

Taxonomic Distribution of Ecto- and Endomycorrhizae among Woody Species in Korea¹

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韓國의 木本植物의 外生 및 内生菌根에 關한 分類學的 分布 調査¹

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ABSTRACT

Taxonomic distribution of ecto- and endomycorrhizae among woody species growing in Korea was studied and their distribution was compared with foreign literature. Most of root samples were collected from Central Branch Station of Forest Research Institute in Gwangneung, Gyeonggi-do. A total of 32 families, 63 genera, 102 species were examined. Ectomycorrhizae were observed in the following 13 genera of Pinaceae (*Pinus*, *Larix*, *Picea*, *Abies*), Salicaceae (*Populus*, *Salix*), Betulaceae (*Alnus*, *Betula*, *Carpinus*, *Corylus*), Fagaceae (*Quercus*, *Castanea*), and Tiliaceae (*Tilia*). Endomycorrhizae (vesicular-arbuscular) were observed in *Populus* and the rest of the 49 genera. *Rhus* was the only genus which did not have either ecto- or endomycorrhizae, but needs further study to confirm it. *Populus* was the only genus with both ecto- and endomycorrhizae, while foreign literature listed following genera having both mycorrhizal types: *Juniperus*, *Cupressus*, *Populus*, *Salix*, *Juglans*, *Alnus*, *Ulmus*, *Malus*, *Pyrus*, and *Tilia*. *Juniperus*, *Ulmus*, *Pyrus*, and *Acer* which were reported to have facultative ectomycorrhizae were free of ectomycorrhizae. Some morphological characteristics of endomycorrhizae are shown in photographs and discussed.

Key Words: ectomycorrhizae, endomycorrhizae, taxonomy of mycorrhizae.

要 約

韓國에서 自生하는 木本植物의 外生菌根과 内生菌根의 分布를 調査하고 그 結果를 外國의 文獻과 比較하였다. 京畿道 光陵 林業試驗場에서 대부분의 뿌리 表本을 採取하였으며, 모두 32科, 63屬, 102種을 觀察하였다. 外生菌根은 소나무科(*Pinus*, *Larix*, *Picea*, *Abies*), 버드나무科(*Populus*, *Salix*), 자작나무科(*Alnus*, *Betula*, *Carpinus*, *Corylus*), 참나무科(*Quercus*, *Castanea*), 피나무科(*Tilia*)의 13屬에서 觀察되었으며, 内生菌根은 위의 13屬中 *Populus*와 나머지 49屬에서 觀察되었다. 外生이나 内生菌根을 전혀 가지지 않은 屬은 *Rhus* 뿐이었으나, 이 屬은 좀더 자세한 觀察을 要한다. 外生과 内生을 함께 가지고 있는 屬은 *Populus* 뿐이었으나, 外國文獻에는 그밖에도 *Juniperus*, *Cupressus*, *Populus*, *Salix*, *Juglans*, *Alnus*, *Ulmus*, *Malus*, *Pyrus*, *Tilia* 등이 收錄되어 있다. 偶發적으로 外生菌根을 形成한다고 하는 *Juniperus*, *Ulmus*, *Pyrus*, *Acer* 등에서는 外生

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菌根을 觀察할 수 없었다. 몇 樹種의 内生菌根의 形態學的 特性을 顯微鏡 寫眞과 함께 說明하였다.

INTRODUCTION

Mycorrhizal association is a common characteristic of the most of the world's present species of vascular plants. Trappe(1977) estimated that about 95% of the present vascular plants belong to families that are characteristically mycorrhizal. Ectomycorrhizae are found mostly on woody plants, while endomycorrhizae are a common feature of many woody plants and most herbaceous plants including agronomic crops.

Information on the taxonomic distribution of mycorrhizal plants is an essential step toward understanding of roles of mycorrhizae in a complex forest ecosystem (Malloch and Malloch, 1981), a desert (Rose, 1981), and a semi-arid region (Reeves *et al.*, 1979). In addition, knowledge on the identification of mycorrhizal fungi in different forest types (Lee and Kim, 1983) and on tree host range of specific mycorrhizal fungi (Marx, 1977) will be applied to enhancement of forest productivity through manipulation of symbiosis between woody plants and fungi.

Lee *et al.*(1981) surveyed ectomycorrhizal distribution in one-hundred woody plants in Korea and found ectomycorrhizae in Pinaceae, Salicaceae, Betulaceae, Fagaceae, Ulmaceae, and Tiliaceae. They were unable to find ectomycorrhizae in Cupressaceae, Rosaceae, *Juglans*, and *Acer* which appeared to have facultative ectomycorrhizae. Occurrence of endomycorrhizae in woody plants in Korea has not been previously investigated.

This paper reports taxonomic distribution of endo- and ectomycorrhizae among woody plants growing in Korea and compares the results with foreign literature. Morphological characteristics of endomycorrhizae in some plants are also reported.

MATERIALS AND METHODS

Most of the plants used in this study were col-

lected from a part of experimental forests in Gwangneung, Gyeonggido, where the Central Branch Station of Forest Research Institute was located and has been maintained as an arboretum for indigenous and exotic species. Root samples were collected in September 1980 from the top 10 to 15 cm soil, and brought to the lab in a fixative (standard formalin, acetic acid, and alcohol solution). Ectomycorrhizae were observed under a dissecting microscope for external morphology and under a compound light microscope for internal morphology after hand sectioning. Criteria described by Wilcox (1968) were used for differentiation of mycorrhizal roots from non-mycorrhizal ones. Most of the data on ectomycorrhiza were published previously (Lee *et al.*, 1981) but some data on the previous paper were corrected and substantiated through further observations.

Endomycorrhizae (vesicular-arbuscular) were studied with a technique modified from Kormanik *et al.*(1980). Root samples were placed in test tubes with 10% KOH solution enough to cover the sample, and heated at 90°C for one hour in a water bath. In case an autoclave is available, the test tubes can be autoclaved at 15 psi for 10 min to achieve the same results as hot water bath. After KOH solution was poured off, root samples were rinsed well with water and alkaline H₂O₂ was added to the tube. Alkaline H₂O₂ solution was made daily as needed by adding 3 ml of NH₄OH to 30 ml of 10% H₂O₂ and 567 ml of water. The samples were left in the H₂O₂ solution at room temperature for 10 to 60 minutes until roots were bleached. After rinsing the samples with water, 1% HCl solution was added to the tubes and left for 3-4 minutes. Then the HCl solution was poured off (do not rinse in this case) and lactophenol staining solution containing 0.01% trypan blue was added. The lactophenol staining solution was a mixture of equal parts of phenol (300g or 250ml), lactic acid (250ml), glycerin (250ml) and water (300ml). Two percent trypan blue stock solution was made by

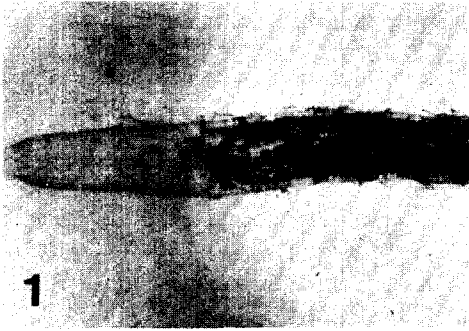


Fig. 1. Vesicular-arbuscular endomycorrhizal development in *Ulmus davidiana* var. *japonica*. Fungal structures were heavily stained by trypan blue and shown as dark color. Mycorrhizal development is limited to a few millimeters behind the root tip.

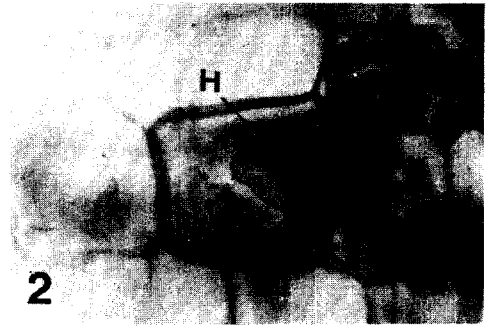


Fig. 2. Intracellular hyphae (H) in the cortex of *Acer palmatum*. Hyphal development had a tendency to be compartmentalized within individual cells in this host.

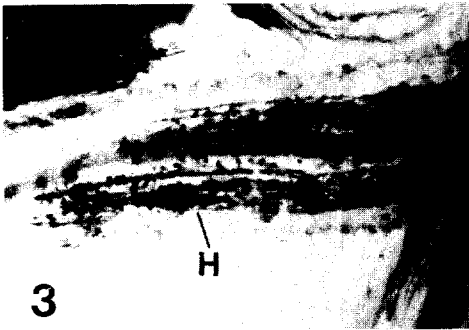


Fig. 3. Hyphal strands (H) developed along the long axis of the root of *Fraxinus mandshurica*. Arbuscules are attached laterally to the hyphae and shown here as small dots.

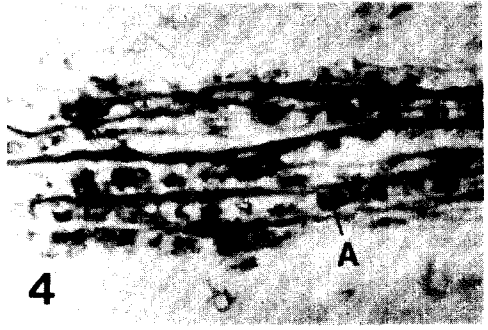


Fig. 4. Abundant arbuscules (A) shown as clumps along the running hyphae in *F. mandshurica*.



Fig. 5. A closer view of functioning young arbuscules which occupy almost entire space of individual cells of *F. mandshurica*. Arbuscules are formed by fine branching from primary hyphae.

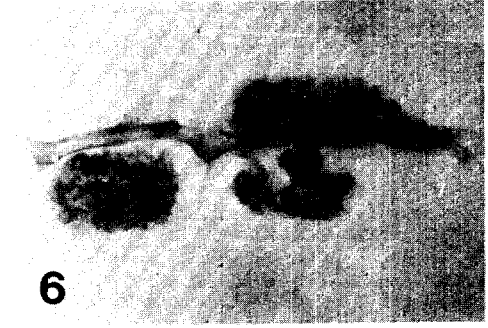


Fig. 6. Disintegrating old arbuscules in the same species. Unfunctioning old arbuscules are digested and their contents absorbed by the host cells.

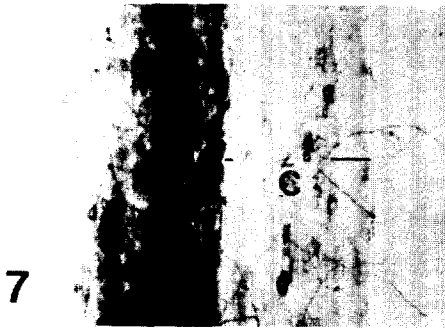


Fig. 7. Growth of arbuscules are generally restricted to near the endodermis (*Liriodendron tulipifera*). See also Fig. 3 for the arbuscules near the endodermis of squashed root samples. C indicates cortical region.

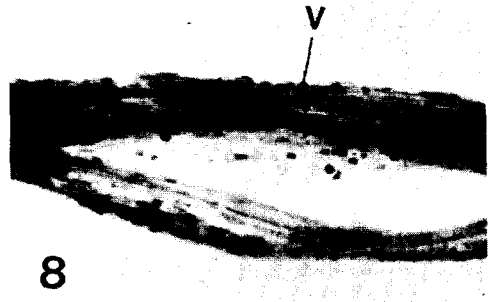


Fig. 8. Formation of abundant vesicles (V) in *Ulmus pumila*.

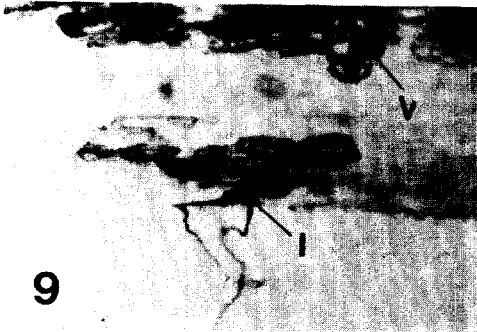


Fig. 9. An infection point (I) on the surface of root. This infection was probably initiated by a germinating fungal spore in the nearby soil. Hyphal growth from this single infection can be easily recognized by isolated hyphal growth in this region. A kidney-shaped vesicle (V) is visible in the cortex (*Liriodendron tulipifera*).

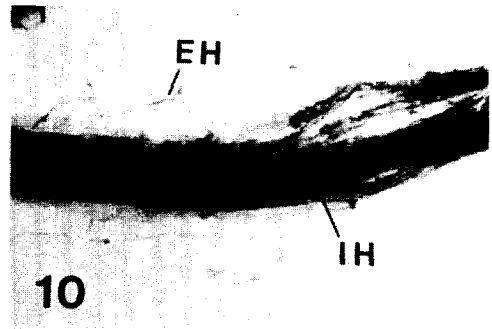


Fig. 10. Development of abundant external hyphae (EH) on the root surface of *Forsythia koreana*. These external hyphae grow into the soil and absorb soil nutrients probably more efficiently than root hairs. Also note heavy staining of internal hyphae (IH) in the cortical region of a squashed root sample.

dissolving 2g of trypan blue in 100ml of water and appropriate amount of the stock solution was used to make 0.01% trypan blue in lactophenol staining solution. The staining solution with roots in test tubes was heated at 90°C for 10-60 minutes until the roots were satisfactorily stained. After proper staining, the root samples were transferred to a new lactophenol solution without trypan blue

to remove excess stain and to store for a maximum period of 4-6 weeks if needed. The stained roots were observed under a compound light microscope for fungal structure.

Taxonomic classification of host plants was based on a system by Lee (1979). A total of 32 families, 63 genera, 102 species were examined.

RESULTS AND DISCUSSION

During the present study on endomycorrhizae, we reviewed some work on ectomycorrhizae reported previously (Lee *et al.*, 1981) and found that *Celtis jessoensis* in Ulmaceae was free of ectomycorrhizae. Two other genera of Ulmaceae, *Ulmus* and *Zelkova*, also did not have ectomycorrhizae. Therefore, Ulmaceae may at present time be considered to have facultative ectomycorrhizae (Meyer, 1973) rather than to have ectomycorrhizae in common.

In contrast to ectomycorrhizae, endomycorrhizae can not be identified by naked eyes, but need to be observed under a high power microscope after a proper staining. Fig. 1 shows vesicular-arbuscular (V-A) endomycorrhizal development near a root tip in *Ulmus davidiana* var. *japonica*. Typically endomycorrhizae develop along the elongating roots, but their development is limited to a few millimeters behind the root tip (Fig. 1). One of the major features of endomycorrhizae is hyphal penetration of root cortical cells (Fig. 2). In *Acer palmatum* fungal hyphae have a tendency to be compartmentalized within individual cells, while *Fraxinus mandshurica* has abundant hyphal strands running along the long axis of roots (Fig. 3). Arbuscules are developed along these hyphae (Fig. 3) and are attached laterally to the hyphae as small clumps (Fig. 4). Arbuscules are finely branched hyphae with smaller diameter than primary hyphae from which arbuscules are originated (Fig. 5). Arbuscules are considered to function as haustoria which absorb nutrients from the host. However, they disintegrate later (Fig. 6) and their contents are digested and absorbed by the host cells. Arbuscules are generally restricted near the endodermis, and fungal hyphae do not penetrate the stele (Fig. 7).

Formation of vesicles is another characteristic of vesicular-arbuscular (VA) endomycorrhizae formed by non-septate fungi. Fig. 8 shows abundant vesicles in *Ulmus pumila*. Vesicles are formed within cortical cells or near root surface by swelling

of terminal hyphae and are believed to contain storage materials. Infection of roots is usually initiated by fungal penetration of more than one hypha at various points along the root surface, and eventually results in interconnection and proliferation of hyphae in the cortex. In some cases, isolated infection point on the surface of epidermis is recognizable (Fig. 9). Fungal hyphae proliferate in the cortex and also grow from the root surface into the soil (Fig. 10), so that abundant hyphae around the root surface efficiently absorb nutrients in the soil. Presumably these extensive hyphae network may work better than root hairs in absorbing nutrients which are not readily available to the plants such as phosphorus.

Table 1 shows occurrence of endo- and ectomycorrhizae in the woody species in Korea. Data on ectomycorrhizae are mostly same as previous report (Lee *et al.*, 1981), except that *Corylus avellana* is ectomycorrhizal and *Morus alba* is non-ectomycorrhizal. Another difference is that as mentioned earlier *Celtis jessoensis* is non-ectomycorrhizal. Endomycorrhizae were found in most of the species except in exclusively ectomycorrhizal genera (*Pinus*, *Larix*, *Picea*, *Abies*, *Betula*, *Carpinus*, *Corylus*, *Quercus*, *Castanea*). In addition, *Salix*, *Tilia*, and *Alnus* which were reported in the literature to have both ecto and endo (Meyer, 1973; Rose, 1980) did not have endomycorrhizae. *Populus* had both ecto- and endomycorrhizae, which agreed with literature (Vozzo and Hacskaylo, 1974). *Rhus chinensis* was the only species which did not have either ecto- or endomycorrhizae, but should be studied further to confirm the present survey. Other species which are not mentioned above had endomycorrhizae.

Table 2 summarizes occurrence of ecto- and endomycorrhizae in 63 genera of woody plants in Korea. Meyer (1973) listed 8 genera to have both ecto- and endo-mycorrhizae: *Juniperus*, *Cupressus*, *Salix*, *Malus*, *Pyrus*, *Tilia*, *Eucalyptus*, and *Arbutus*. If information in Meyer (1973), Kormanik *et al.* (1977), Trappe (1979), Vozzo and Hacskaylo (1974), and Rose (1980) are combined, *Populus*, *Juglans*, *Alnus*, *Ulmus*, and some leguminous

Table 1. Occurrence of endo- and ecto-mycorrhizae in the selected woody species in Korea. Literature survey included Kormanik *et al.* (1977), Meyer (1973), Rose (1980), Trappe (1979), Vozzo and Hacskaylo (1974), and Wilcox (1982).

Family name	Species name		Endo.	Ecto.	In literature
Ginkgoaceae	<i>Ginkgo</i>	<i>biloba</i>	+	-	
Taxaceae	<i>Taxus</i>	<i>cuspidata</i>	+	-	
	<i>Cephalotaxus</i>	<i>koreana</i>	+	-	
Pinaceae	<i>Pinus</i>	<i>koraiensis</i>	-	+	Obligate and exclusive ectomycorrhizal family. Ectendomycorrhizae are common.
	<i>P.</i>	<i>thunbergii</i>	-	+	
	<i>P.</i>	<i>densiflora</i>	-	+	
	<i>P.</i>	<i>rigida</i>	-	+	
	<i>P.</i>	<i>taeda</i>	-	+	
	<i>P.</i>	<i>strobilus</i>	-	+	
	<i>P.</i>	<i>banksiana</i>	-	+	
	<i>P.</i>	<i>sylvestris</i>	-	+	
	<i>Larix</i>	<i>leptolepis</i>	-	+	
	<i>Picea</i>	<i>abies</i>	-	+	
	<i>P.</i>	<i>koraiensis</i>	-	+	
	<i>Abies</i>	<i>holophylla</i>	-	+	
	<i>A.</i>	<i>nephrolepis</i>	-	+	
	<i>A.</i>	<i>koreana</i>	-	+	
Taxodiaceae	<i>Cryptomeria</i>	<i>japonica</i>	+	-	
	<i>Metasequoia</i>	<i>glytostroboides</i>	+	-	
Cupressaceae	<i>Thuja</i>	<i>occidentalis</i>	+	-	
	<i>Chamaecyparis</i>	<i>pisifera</i>	+	-	
	<i>C.</i>	<i>obtusa</i>	+	-	
	<i>Juniperus</i>	<i>chinensis</i>	+	-	Facultative ecto.
	<i>J.</i>	<i>chinensis</i> var. <i>sargentii</i>	+	-	Both ecto and endo.
	<i>J.</i>	<i>rigida</i>	+	-	
Salicaceae	<i>Populus</i>	<i>euramericana</i>	+	+	Ecto family.
	<i>P.</i>	<i>tomentiglandulosa</i>	+	+	Both ecto and endo.
	<i>Salix</i>	<i>graciliglans</i>	-	+	Both ecto and endo (facultative ecto).
Juglandaceae	<i>Juglans</i>	<i>mandshurica</i>	+	-	
	<i>J.</i>	<i>sinensis</i>	+	-	Both ecto and endo.
Betulaceae	<i>Alnus</i>	<i>japonica</i>	-	+	Ecto family.
	<i>A.</i>	<i>hirsuta</i> var. <i>sibirica</i>	-	+	Both ecto and endo. Facultative ecto.
	<i>Betula</i>	<i>platyphylla</i>	-	+	
	<i>B.</i>	<i>platyphylla</i> var. <i>japonica</i>	-	+	
	<i>B.</i>	<i>verrucosa</i>	-	+	
	<i>Carpinus</i>	<i>laxiflora</i>	-	+	Ecto only.
	<i>C.</i>	<i>cordata</i>	-	+	
	<i>Corylus</i>	<i>avellana</i>	-	+	Facultative ecto.
Fagaceae	<i>Quercus</i>	<i>aliena</i>	-	+	Ecto family.
	<i>Q.</i>	<i>acutissima</i>	-	+	
	<i>Q.</i>	<i>grosseserrata</i>	-	+	
	<i>Q.</i>	<i>mongolica</i>	-	+	
	<i>Q.</i>	<i>rubra</i>	-	+	
	<i>Q.</i>	<i>serrata</i>	-	+	
	<i>Q.</i>	<i>variabilis</i>	-	+	
	<i>Castanea</i>	<i>crenata</i>	-	+	ecto.

Family name	Species name	Endo.	Ecto.	In literature	
Ulmaceae	<i>Celtis jessoensis</i>	+	-		
	<i>Ulmus davidiana</i>	+	-	Facultative ecto.	
	<i>U. pumila</i>	+	-	Both ecto and endo.	
	<i>Zelkova serrata</i>	+	-		
Moraceae	<i>Morus alba</i>	+	-		
Berberidaceae	<i>Berberis koreana</i>	+	-		
Cercidiphyllaceae	<i>Cercidiphyllum japonicum</i>	+	-		
Magnoliaceae	<i>Magnolia sieboldii</i>	+	-		
	<i>Liriodendron tulipifera</i>	+	-		
Leguminosae	<i>Amorpha fruticosa</i>	+	-		
	<i>Lespedeza maximowiczii</i>	+	-		
	<i>L. cyrtobotrya</i>	+	-		
	<i>Maackia amurensis</i>	+	-		
	<i>Robinia pseudoacacia</i>	+	-		
Platanaceae	<i>Platanus occidentalis</i>	+	-		
Rosaceae	<i>Crataegus pinnatifida</i>	+	-	Ecto.	
	<i>Malus baccata</i>	+	-	Both ecto and endo.	
	<i>Prunus armeniaca</i> var. <i>ansu</i>	+	-		
	<i>P. padus</i>	+	-		
	<i>Pyrus pyrifolia</i>	+	-	Both ecto and endo.	
	<i>Sorbaria sorbifolia</i> var. <i>stellipila</i>	+	-	(Facultative ecto)	
	<i>Spiraea salicifolia</i>	+	-		
	<i>S. prunifolia</i> var. <i>simpliciflora</i>	+	-		
	<i>S. pubescens</i>	+	-		
	<i>Stephanandra incisa</i>	+	-		
	Saxifragaceae	<i>Philadelphus schrenckii</i>	+	-	
	Euphorbiaceae	<i>Securinega suffruticosa</i>	+	-	
Rutaceae	<i>Phellodendron amurense</i>	+	-		
	<i>Evodia daniellii</i>	+	-		
	<i>Zanthoxylum schinifolium</i>	+	-		
Anacardiaceae	<i>Rhus chinensis</i>	-	-		
Aceraceae	<i>Acer saccharum</i>	+	-	Facultative ecto.	
	<i>A. palmatum</i>	+	-		
	<i>A. saccharinum</i>	+	-		
	<i>A. ginnala</i>	+	-		
	<i>A. mono</i>	+	-		
	<i>A. triflorum</i>	+	-		
Staphyleaceae	<i>Staphylea bumalda</i>	+	-		
Tiliaceae	<i>Tilia amurensis</i>	-	+	Both ecto and endo.	
	<i>Tilia megaphylla</i>	-	+		
Elaeagnaceae	<i>Elaeagnus umbellata</i>	+	-		
Araliaceae	<i>Acanthopanax sessiliflorus</i>	+	-		
Cornaceae	<i>Cornus controversa</i>	+	-		
	<i>C. officinalis</i>	+	-		
	<i>C. kousa</i>	+	-		
Styracaceae	<i>Styrax japonica</i>	+	-		
Oleaceae	<i>Fraxinus mandshurica</i>	+	-		
	<i>F. rhynchophylla</i>	+	-		
	<i>Ligustrum obtusifolium</i>	+	-		
	<i>Forsythia koreana</i>	+	-		
	<i>Syringa dilatata</i>	+	-		

Family name	Species name		Endo.	Ecto.	In literature
Scrophulariaceae	<i>Paulownia</i>	<i>coreana</i>	+	-	
	<i>P.</i>	<i>tomentosa</i>	+	-	
Bignoniaceae	<i>Catalpa</i>	<i>bignonioides</i>	+	-	
Caprifoliaceae	<i>Viburnum</i>	<i>koreanum</i>	+	-	
	<i>Weigela</i>	<i>subsessilis</i>	+	-	

Table 2. Summary of occurrence of ectomycorrhizae and endomycorrhizae in 63 genera of woody plants in Korea. Literature surveyed was same as in Table 1.

Class	In the present survey	In literature
Ectomycorrhizae only	<i>Pinus, Larix, Picea, Abies, Salix, Alnus, Betula, Carpinus, Corylus, Quercus, Castanea, Tilia</i>	<i>Pinus, Larix, Picea, Abies, Betula, Carpinus, Corylus, Quercus, Castanea</i>
Endomycorrhizae only	All the genera listed in Table 1 except above 12 genera (ecto only), <i>Populus</i> , and <i>Rhus</i>	Many woody plants have endo.
Both ecto. and endo.	<i>Populus</i>	<i>Juniperus, Cupressus, Populus, Salix, Juglans, Alnus, Ulmus, Malus, Pyrus, Tilia, Eucalyptus, Arbutus</i> , and some leguminous woody plants
No mycorrhizal	<i>Rhus</i>	No specific trees listed as non-mycorrhizal
Facultative ecto.	Facultative nature not confirmed, but <i>Juniperus, Ulmus, Pyrus, Acer</i> were free of ecto, and instead had endo.	<i>Cupressus, Juniperus, Salix, Betula, Corylus, Alnus, Ulmus, Pyrus, Acer, Eucalyptus</i>

woody plants also have both ecto- and endomycorrhizae. Further study is needed to confirm above information. We were not able to find ectomycorrhizae in *Juniperus, Ulmus, Pyrus*, or *Acer* which were reported to have facultative ectomycorrhizae (Meyer, 1973). Above four genera and *Juglans* and *Malus* need to be studied to confirm ectomycorrhizae.

Present survey indicated that most woody plants had either ecto- or endomycorrhizae or both. It was clear to the authors that mycorrhizal association was ubiquitous in woody plants and suggests im-

portant roles in forest ecosystem. Mycorrhizae have been known to benefit hosts by aiding in absorption of inorganic nutrients, especially phosphorus, supplying hosts with growth-regulating substances, deterring root pathogens, decreasing soil toxicity, and increasing host plant resistance to drought and extreme soil temperatures (Slankis, 1974). Information on the taxonomic distribution of mycorrhizae can be applied to various disciplines of forestry, such as contribution of mycorrhizal fungi to maintenance of ecological balance in complex forest ecosystem (Ogawa, 1977), and growth stimulation

of host plants through artificial inoculation (Koo *et al.*, 1982).

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