모래발 버섯菌(Pisolithus tinctorius) (Pt)을 포플러에 人工接種하여 寄主의 生長促進과 活着率 增進效果를 調査하였다. 蛭石, 泥炭이끼, MMN培地(Modified Melin-Norkrans)를 添加한 1ℓ 유리 容器에 Pt 菌을 大量으로 培養하여 Methyl bromide(50g/m²)로 煎蒸한 苗圃土壤에 混合하고, 은수원사시(Populus alba×P. glandulosa)를 挿木하였다. 1×2m 크기의 挿木床에 50個의 挿穗를 使用했으며, 挿木當年 가을에 樹高, 乾重量(地上部), 活着率을 調査하였다. Pt 로 接種된 苗木은 接種안된 苗木보다 樹高生長에서 19% 더 빨리 자랐으며, 地上部의 乾重量에서 49% 더 많았다. 挿木 活着率도 接種된 苗木

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이 接種안된 苗木보다 20% 增加하였다. 接種된 苗木에는 Pt菌의 典型的인 黃褐色을 띤 細根이 많이 發達 했으며, 교불교불한 root tip 形態를 보였다. 接種된 挿木床에서 9月 中旬에 Pt菌의 子實體가 生産되어 서,接種과 菌根形成이 成功的이었음을 立證해 주었다.

### INTRODUCTION

It has been well documented that forest trees as well as agronomic plants form mycorrhizae with certain soil fungi and that host plants are benefited by this unique symbiosis through enhanced nutrient absorption, determent of root pathogens, and increase in host resistance to drought, high soil temperature and acidity (Harley and Smith, 1983). Recent advancement in the application of mycorrhizal research to agriculture has been remarkable (Molina, 1985). Particularly in forestry an extensive research on an ectomycorrhizal fungus, *Pisolithus tinctorius*, has resulted in commercial production of this fungal inoculum and of appropriate machinery for mechanized inoculation to bare-root nurseries (Cordell, 1985).

The potential of *Pisolithus tinctorius* (Pt) for growth stimulation of host plants has been demonstrated in U.S.A. by Marx *et al.* (1982) and in Korea using four pine species in nurseny conditions (Lee and Koo, 1983), in greenhouse (Lee, 1984) and with containerized oak seedlings (Oh, 1984). The objective of present study was to test the effectiveness of Pt in growth enhancement and survival of poplar cuttings in nursery condition.

## MATERIALS AND METHODS

Pisolithus tinctorius (Pt) #250 was obtained as mycelium in tube slants from the Institute for Mycorrhizal Research and Development (USDA Forest Service) in Athens, Georgia, U.S.A. and cultured for about four months in glass bottles containing vermiculite, peat moss and Modified Melin-Norkrans' (MMN) solution as described in Marx and Bryan(1975), and Koo et al. (1982).

In early spring nursery bed was prepared for 1 x 2m microplots. The entire area of nursery bed was

covered with an air-tight canvas tent and fumigated with methyl bromide  $(50g/m^2 \text{ surface})$  for three days. After ten days of gas escape from the fumigated soil, 2l of Pt inoculum (vermiculite-mycelium mixture) was added to each microplot  $(1 \times 2m)$  for inoculation treatment. For control treatment, same amount of vermiculite without mycelium was added, and the inoculum was mixed thoroughly with top soil. Fifty *Populus alba*  $\times$  *P. glandulosa* cuttings (25cm long) prepared from a single clone were placed in each microplot and replicated three times.

At the end of the first growing season, height growth and survival of cuttings were measured in the field. Above-ground biomass was determined after air-drying the harvested shoots. Survival was defined as cuttings grown to acceptable size at the end of the first growing season.

### RESULTS AND DISCUSSION

Inoculated cutting developed abundant fine root system with golden brown zigzag tips, while uninoculated plants had relatively short and straight fine roots with light brown color (Photos 1 and 2). In mid-September a sporocarp of Pt was produced from an inoculated plot, suggesting successful establishment of mycorrhiza between poplar and Pt fungus.

Table 1 shows height growth, dry weight, and survival rate of *Populus alba*  $\times$  *P. glandulosa* cuttings inoculated with *Pisolithus tinctorius* (Pt). Height growth and survival of the cuttings were relatively poor regardless of the treatment, because cuttings stored in a cold room were placed in the nursery during second week of May and were exposed to dry months after cutting. In average inoculated cuttings grew 19% faster in height than uninoculated cuttings (105 versus 125cm). The effect of inoculation was not shown in Block II, probably

due to inadequate conditions for successful mycorrhizal formation. Dry weight of inoculated cuttings increased by 49% over uninoculated cuttings (23.1 versus 34.3g per tree). Particularly in Block III inoculated cuttings were more than two times heavier than uninoculated cuttings (26.0 versus 55.8g per cutting). This experiment suggested that Pt had potential to stimulate growth of poplar



Photo 1. Populus alba × P. glandulosa roots in fumigated plot



Photo 2. Populus alba × P. glandulosa roots in Ptinoculated plot

Table 1. Height growth(cm), dry weight(g/plant), and survival rate(%) at the end of the first growing season of Populus alba × P. glandulosa cuttings in the nursery inoculated with ectomycorrhizal fungus, Pisolithus tinctorius (Pt). Each number is an average of 50 cuttings per microplot of 1 × 2m.

Treatment -	Block		<b></b>	
	I	П	III	Average
(Height)	Unit : cm			
Fumigation	108	96	110	105
Pt inoculation	133	95	148	125
(Dry Weight)	Unit: g/plant			
Fumigation	24.2	18.9	26.0	23.1
Pt inoculation	30.3	16.8	55.8	34.3
(Survival Rate)	Unit:%			
Fumigation	38	60	64	54
Pt inoculation	62	76	56	65

host plants as much as the case with pine seedlings shown by Lee and Koo (1983). Survival rate of inoculated cuttings was also improved by 20% over that of uninoculated cuttings (54 versus 65%).

The present experiment did not include the treatment of natural mycorrhizal fungi in unfumigated soil, but tried to compare the effect of Pt after soil fumigation. However, fumigated plots were gradually invaded by natural fungi, with natural mycorrhizae being formed during the late stage of the present experimental period.

Pt has been reported to have a worldwide distribution and to become mycorrhizal with over 40 woody plants (Marx, 1977). The same fungus was reported to occur naturally in Korea and its potential for mycorrhizal inoculum to pines was also tested (Lee and Koo, 1984). They showed that American strain of Pt was better source of inoculum than Korean strain. The same American Pt has been proved to stimulate dry weight increase of four pine species by more than two times (Lee and Koo, 1983; Koo et al., 1982). The present experiment demonstrated the benefit of Pt to the growth and survival of host plant, Populus alba × P. glandulosa. The author has not been able to find reports on the effect of Pt inoculotion on the growth of poplar species, even though there are some reports on the formation of endomycorrhizae in hybrid poplars (Walker, 1979; McNabb and Walker, 1979). Populus alba × P. glandulosa hybrid developed by Institute of Forest Genetics in Korea has been reported to adapt successfully to relatively poor soils in upper hill where moisture and nutrients are often limited for good growth (Noh and Lee, 1983). Successful mycorrhizal formation of this hybrid with Pt suggests that range of site conditions for this hybrid could be broadened through manipulation of Pt which is known to successfully adapt wide range of adverse soil conditions, such as drought, low pH, high soil temperature, and soil toxins (Marx, 1980; Berry and Marx, 1978).

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76

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