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Endomycorrhizal Fungi identified on the Soils in Forest and Coast Areas

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산림 및 해안지역에서 발견된 내생균근

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ABSTRACT: The presence of endomycorrhizal fungi was examined on the soils collected from the followings; *Cryptomeria japonica* dominant forest (Wan San Park, Jun Ju city) and two coast areas (*Digitaria sanginalis* dominant; Sin Chang Ri, Young Il Kun and *Pragmited communis* dominant; Sap Kyo Cheon, A San). Six species in Endogonales were identified; *Glomus intraradices, G. occulum, G. clarum, Acaulospora bireticulata, Scutellospora aurigloba, and Sc. gilmorei.*

KEYWORDS: Mycorrhiza, Endomycorrhiza, *Glomus*, *Acaulospora*, *Scutellospora*.

Mycorrhizae, terming symbiotic associationbetween fungi and plant roots, are consisted of ectomycorrhiza and endomycorrhiza. Endomycorrhiza is different from ectomycorrhiza in infection modes of plant roots, and directly infects the plant roots at the cortical cells (Schenck, 82). Otherwise, ectomycorrhiza covers the plant roots, to form the mantle in the plant root. thus, mycorrhiza plays important roles in the plants. Ectomycorrhizae provide the phytohormone or unknown nutrients for plants (Schenck, 82). Endomycorrhizae are similar to N_2-fization bacteria in plant nutritions and convert inoganic from organic phosphate for plant roots (Jensen, 82). Therefore, little has been known for endomycorrhiza in Korea.

The application of endomycorrhiza is great in the agriculture: Supplying mineral nutrition and preventingplant disease by ecological niche (Hussey and Roncadrol, 82). Particularly, endomycorrhizae have been employed in the forage producing or small grain producing regions, where the industrial fertilization was economically unfeasable in the agriculture (Trappe, 81; Abbott *et al.*, 83). So the inoculum of mycorrhizal fungi was emphasized in the above regions, especially wheat or barley cultivated regions in the U.S., or wool producing areas in New Zealand (Hall, 85). Otherwise, the usage of endomycorrhizae has been also suggested in the areas abundant of the root diseases. Supposed that the mycorrhizal fungi colonize around the plant roots in the early stage, the pathogenic organisms did not have chance to infect the plant roots in the system of pathogen 'waiting game for plants'. Around the plant roots, the mycorrhizal fungi are supposed to clean up the plant extudate nutrients, which the plant secrets near the root soils and the pathogen fungi would utilize for germination. The mycorrhizal fungi also infect the plant roots in the curticle, and may plays important roles in plant immune systems (Sylvia and Sinclair, 83). Thus, the basic studies for endomycorrhiza have been emphasized but have not been done in Korean agriculture.

This experiment was first to identify the endomycorrhizal fungi from the soil samples collected from the various areas in Korea. The three

genera, including six species of endomycorrhizae were identified. We will also try to identify all VA-mycorrhizae, which might exist in Korea.

Materials and Methods

Collection of soil samples

For isolation of mycorrhizal spores (referred to "chlamydospores and azygospores" in this work), the samples of three different soils were collected. The roots of plants described in the below were confirmed in the soils before the soils were collected. The soils collected were carried into our laboratory in the polyethene bags, and stored in the room temperature. From October 1987 to August 1988, the slit soils referred 'A soils' were collected at the Park of Wan San in Jun Ju (Jun Puk), where *Cryptomeria japonica* (L. Fil.) D Don were dominant. On August of 1988, the clay soils referred 'B soils' were collected at *Phragmites communis* Trin dominant area at Sap Kyu Cheon (A-San Kun, Chung Nam), facing the artificial Lake of Sap Kyu Cheon. On June of both 1987 and 1988, the sand soils referred 'C soils' were collected at the *Digitaria sanguinalis* (L.) Scop. dominant area of Shin Chang Ri (Wol Seong Kun, Kyoung Puk), facing the Sea of Dong Hae.

Spore collections

For the isolation of spores, the sieving method described by Trappe (1982) and schenck (1982) was first employed. The result indicated that a lot of organic matter and only few spores were observed. The modified methods including the sieving and centrifuge methods (Hall, 1985) were employed. The twenty grams of soil samples was dissolved in 0.0818 M sodium hexametaphosphate at the 250 *ml* mass cylinders, and shaked for 15sec (McKenney and Lindsey, 87). The suspensor in the mass cylinder was poured in the sieves of 300, 212, 150, 90 and 38 μm. The 0.0818 M sodium hexametaphosphate was poured at the sedimented soils in the mass cylinder, again and shaked for 2 mins. The suspensor was collected by five different sized (mentioned in the above) sieves. The collected spores were treated with the 50% sucrose centrifuge, to remove the organic matters and the sands. The spores and the clay matters collected from the sieves were put in the 30 m*l* polyethene tube with the water and centrifuged at 3000rpm for five mins. The sedimented portion in the tube was shaked with 50% sucrose solution and centifuged at 3000rpm for five mins. The suspensor was collected, again, with the same sized sieve or poured in the filterpaper. The spores collected by the both methods were observed under the dissection microscope and picked up on the clean slide by the needle. The cover glass was fixed on the spore collected slides with the Miracle mounting solution (See Hall: 1985).

Obervations of spores

The spores collected by the modified methods were stored in the five cm (dia) petri dishes and picked at the slides when neccessary. The spores at the permenant slide made by Miracle mountings were observed and measured by compound microscopes (Olympus and Lietz) with the sizes of spore and thickness of spore wall layers. The identifications of spores were, at the level of genera, easily made by the description of Trappe's (1982) and Hall's (1985) slide guides. The identification of the endomycorrhizal species collected was absolutely based on Trappe's keys, which described the sizes and thickness of spores. The taxonomical names of six species collected in Korea followed the recent work done by Schenck and Perez (1988).

Identification of plants

The plants observed in the three areas (A, B and C) were identified by the Korean Plant flora described by Lee (1985) and Park (1980) for seed plants and Pteriophyta, respectively.

Results and Discussion

Spore collection

The A soil was employed for the experiment of spore collections, because very various spores were found in this soil. First, the sieving method described in the above resulted that so much organic matter and clay were included in the collecting petri dishes. To remove these materials, the centrifuge method was employed for the A soil, but few spores were found in this method. The modified method indicated in the above removed the organic matter and the heavy clay in this experiment. Also, this method for spore collections was applied

for B and C soils including the fine clay and sand, and completely remove the heavy materials.

The areas of soil collections

The area A (referred 'A soil') was the region of *Cryptomeria japonica* artificially planted, same aged (about 60 to 70 year old, estimated), and even distributed community. The understory plant systems at *Cryptomeria japonica* were considerably disturbed by the Jun Ju residents (Lee; 1988) and the typical dominant plants were *Persicaria pubescens* hara, *Preridium aquilium* var. *latiusculum* (Desv.) Underw., and *stephanandra incisa* Zabel. The slope of *Cryptomeria japonica* growing area at Wan San Public Park was so steep that these trees were expected to be difficult to grow in this area. Otherwise, the areas referred 'B and C soil' was only one species growing regions: The B area was located at the car-road on the bulwark (for sea water attacking) and only a species of *Phragmites commun* growing area near Sap Kyu Cheon. The C area was the sand soil facing the Dong Hae and only one species of the *Digitaria sanguinalis* growing soil. The two areas referred in the above were considered to be quite a simple community system as compared with the A area. The three areas selected for this experiment were consistent with the areas reported by the endomycorrhizal workers (Rothwell and Trappe, 1979; Schenck, 1982).

Identifications of endomycorrhizal fungi

The following species were identified as based on Trappe's made Synoptic keys with the size measurements and thickness of wall layers. The names of species selected by Trappe's were also reviewed with the descriptions of the references mentioned in each species.

1) *Acaulospora bireticulata* Rothwell & Trappe.

Rothwell & Trappe (1979) *Mycotaxon* **8:** 471-475, Schenck & Perez (1988) INVAM: 64, Hall (1984) VA mycorrhizal Fungi: 87, Trappe (1982) *Phytopathology* **72:** 1105, Walker (1981) *Mycotoxon* **12:** 520, Schenck & Smith (1982) *Mycologia* **74:**88.

Specimens examined: The soils collected from Shin Chang Ri and Sap Kyu Cheon; KNUE-S11, S12 AND S13.

Description of species

1. Azygospores, ovoid or occasionally globose,170-180 (L)×190-220μm, hyaline and thin wall layer.

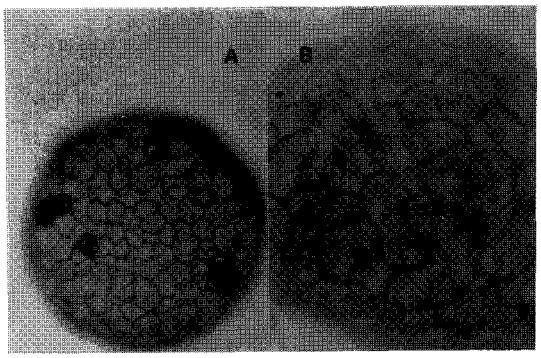

Fig.1. Azygospore of *Acaulospora bireticulata* A) 8×25 B) 8×40, polygonal projector

2. Reticulated surfaces with hexagonal shaped holes (25-27.5μm dia. length).

3. None perceptible hyphae.

Comments: Azygospores of this species were observed in the soils collected from two coast area, which was consistent with the soil reported by Tothwell and Trappe's (1979) descriptions. Norphological chatracteristics of this species was similar to that of Hall's slides (3.7 see Hall and Abbott., 1981) and to that of other description, but the size of the azygospore collected here was a little bigger than that of the above description. Clear features of this was that there was no subtanding hyphae around the azygospore and that there were also polygonal projectors in the surface ornamentation (Fig. 1).

2) *Glomus clarum* Nicolson & Schenck.

Nicolson & Schenck (1979) *Mycologia* **71:** 179, Trappe (1982) *Phytophthology* **82:** 1106, Schenck & Perez (1988) INVAM: 121, Hall (1984) VA mycorrhizal fungi: 86.

Specimens examinated: The soils collected from the communities of *Cryptomeria japonica* (Jeon Ju); KNUE S8 and S9.

Descriptions of species

1. Chlamydosporees formed singly, ovate or globose 132-142μm×135-145μm.

2. Surface ornamenysyion; cerebriform folds double layers-equal alyers (15μm).

3. Single Subtending hyphae but branched (17.5μm dia.), hyphae wall 4-5μm.

Comments: As Fig. 2 was shown, the features

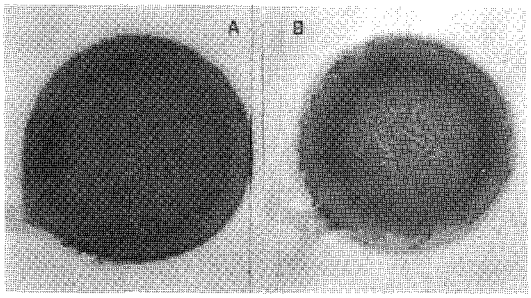

Fig.2. Chlamydospore of *Glomus clarum* A cerebriform fold surface A) 8×40 B) 8×40

of this chlamydospores were the cerebriform fold in the surface ornamentation. the shape, the longest dimension the wall thickness of this observed chlamydospore and subtanding hypha were consistent with Trappe's (1982) keys and Schenck and Perez's (1988) description. However, the cerebriform fold in the surface ornamentation was quite different from the Figures provided by Schenck and Perez (1988).

3) *Glomus intraradices* Schenck & Smith

Schenck & Smith (1982) *Mycologia* **74:** 77-92, Schenck & Perez (1988) INVAM: 154, Trappe (1982) *Phytopathology* **72:** 1106, Hall (1984) VAmycorrhizal fungi: 85; 86.

Speciments examinated: The soils collected from the communities of *Cryptomeria japonica* (Jeon Ju): NNUE-S7

Descriptions of species

1. Chlamydospores formed singly; ovoid 132.5-430um × 132.5-137.5μm

2. smooth to dull roughened surface ornamentation with several pins attached. Two wall layers thick 10-12.5μm

3. single subtending hyphae with cylindric but a little contstricted hyphae at the point attachment

Comments: The size, the wall layers and the subtanding hypha of this chlamydospore were consistent with the above descriptions (Fig. 3). The cross lines in the wall layer are clear, but not mentioned in any descriptions

4) *Glomus occultum* Walker

Walker (1982) *Mycotaxon* **15:** 49-61, Schenck & Perez (1988) INVAM; 173, Trappe (1982) *Phytopathology* **72:** 1106.

Specimens examinated: The soils collected from the communities of *Cryptomeria japonica* (Jeon Ju); KNUE S4, S5, S2, S3 and S7

Description of species

1. chlamydospores borne singly or in loose cluster in the young state, ovoid shape 122.5-125.0μm×125.0-132.5μm.

2. smooth surface and double wall layers-equal (total 12-17.5μm)

3. one subtending hyphae with cylinderic or blared towards point of attachment dia 10-12μm

Comments: The small and transparent

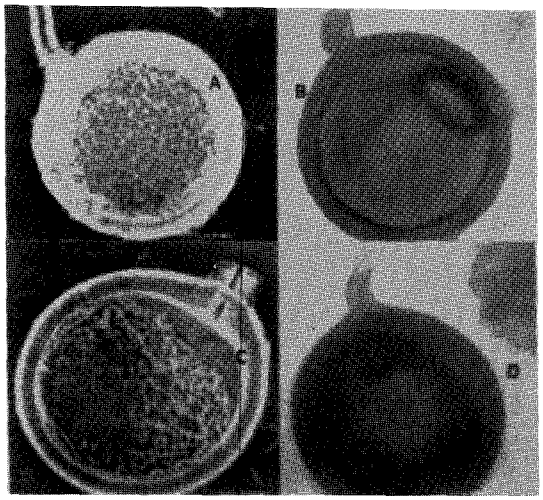

Fig.3. Chlamydospore of *Glomus intraradices* A) 8×40 B) 8×40 C) 8×100 D) 8×40

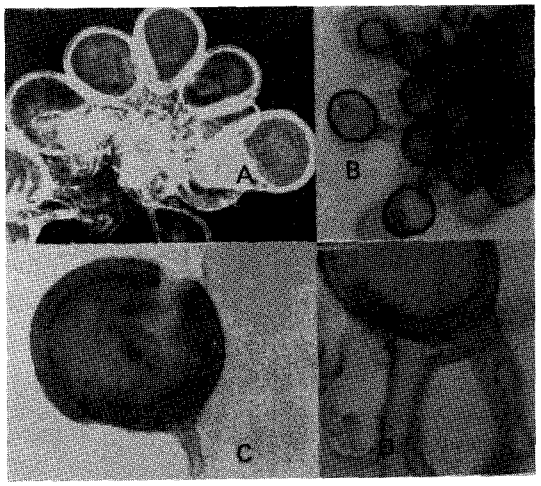

Fig.4. Chlamydospore of *Glomus occultum* A) Young spore, 8×40 B) Young spore, 8×40 C) Mature spore, 8×40 D) Young spore, 8×100

chlamydospores of this species were aggregated with eight or ten chlamydospores. The mature chlamydospores observed in Figure-4 were different from the young ones at the color and the size of this chlamydospore; the mature spore is yellow but the young spore is hyaline. The attachment of subtanding hyphae in this chlamydospore was so distinguishable.

5) *Scutellospora aurigloba* Walker & Sanders

Hall (1977) *Trans. Br. Mycol. Soc.* **68:** 341-356, Trappe (1982) *Phytopathology* **82:** 1105, Schenck & Perez (1988) INVAM; 208, Hall (1984) VA mycorrhizal fungi: 84, Nicolson & Schenck (1979) *Mycologia* **71:** 197. Schenck & Smith (1982) *Mycologia* **74:** 88.

Specimens examinated: The soils collected from the communities of *Cryptomeria japonica* (Jeon Ju); KNUE S-23, S-20, S-21, S-22

Description of species

1. azygospores, globose 274-613μm (L)×210-604μm (W) bright yellow and shinning. 2μm dia×1.5μm high small projectors distributed in the wrinkled spore surface

2. Spore wall, 4-5 layers (total 13-14μm); inner layer thiner than the outer layer

3. Bulbous hyphae 53-57.5μm dia in the longest

Comment: The small projectors were all over distributed in the surface of this azygospore (Fig.5, A, D and F). The azygospore of this has the bulbous suspensor and auxiliary cells (Figure-5, E) as the features of this species described. This has several wall layers as Trappe's (1982) and Schenck and Perez's (1988) descriptions. Hall and Abbott (1982) described this species in detail (2.7). The vesicles of this azygospore were found in Fugure-5 (E).

6) *Scutellospora gilmorei* (Trappe & Gerd.) Walker & Sanders

Schneck & Perez (1988) INVAM; 216, Gerdeman & Trappe (1974) *Mycologia Memoir* **5:** 76, Trappe (1982) *Phytopathol.* **72:** 1105, Schenck & Smith (1979) *Mycologia* **74:** 88, Hall (1984) VA mycorrhizal Fungi :84.

specimens examinated: The sand soils collected Shin Chang Ri coast: KNUE S-26, S-26-2, S-26-3, S26-4, S26-5, S26-6, S29

Descriptions of species

1. Azygospores formed singly in the soil, 250-310μm (L)×200-220μm (W), globose or occasionlly ellipsoid, spore wall consisted of several wall layers, total thick 9-11μm. The total outer wall layer is hyaline and thick (each layer less than 1 μm

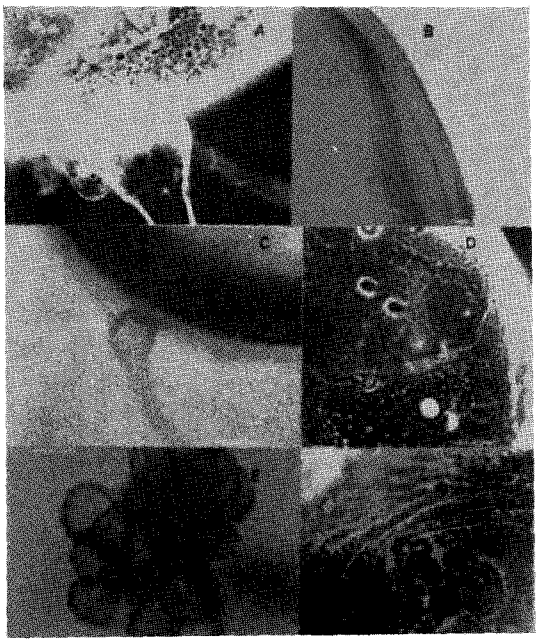

Fig.5. Azygospore of *Scutellospora aurigloba* A) Bulous suspensor, 8×40 B) Wall layer, 8×40 C) Spore surface, 8×40 D) Bulbous suspensor, 8×25 E) Auxiliary cells, 8×25 F) Spore surface, 8×40

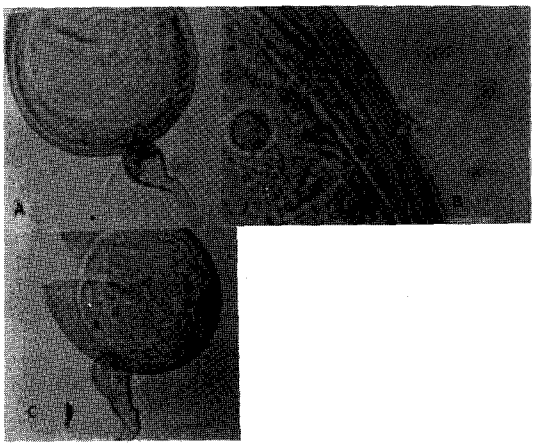

Fig.6. Azygospore of *Scutellospora gilmorei* A) Whole spore, 8×25 B) Spore wall layer, 8×100 C) Broken spore, 8×25

+ composed of more than four layers) and the inner layer thicker than the individual outer wall layer.

2. The bulbous hyphae attached to azygospore and its diameter 30-50μm

Comments: The azygospores of this species (Fig. 6) were wimilar to that of *Sc. aurigloba* described in the above (wall layers, surface morphology and bulbous suspensor). The auxiliary cell was not found in this soil. The pictures supplied by Hall and Abbott (1981) were compared here.

摘 要

전주시 완산공원(*Cryptomeria japonica* dominant forest) 및 두 해안지역(경북 신창리, *Digitaria sanguinalis* dominant ; 아산군 삽교천 *Pragmited communis dominant*)에서 채취한 토양시료로부터 내생균근 균을 관찰하였다. Endogonales에 속하는 3개 속(Acaulospora, Glomus, Scutellospora)에 6개 종이 동정되었다 ; *Aculospora bireticulata*, *Glomus clarum*, *Glomus intraradices*, *Glomus occultum*, *Scutellospora aurigloba*, *Scutellospora gilmorei*.

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