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Note

First Report of Two Nematode-trapping Fungi, *Monacrosporium ullum* sp. nov. and *Arthrobotrys amerospora*, from Korea

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***Monacrosporium ullum* sp. nov. the captures nematodes on adhesive spherical knobs was isolated from soil around *Codonopsis lanceolata* at Ulleung island, Korea. The spindle-shaped conidia sized 17-44 μm (26.5 μm) long, 7-10 μm (8.1 μm) wide, containing from 1 to 4 cross-walls but most often divided by 2 septa (47%). Resting bodies sized 57×30 μm. *Arthrobotrys amerospora* has almost spherical non-septate conidia with a small truncate protuberance at the base and sized 20-27 μm (23.3) μm long and 11-17 (14.1) μm wide. Conidiophores are somewhat longer 362.8 μm (311-418 μm) than its original description (75-250 μm).**

Keywords : adhesive knob, adhesive network, predatory fungi, nematophagous fungi

During a survey of nematophagous fungi in Korea, a putative new and an unrecorded species of nematode-trapping fungus were isolated. Description of these species are presented in this paper.

Soil and water mixture (20 g soil + 40 ml water) were placed in a beaker and well mixed for a minutes (Duddington, 1955; Kim et al., 1977). The soil mixtures were placed on four locations each in 0.2 ml on the surface of 1/2 strength cornmeal agar in a Petri dish. Nematophagous fungi were observed on the agar surface under a stereomicroscope (x50). These were examined in water and cotton blue lactophenol mount.

The fungal isolate was grown in MY medium (2% malt extract, 0.2% yeast extract) for DNA extraction. DNA was extracted from lyophilized tissue of the fungal isolate using the CTAB (cetyltrimethyl ammonium bromide) procedure as described previously (White et al., 1990). The extracted DNA was suspended in TE buffer (10 mM Tris-HCl, 10 mM EDTA, pH 8.0). The primers ITS1 (5-TCCGTA-GGTGAACCTGCGG-3) and ITS4 (5-GGAAGTAAAAG-TCGTAACAAGG-3) were used to amplify the ITS region I and 5.8S rDNAs, the ITS region II, and a portion of the 28S rDNA (White et al., 1990). Each reaction was conducted by *ExTaq* Premix (TaKaRa, Japan) and 0.5 pM of ITS 1 and 4 primers were used. The thermal cycling parameters were initial denaturation at 94°C for 4 min followed by 30 cycles of denaturation at 94°C for 30s, annealing at 56°C for 30s and extension at 72°C for 100s; a final extension at 72°C for 10 min was done at the end of the amplification. PCR products were electrophoresed in a 2% agarose gel (TaKaRa, Japan) and expected products were purified with a Gel Purification Kit (Bioneer, Korea). The purified PCR products were ligated to pGEM-T easy vector system (Promega, USA) and transformed to *E. coli* strain DH5α. Plasmid DNA carrying an insert DNA was extracted from white colony and used as the template for DNA sequencing. DNA sequencing was carried out in an ABI 3700 (Applied Biosystems, Japan) using the ABI Prism® BigDye™ Terminator Cycle Sequencing Ready Reaction Kit. The nucleotide sequences were compared visually and aligned using Clustal X (ver. 1.8, Thompson et al., 1997), and a phylogenetic tree was constructed from genetic distance values calculated using the neighbor joining method (Saitou and Nei, 1987). The phylogenetic tree was

Table 1. Species NCBI Genbank access No.

Species	NCBI Genbank access No.
Monacrosporium candidum	AY965749
M. drechsleri	AY695063
M. ellipsosporum	AY695065
M. haptotylum	AF106523
M. leptosporum	AF106529
M. lysipagum	AY695067
M. mammillatum	AY902794
M. parvicolle	AY965748
M. phymatopagum	U51970
M. robustum	AY965755
M. sclerohyphum	AY902806
M. shuzhengense	AY965752
M. sichuanense	AY902795
M. tentaculatum	AF106531
M. yunnanense	AY965757
M. ullum	**this work**

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visualized and edited by TREEVIEW software version 1.6.6 (URL: http://taxonomy.zoology.gla.ac.uk/rod/rod.html).

***Monacrosporium ullum* sp. nov.** Mycelium sparsum; hyphis incoloratis, mediocriter septatis, plerumque 2.5-3.9 μm crassis, hic illic ex ramulo recto vel curvato, 6-26 μm longo, 2.6-3.6 μm crasso, in 1-2 cellulis consistente, bullas tenaces globosas vel ellipsoideas 7.7-11.8 μm longas, 7.1-9.6 μm crassas, singillatim emittentibus; his bullis ad vermiculos nematoideos inhaerentibus, its animalia tenentibus, integumentum eorum perforantibus, tuber mortiferum intrudentibus. Hyphae fertiles incoloratae, erectae, 65-99 μm altae; conidiis incoloratis, vulgo fusoideis, apice anguste rotundatis, basi truncatis, plerumque 17-44 μm (saepius circa 26.5 μm) longis, 7-10 μm (saepius circa 8.1 μm) crassis, 1-4 septatis, saepissime diseptatis (47%). Corpora perdurantia saepe 57×30 μm. Vermiculos nematoideos multarum specierum capiens consumensque

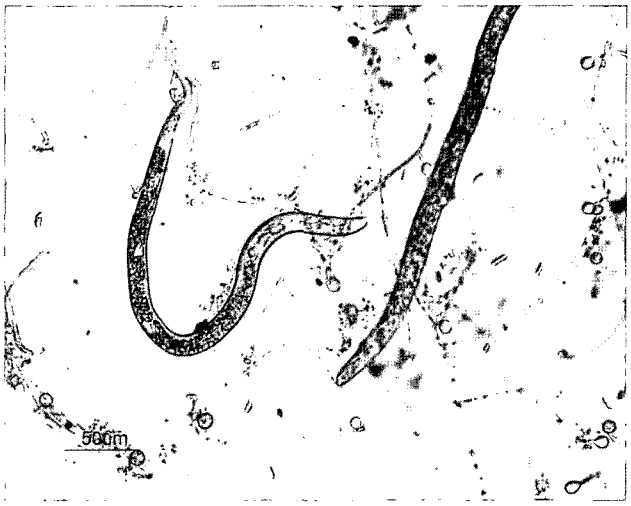

Fig. 1. Juveniles of root-knot nematode captured by *Monacrosporium ullum* sp. nov.

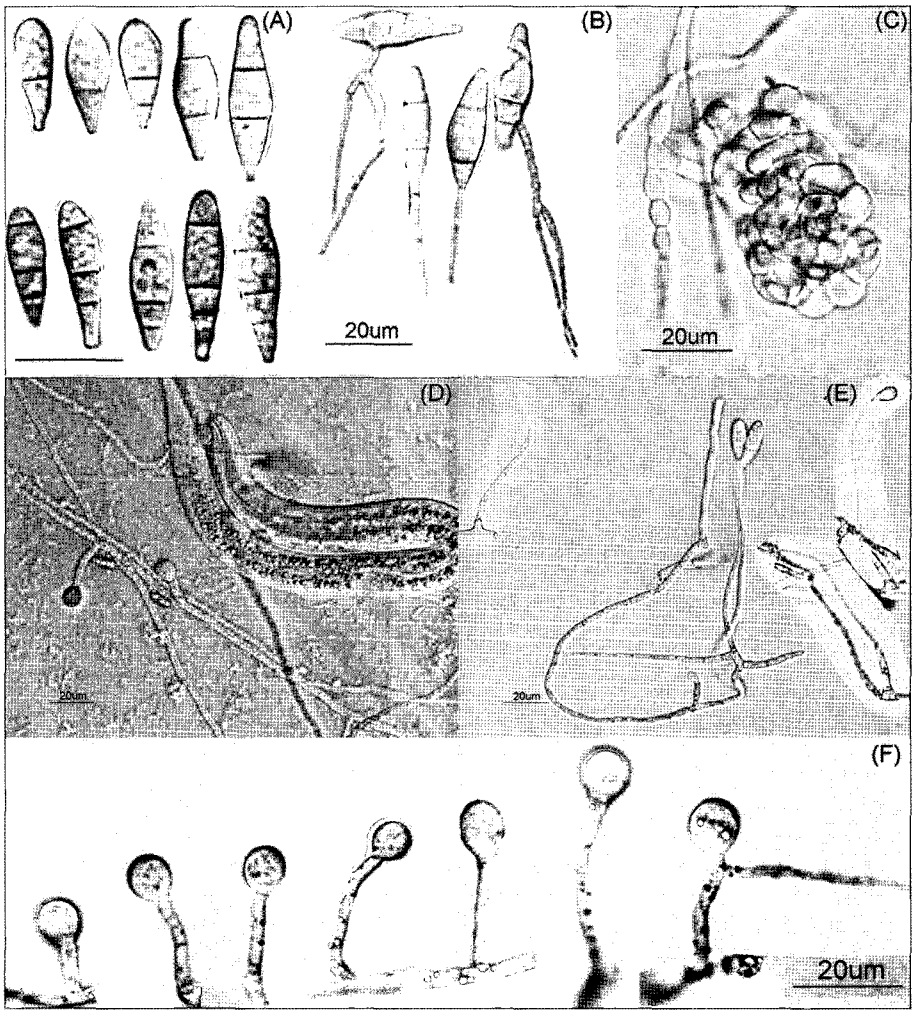

Fig. 2. *Monacrosporium ullum* sp. nov. (A) Conidia, (B) Germinating conidia, (C) Chlamydospore, (D) Captured nematode on adhesive knob, (E) Conidiophores, and (F) Adhesive knobs.

habitat in materiis plantarum putrescentibus prope Ulleung island, Korea.

Mycelium scanty; vegetative hyphae colorless, septate at moderate intervals, mostly 2.5 to 3.9 μm wide, often especially in the presence of nematodes giving rise on straight or curved stalks, 6 to 26 μm long and 2.6 to 3.6 μm wide, usually uniseptate or biseptate, to solitary adhesive knobs, globose or ellipsoidal in shape, commonly only 7.7 to 11.8 μm long and 7.1 to 9.6 μm wide; these knobs capturing nematodes including plant-parasitic nematodes (Fig. 1). Conidiophores colorless, erect, frequently 65 to 99 μm high; conidia colorless, commonly spindle-shaped narrowly rounded at the tip, truncate at the base, mostly 17 to 44 μm (average about 26.5 μm) long, 7 to 10 μm (average about 8.1 μm) wide, containing from 1 to 4 cross-walls but most often divided by 2 septa into 3 cells (47%) after being germinated sometimes putting forth a branched hyphae (Fig. 2). Resting bodies formed sometimes sized 57×30 μm. Capturing and consuming nematodes of different species including plant-parasitic nematodes, it occurs in decaying plant materials from Ulleung island, Korea.

The fungus that captures nematodes on adhesive spherical knobs was isolated from soil around *Codonopsis lanceolata* (Sieb. & Zucc.) Trautv. at Ulleung island, Korea. The fungus named *Monacrosporium ullum* resembles *M. fusiformis* in 2-3 septate conidia and spore shape but differ in their trapping organ (adhesive knob vs. 3-D networks) and size of conidia (16.8-43.7 (26.5) μm×6.8-9.9 (8.1) μm) is much smaller than that of *M. fusiformis* (29.5-48.5×10.5-15.5 μm) (Cooke & Dickinson, 1965). Conidial size of *M. ullum* was similar to that of *M. candidum* (26-52×5.5-11.5 μm), but difference between them was that the new fungus produces 1-4 septate conidia (mainly in 2 septate), while *M. candidum* produces 4-6 septate conidia. *M. candidum* has

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TCCGTAGGTGAACCTGCGGAAGGATCATTA/CCAATCATGTCTCCCAGCCGAAAGGTTGGTGCAGGCGCCTTAACCCTTTG
                          18S rDNA/ITS I
TGAACCAAAAAACCTTTCGCTTCGGCAGCAGCTCGGTTGGCAACAGCCTCTGCGTCAGCCTGCCGGTAGCACCAATCATC
AAAACTTGCAGTTAATAACATTGTCTGATTACCAAATTTTCGAATGAAAATC/AAAACTTTCAACAACGGATCTCTTGGTT
                                                    5.8S
CCCGCATCGATGAAGAACGCAGCGAAACGCGATAGTTAATGTGAATTGCAGAATTCAGTGAATCATCGAGTCTTTGAACG
CACATTGCGCCCATTGGTATTCCATTGGGCATGTCTGTTTGAGCGTC/ATTACAACCCTCGGTCACCACCGGTTTTGAGCA
                                                  ITS II
AGCGAGGTCTCCGGACCCGGCTGGCTTTAAAGTTGTAAGCTCTGCTGGCTGCCAGGCCCAACCAGAACATAGTAAAATCA
TGCTTGTTCACGGTTTGCGGTCGAAGCGGTACGGCCTGAACAATACCTACCACCTCTTAGGTT/TGACCTCAGATCAGACA
                                                                  28S rDNA
AGGATACCCGCTGAACTTAAGCATATCAATAAGCGGAGGA
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Fig. 3. Nucleoltide sequences of Inter Trancribed Spacer and 5.8S rDNA from *M. ullum*.

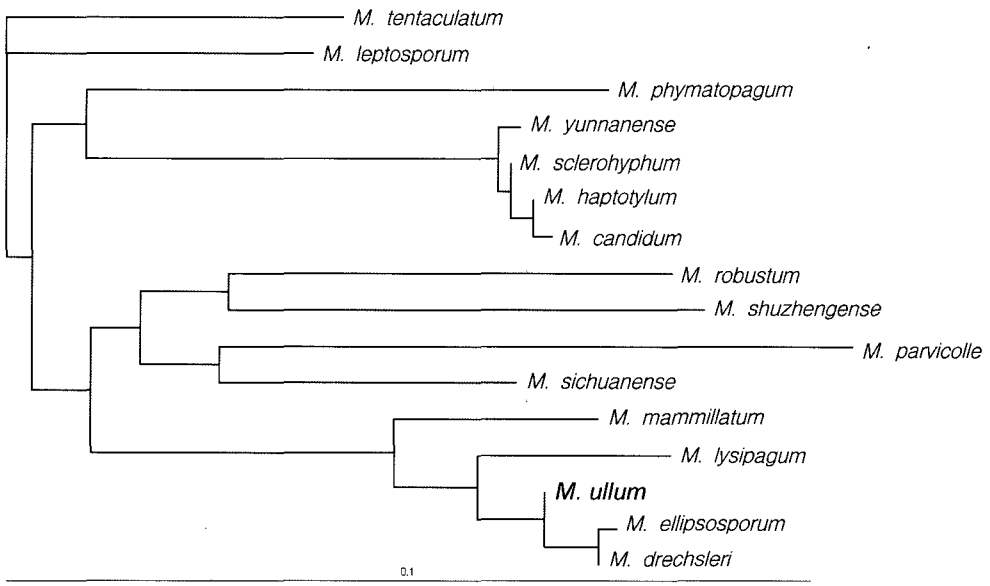

Fig. 4. The phylogenetic tree based on the nucleotide sequences of the ITS and 5.8S regions of *Monacrosporium* by Neighbor-Joining method.

both adhesive knob and non-constructing ring as trapping organ and the height of conidiophore is 150-400 μm (Drechsler, 1937, 1944). Among the genus of *Monacrosporium* with adhesive knobs on stalk, *M. ullum* has the smallest conidia (26.5×8.1 μm) compared to those of 40-55×10-17 μm of *M. drechsleri, M. haptotylum, M. mammillatum, M. mutabilis*, and *M. ellipsosporum*.

In a phylogeny analysis, *M. ullum* is most closely related to *M. ellipsosporum* and *M. drechsleri* followed by *M. mammillatum* and *M. lysipagum* (Figs. 3~4).

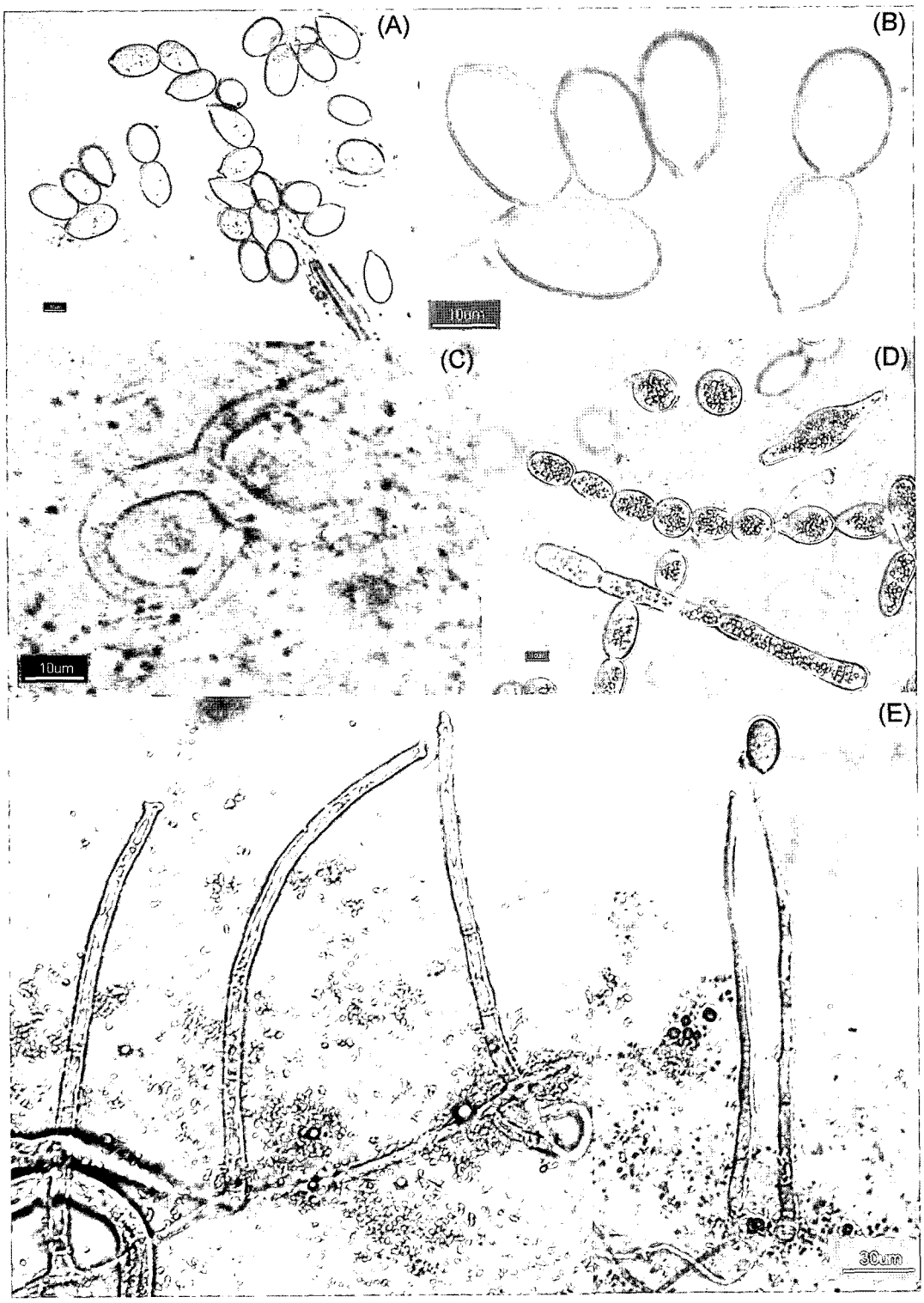

Fig. 5. *Arthrobotrys amerospora.* (A, B) Conidia, (C) adhesive network, (D) Chlamydospores, and (E) Conidiophore.

Table 2. Comparisons of *Arthrobotrys amerospora* isolates from Korea with the original description (Schenck et al., 1976).

Characters	Korean isolate (μm)	Original description (μm)
Conidium length	23.3 (20-27)	23.6 (15-31)
width	14.1 (11-17)	15.9 (10-20)
septum	0	0
Conidiophore length	362.8 (311-418)	75-250
Chlamydospores	15-72×10-20	18-50×18-23

***Arthrobotrys amerospora* Schenck, Kendrick et Pramer.** This species was described by Schenck et al. (1976) on nematode infested agar as producing 2-10 conidia which were hyaline, obovoid, one-celled, 15-31 (23.6) μm long and 10-20 (15.9) μm wide, with a small truncate protuberance at the base. The conidiophores were simple, elect, septate, unbranched, 75-250 μm long, producing 2-10 conidia, conspicuous denticles at and near the apex. Chlamydospore yellowish, smooth walled, spherical to elongate-ellipsoidal, 18-50×18-23 μm, usually intercalary, single or in chain.

A fungus subsequently identified as *A. amerospora* was isolated from soil from commercial greenhouse at Sangju, Korea. Unlike the fungus described by Schenck et al. its conidiophores were much longer 362.8 μm (311-418 μm) (Fig. 5) and there were abundant chlamydospores in two months old Potato dextrose agar culture, which were usually intercalary, single or in chain. Despite the differences noted above between this fungus and Schenck's original description of *A. amerospora* it seems clear to be the same species. *A. amerospora* is immediately recognized by almost spherical non-septate conidia with relatively short conidiophore. This is the first record of *Arthrobotrys* with non-septate conidia in Korea.

Acknowledgements

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