

## Endophytic Fungi Isolated from the Marine Macroalga *Dictyopteris pacifica* in Korea

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### ABSTRACT

The exploration of endophytic fungi associated with seaweeds has garnered significant interest due to their crucial ecological functions and potential as sources of valuable bioactive compounds. In this study, we isolated and identified endophytic fungi from the brown seaweed *Dictyopteris pacifica*, collected from the intertidal zone of Yeongdoek, Gyeongsangbuk-do in Korea. Through morphological examination and molecular phylogenetic analysis using multiple molecular markers, including ITS, LSU, SSU, TEF1, TUB2, and RPB2 sequences, we identified three fungal species not previously recorded in Korea: *Emericellopsis fuci*, *Neoarthrimum lithocarpicola*, and *Periconia chimonanthi*. Detailed descriptions of morphological characteristics and phylogenetic analyses are provided. This study represents the first report of these endophytic fungi isolated from *D. pacifica* in Korea, thereby enhancing the understanding of the diversity of seaweed-associated endophytic fungi in the region.

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### 1. Introduction

*Dictyopteris pacifica* is a flattened, branching thalloid brown alga that attaches to rocks and is distributed along the coasts of Korea, China, Japan, and Taiwan [1]. This species is rich in niacin and folic acid, crucial for human energy metabolism, as well as lipoic acid, which may serve as an adjunct in treating various diseases [2, 3]. Furthermore, the genus *Dictyopteris* is prevalent in tropical, subtropical, and temperate marine regions renowned for its chemical diversity and bioactive properties [4].

Endophytes are microorganisms that colonize plant tissues without causing visible pathological symptoms in the host [5–7]. Initially defined in relation to terrestrial plants, the term “endophyte” now encompasses organisms involved in mutualistic associations with a variety of hosts [8]. Studies have shown that nearly all plant species in natural ecosystems harbor symbiotic relationship with fungal endophytes [9–12]. In the marine environment, endophytes associate with seaweeds, forming groups known as Marine Algicolous Fungi (MAFs) which establish symbiotic relationship [13]. These marine endophytes obtain nutrition and protection from their seaweed hosts, while producing secondary metabolites that enhance the host’s tolerance to environmental stress and defend against herbivores,

thereby playing a critical role in the host’s ecological adaptation [14, 15].

In recent years, endophytes have attracted attention as sources of bioactive natural products with potential applications in pharmaceuticals, agriculture, and industry [16]. Seaweed endophytes, in particular, are valued for their resilience in harsh marine environments, including exposure to sunlight, salinity fluctuations, and tides, which drives their ability to produce novel secondary metabolites [17, 18]. These bioactive compounds are increasingly recognized for their potential as environmentally friendly and sustainable natural resources [19].

Although studies on the diversity of fungi in Korean marine ecosystems have gradually increased [20–22], research on endophytic fungi associated with seaweeds are almost nonexistent, leaving significant gaps in understanding the symbiotic relationships and ecological roles of these fungi remains limited [23]. In this study, we present the morphological characteristics and molecular phylogenetic analysis of three previously unrecorded endophytic fungal species isolated from *Dictyopteris pacifica* collected in Yeongdoek, Korea: *Emericellopsis fuci*, *Neoarthrimum lithocarpicola*, and *Periconia chimonanthi*. This study enhances the understanding of the diversity of endophytic fungi associated with seaweeds in Korea.

## 2. Materials and methods

### 2.1. Sample collection and preparation

In May 2024, samples of *D. pacifica* were collected from the intertidal zone of Yeongdeok, Korea (36°22'52.6"N 129°24'20.5"E). Only healthy specimens without damage or disease symptoms were selected. The collected samples were placed in clean plastic bags containing seawater and transported to the laboratory within 12 h. Upon arrival, samples were immediately subjected to surface sterilization without any storage period.

### 2.2. Surface sterilization and fungal isolation

To eliminate surface contaminants, samples were first rinsed with tap water [16]. Subsequently, the samples were trimmed into smaller sections measuring 1.5 cm × 0.5 cm, suitable for culturing. Surface sterilization was achieved by immersing the trimmed sections in 70% ethanol for 15 s, followed by rinsing with sterile seawater to remove any residual ethanol [24]. After sterilization, the samples were blotted dry with sterile cotton [25] and placed onto Dichloran Rose Bengal Chloramphenicol (DRBC; MCell, Seoul, Korea) medium supplemented with seawater, as well as on Potato Dextrose Agar (PDA; Difco Lab., Detroit, USA) medium supplemented with chloramphenicol [16]. The inoculated plates were incubated at 25°C in the dark to allow the growth of endophytic fungi. Once hyphal growth was observed, the fungi were subcultured on PDA medium to obtain pure isolates [26].

### 2.3. Culturing and morphological characterization

Mycelia emerging from within the segments of *D. pacifica* were transferred to fresh media to establish pure fungal cultures. These isolates were cultured on Malt Extract Agar (MEA; Kisan Bio, Seoul, Korea) and PDA at 25°C in darkness to assess colony morphology. Colony characteristics, including color, texture, and margin, were observed visually. Spore-forming structures were examined under a light microscope (Axio Imager A2, Carl Zeiss, Oberkochen, Germany) to document morphological features.

### 2.4. DNA extraction, PCR and sequencing

For molecular phylogenetic identification, genomic DNA was extracted from the mycelia using the HiGene Genomic DNA Prep Kit (Biofact, Daejeon, Korea). Specific DNA regions essential for strain identification were amplified by Polymerase Chain

Reaction (PCR) using universally standard primers. The primers used and the corresponding PCR conditions are detailed in Table 1 [27–32]. DNA sequencing was performed by Solgent Co., Ltd. (Daejeon, Korea). The obtained sequences were compared with those in the National Center for Biotechnology Information (NCBI) database using the Basic Local Alignment Search Tool (BLAST).

### 2.5. Phylogenetic analysis

The analysis incorporated combined sequences of ITS, LSU, SSU, TEF1, TUB2, and RPB2 as applicable. Phylogenetic trees were constructed using the Maximum likelihood in MEGA11 software [33]. The chosen evolutionary models for the analysis were TN93+G, K2+G and TN93+G for strains 24S071, 24S082 and 24S115, respectively. Bootstrapping was performed with 1000 replicates to assess the robustness of the tree topology. The isolated strains were deposited in the National Institute of Biological Resources (NIBR), and their sequences were registered with NCBI under accession numbers.

## 3. Results

### 3.1. Phylogenetic analysis

The identification of the isolated fungal strains was corroborated through comprehensive molecular phylogenetic analysis using multiple genetic markers. The ITS sequence of strain KNUE24S071 exhibited 99.82% identity with *Emericellopsis fuci* KO01 (GenBank accession PP125170), the LSU sequence was 100% identical to *E. fuci* CBS127350 (GenBank accession MH875970), the RPB2 sequence matched *E. fuci* CBS:113887 (GenBank accession OQ453954),

**Table 1.** DNA region, primer sets, and PCR condition for molecular phylogenetic analysis.

| DNA region | Primer pair                | PCR condition                          | Fungal strains           |
|------------|----------------------------|--|--------------------------|
| ITS        | ITS1F/ITS4 [27]            | 95 °C: 20 s, 50 °C: 40 s, <sup>a</sup> | KNEU24S071               |
|            |                            | 72 °C: 60 s (35 cycles) <sup>d</sup>   | KNUE24S082<br>KNNE24S115 |
| LSU        | LR0R/LR5 [28]              | 95 °C: 20 s, 44 °C: 40 s, <sup>a</sup> | KNEU24S071               |
|            |                            | 72 °C: 60 s (35 cycles) <sup>d</sup>   | KNUE24S082<br>KNUE24S115 |
| RPB2       | RPB2-5F2/RPB2-7cR [29]     | 95 °C: 45 s, 56 °C: 80 s, <sup>b</sup> | KNEU24S071               |
| SSU        | NS1/NS4 [28]               | 72 °C: 120 s (35 cycles) <sup>e</sup>  | KNUE24S115               |
|            |                            | 95 °C: 20 s, 40 °C: 40 s, <sup>a</sup> |                          |
| TEF1       | EF1-983F/EF-2218R [30, 31] | 72 °C: 60 s (35 cycles) <sup>d</sup>   | KNEU24S071<br>KNUE24S115 |
|            |                            | 95 °C: 30 s, 52 °C: 30 s, <sup>c</sup> |                          |
| TUB2       | Bt2a/Bt2b [32]             | 72 °C: 80 s (35 cycles) <sup>e</sup>   | KNUE24S082               |
|            |                            | 95 °C: 20 s, 55 °C: 40 s, <sup>a</sup> |                          |
|            |                            | 72 °C: 60 s (35 cycles) <sup>e</sup>   |                          |

<sup>a</sup>Initiation step of 95 °C: 2 min.

<sup>b</sup>Initiation step of 95 °C: 5 min.

<sup>c</sup>Initiation step of 94 °C: 5 min.

<sup>d</sup>Final elongation step of 72 °C: 5 min and final hold at 4 °C.

<sup>e</sup>Final elongation step of 72 °C: 10 min and final hold at 4 °C.

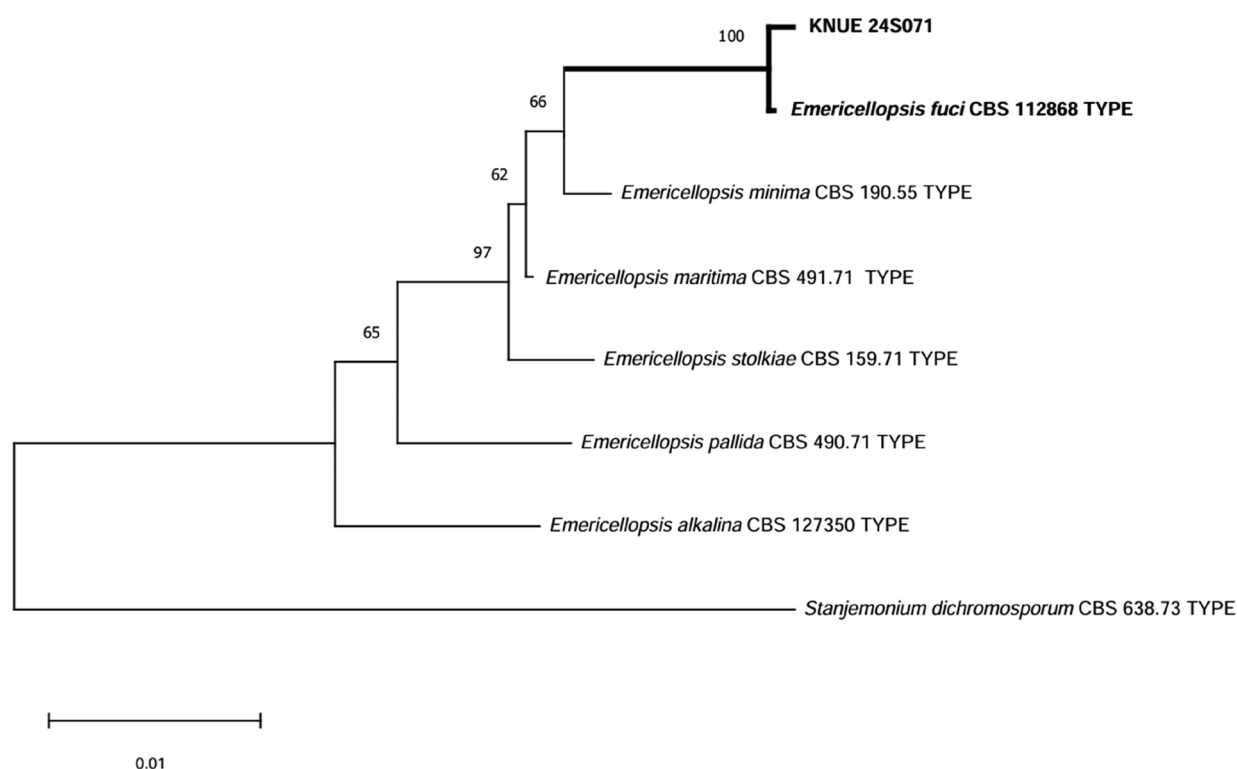
and the TEF1 sequence was identical to *E. fuci* CBS113887 (GenBank accession OQ470861). In the maximum likelihood phylogenetic tree constructed using the combined ITS, LSU, RPB2, and TEF1 sequences, strain KNUE24S071 clustered closely with *E. fuci* CBS 112868 (Figure 1), confirming its identification. The ITS sequence of strain KNUE24S082 showed 99.64% identity with *Neoarthriniium lithocarpicola* CFCC 54456 (GenBank accession NR\_182597), the LSU sequence demonstrated 99.76% identity with the same strain (GenBank accession NG\_149070), and the TUB2 sequence had a 94.16% identity with *N. lithocarpicola* CFCC 54456 (GenBank accession ON456914). Phylogenetic analysis using the combined ITS, LSU, and TUB2 sequences placed KNUE24S082 in a distinct clade alongside *N. lithocarpicola* CFCC 54456 (Figure 2), thereby supporting its classification. For strain KNUE24N115, the ITS sequence revealed a 99.63% identity with *Periconia chimonanthi* KUMCC 20-0266 (GenBank accession NR\_176752), the LSU sequence showed 99.77% identity with the same strain (GenBank accession MW448572), the SSU sequence exhibited 99.90% identity (GenBank accession MW448656), and the TEF1 sequence was 100% identical to *P. chimonanthi* KUMCC 20-0266 (GenBank accession MW460897). In the maximum likelihood phylogenetic tree based on the combined ITS, LSU, SSU, and TEF1 sequences,

strain KNUE24N115 formed a well-supported clade with *P. chimonanthi* KUMCC 20-0266 (Figure 3), confirming its identification.

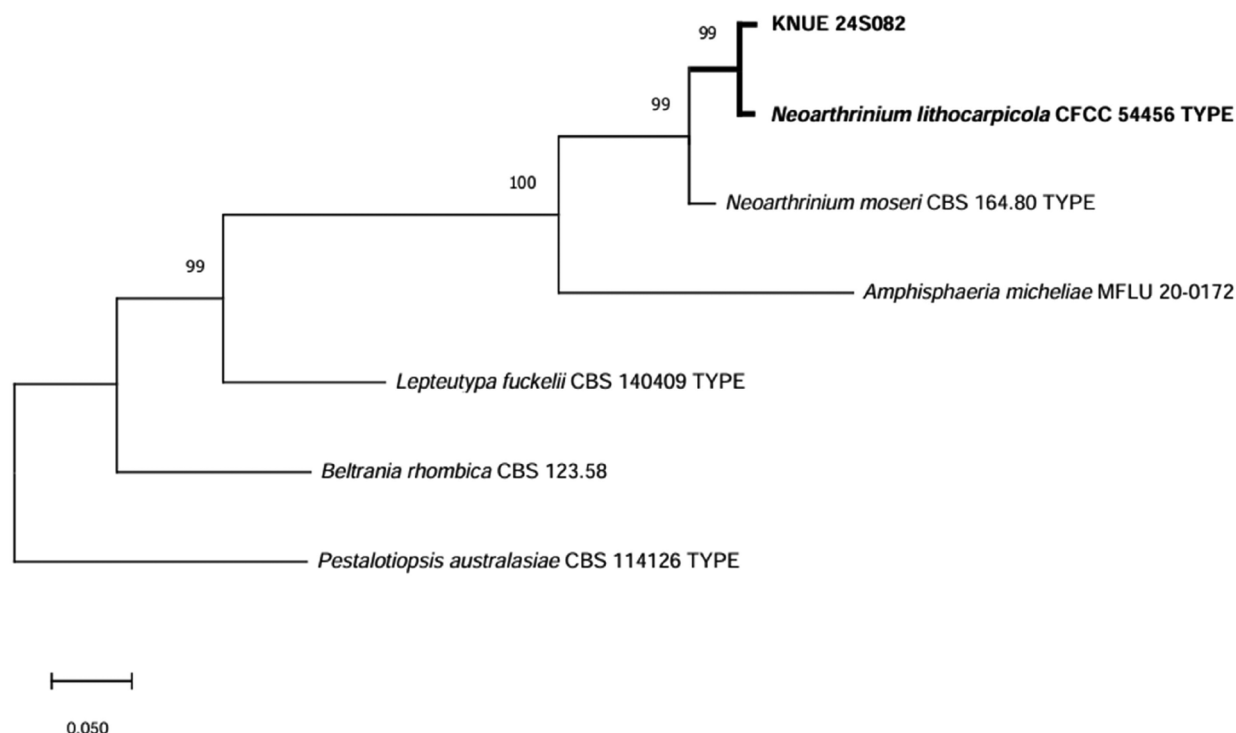
### 3.2. Taxonomy

***Emericellopsis fuci*** (Summerb., Zuccaro & W. Gams) L.W. Hou, L. Cai & Crous, Stud. Mycol. 105: 117 (2023) [MB#845861] (Figure 4).

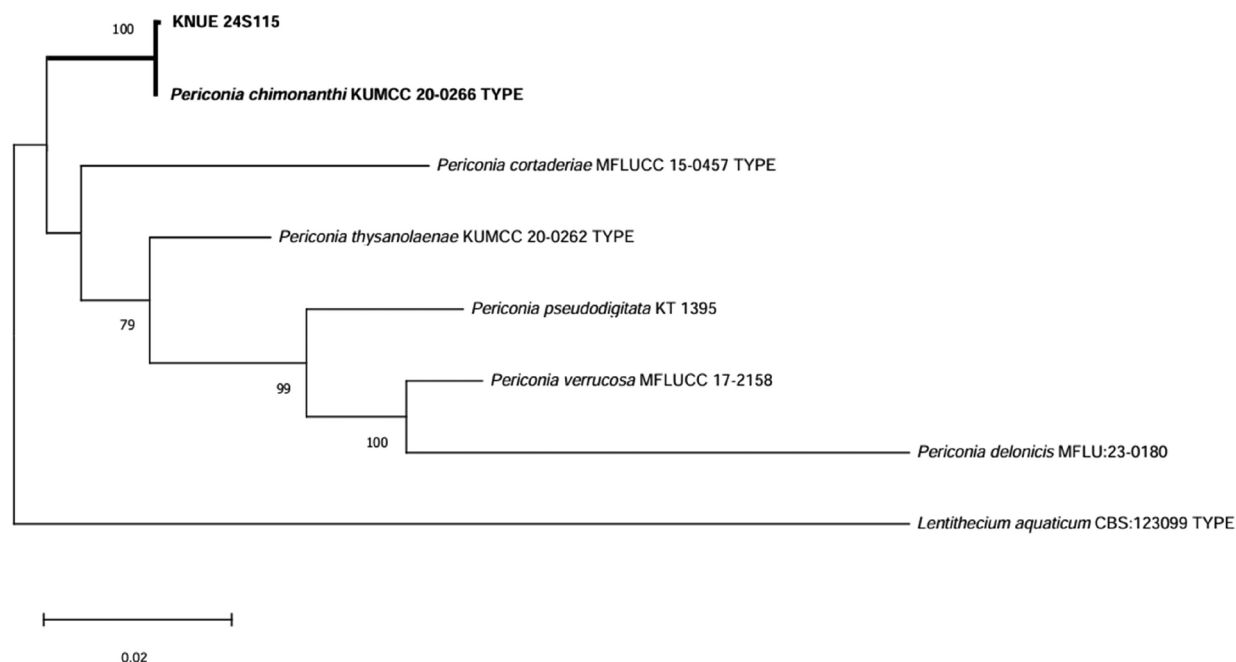
**Morphological characteristics of strain KNUE24S071:** After 7 days of incubation at 25°C in the dark, colonies on MEA reached a diameter of 28.0–31.0 mm. The colony surface was light orange in the center, gradually fading toward the margins. The reverse side of the colony displayed similar coloration to the front. Colonies were circular with an entire margin, flat with low wrinkles observed in the central region, and exhibited an overall pasty texture. On PDA, the colony diameter was 32.0–34.0 mm, and other morphological characteristics were similar to those on MEA. Phialides were varied in length, typically attached singly to the supporting hyphae, although some were relatively longer, consisting of 2–3 contiguous basal cells. Conidia showed high morphological diversity, being ovoid, broadly elliptical, and occasionally club-shaped. The size of the spores was  $(3.67\text{--}5.91(-13.01) \times (2.23\text{--}3.01(-3.95) \mu\text{m}$  ( $n=20$ ).



**Figure 1.** Maximum likelihood tree of *Emericellopsis fuci* KNUE 24S071. The tree is based on concatenated sequences of internal transcribed spacer (ITS), large subunit ribosomal RNA (LSU), RNA polymerase II second largest subunit (RPB2), translation elongation factor 1-alpha (TEF1) DNA sequences. *Stanjemonium dichromosporum* was used as an outgroup. The numbers on the nodes represent bootstrap values greater than 50% (1,000 replicates).



**Figure 2.** Maximum likelihood tree of *Neoarthriniaceae lithocarpicola* KNUE 24S082. The tree is based on concatenated sequences of internal transcribed spacer (ITS), large subunit ribosomal RNA (LSU), beta-tubulin (TUB2) DNA sequences. *Pestalotiopsis australasiae* was used as an outgroup. The numbers on the nodes represent bootstrap values greater than 50% (1,000 replicates).

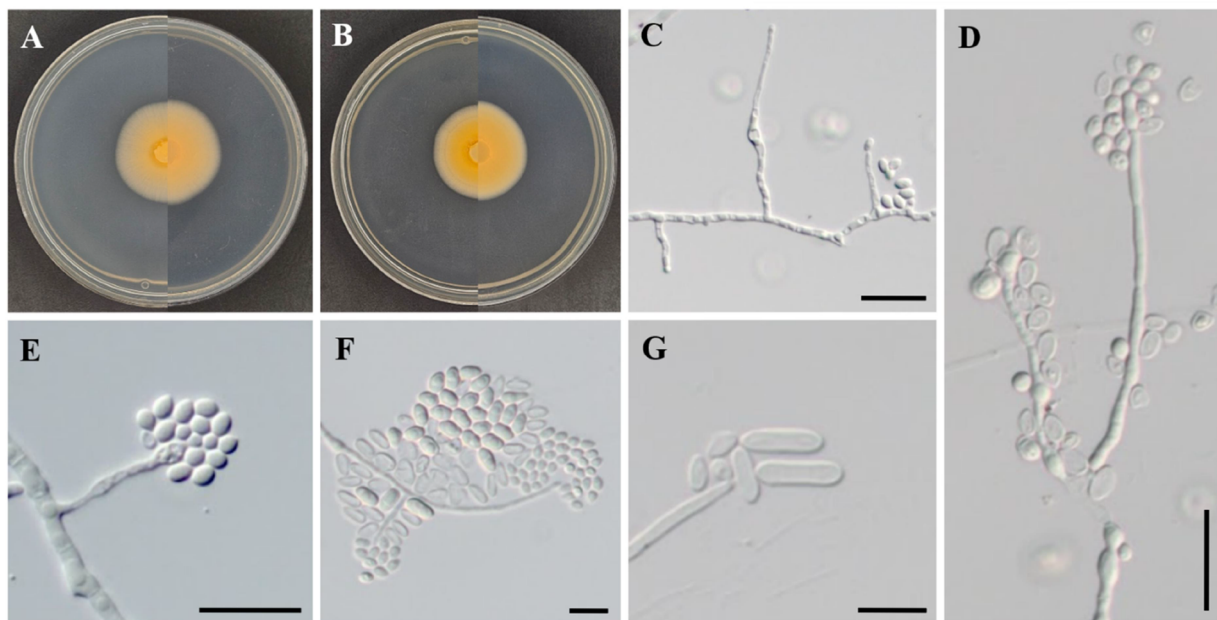


**Figure 3.** Maximum likelihood tree of *Periconia chimonanthi* KNUE 24S115. The tree is based on concatenated sequences of internal transcribed spacer (ITS), large subunit ribosomal RNA (LSU), small subunit ribosomal RNA (SSU), translation elongation factor 1-alpha (TEF1) DNA sequences. *Lentithecium aquaticum* was used as an outgroup. The numbers on the nodes represent bootstrap values greater than 50% (1,000 replicates).

**Specimen examined:** Yeongdeok, Gyeongsangbuk-do, Korea, 36°22'52.644"N 129° 24'20.52"E, May 9, 2024, *E. fuci*, isolated from *D. pacifica*, strain KNUE24P071, NIBRFGC000512612, GenBank No. PQ498693 (ITS), PQ498694 (LSU), PQ537098 (RPB2), and PQ537099 (TEF1).

**Notes:** The basionym *Acremonium fuci* is characterized by relatively large, ovoid conidia, distinguishing it as the second *Acremonium* species documented from the marine environment with notably large conidia [30]. Alongside the holotype, numerous additional specimens have been isolated from brown





**Figure 4.** Morphology of *Emericellopsis fuci* KNUE 24S071. A-B Colony after 7 days of growth at 25°C. The left side shows the front view and the right side shows reverse view, on malt extract agar (A) and potato dextrose agar (B). C-G: Microscopic features of phialides and conidia. C: Phialides of varying lengths. D: Longer phialides. E: Conidiogenous occurring in the phialide. F: Conidia with diverse sizes and shapes. G: Larger conidia. Scale bars: C-E=20 µm, F-G: 10 µm.

**Table 2.** Morphological characteristics of strain KNUE 24S071 and *Emericellopsis fuci* CBS 112868.

| Characteristics    | <i>Emericellopsis fuci</i><br>KNUE 24S071   | <i>Emericellopsis fuci</i><br>CBS 112868 [34]   |
|--------------------|---|---|
| Colony             | MEA, PDA, 25°C, 7 days<br><b>MEA</b> , 28.0–31.0 mm, light orange yellow at center, gradually fading toward the edges, reverse concolorous, circular, even surface or, at colony center folded in to low wrinkles, smooth with a moist texture.<br><b>PDA</b> , 32.0–34.0 mm, similar to those on MEA.            | MEA, OA, 20°C, 10 days<br><b>MEA</b> , 23.0–30.0 mm, pale pinkish orange, reverse concolorous, flat or, at colony center, folded into low wrinkles, overall pasty in texture.<br><b>OA</b> , similar to those on MEA but with a mucoid surface.   |
| Asexual morphology | <b>Phialide</b> : 6.32–80.36 µm, almost attached singly to subtending hyphae, some phialides are relatively long and consist of 2–4 contiguous basal cells.<br><b>Conidia</b> : High diversity, obovoid, widely elliptical, occasionally club-shaped, (3.67–)5.91(–13.01) × (2.23–)3.01(–3.95) µm ( <i>n</i> =20) | <b>Phialides</b> : 7.5–23.5 µm, almost attached singly to subtending hyphae, terminal phialides relatively long (42–57 µm) and in combination with 2–4 contiguous basal cells.<br><b>Conidia</b> : obovoid, broadly ellipsoidal, sometimes broadly clavate, truncated at the base, typically measuring 5–8(–15) × 3.2–5(–6) µm ( $\bar{x}$ =8 × 5 µm) |

MEA: malt extract agar, PDA: potato dextrose agar, OA: oatmeal agar.

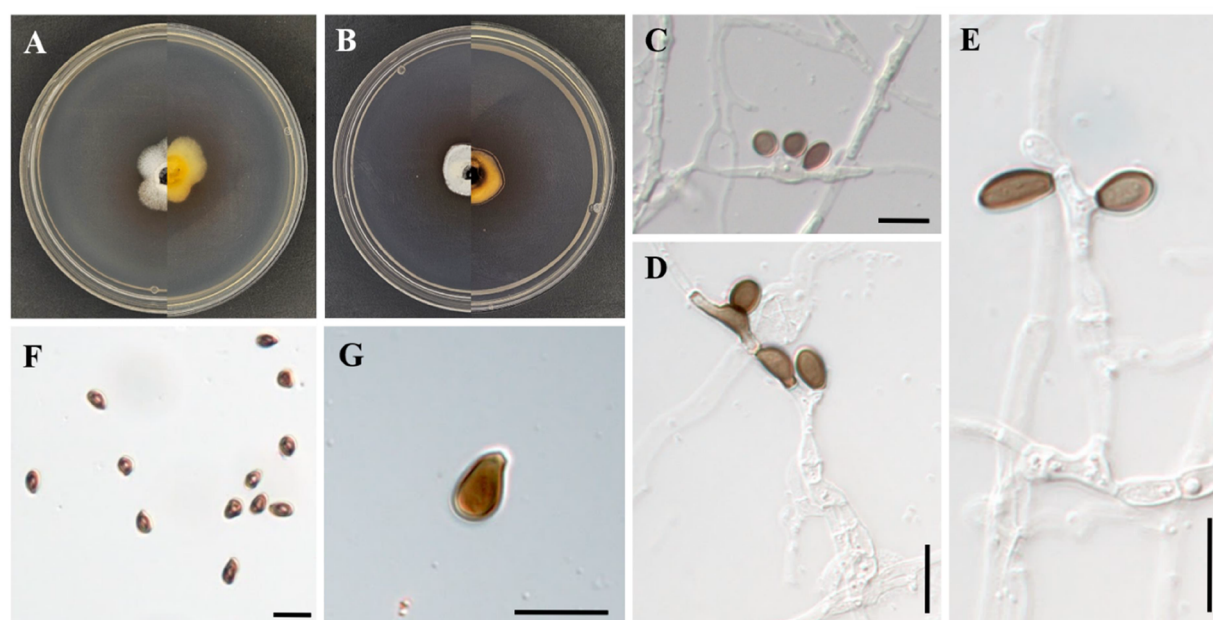
algae in the family Fucaceae. Phylogenetically, *A. fuci* is associated with algae and, as of 2023, has been incorporated into a distinct clade within the genus *Emericellopsis*, thereby reflecting an ecological niche adaptation within this genus [30, 34]. According to Zuccaro et al. [34], conidia of *E. fuci* exhibit rapid germination when exposed to *Fucus serratus* (Fucaceae, brown algae) or its aqueous homogenates, yet fail to germinate in their absence [34] (Table 2).

***Neoarthrimum lithocarpicola*** Ning Jiang, MycoKeys 92: 36 (2022) [MB#843846] (Figure 5).

**Morphological characteristics of strain KNUE24N082:** After 7 days of growth at 25°C in the dark, colonies on MEA reached a diameter of 15.0–30.0 mm. The colony surface was dark grey at the center, becoming bright grey to white toward margin.

The reverse side of the colony showed deep yellow center and pale yellow margin. The overall appearance of the colonies was irregular, with filiform edges, flat, and a cottony texture. On PDA, the colony diameter was 14.0–19.0 mm, with a dark grey center and bright grey to white margin on the surface. The reverse side appeared orange-yellow at the center, fading to pale yellow at the margin. The colonies were irregular but nearly circular in shape, with an entire margin. Conidiophore is cylindrical, septate, sometimes branched. Conidiogenous cells were erect, blastic, and form clusters along the hyphae, appearing hyaline to pale brown. Conidia were brown to dark brown, with ellipsoid, obovoid, acuminate at one end, and polygonal shapes. The size of the spores was (5.9–)6.7(–7.7) × (3.4–)4.4(–5.0) µm (*n*=20).

**Specimen examined:** Yeongdeok, Gyeongsangbuk-do, Korea, 36°22'52.644"N 129° 24'20.52"E, May 9, 2024,



**Figure 5.** Morphology of *Neoarthrinium lithocarpicola* KNUE 24S082. A–B Colony after 7 days of growth at 25°C. The left side shows the front view and the right side shows reverse view, on malt extract agar (A) and potato dextrose agar (B). C–G: Microscopic features of phialides and conidia. C: Conidiogenous cell bearing conidia. D, E: Conidiophores with attached conidia. F, G: Conidia showing diverse shapes. Scale bars = 10 μm.

*N. lithocarpicola* isolated from *D. pacifica*, strain KNUE24P082, NIBRFGC000512625, GenBank No PQ537100 (TUB2), PQ499612 (ITS), PQ555232 (LSU).

**Notes:** The genus *Neoarthrinium* was named due to its morphological similarity to *Arthrinium*, and *Neoarthrinium lithocarpicola*, isolated from *Lithocarpus glaber* (Thunb.) Nakai, was incorporated into the newly established genus *Neoarthrinium* in 2022 [35]. This species has also been reported in terrestrial plants in China and typically exists as a plant endophyte, displaying pathogenic or saprobic traits. Phylogenetically, *N. lithocarpicola* is closely related to *N. moseri*, *N. trachycarpi*, and *N. urticae*, but it is distinguished morphologically by its smaller conidia ( $5\text{--}8.5 \times 4.5\text{--}6\text{ }\mu\text{m}$  in *N. lithocarpicola* versus  $10\text{--}14 \times 3\text{--}4.5\text{ }\mu\text{m}$  in *N. moseri*) [36]. *N. lithocarpicola* can be further differentiated from *N. urticae* by the absence of thick black septa in its conidiophores [37]. While *N. lithocarpicola* shares similar conidiogenous cell and conidial dimensions with *N. trachycarpi*, it is distinguishable by its globose to subglobose conidiogenous cells [38] (Table 3).

***Periconia chimonanthe*** E.F. Yang, H.B. Jiang & Phookamsak, Journal of Fungi 8 (3, no. 243): 18 (2022) [MB#559497] (Figure 6).

**Morphological characteristics of strain KNUE24N115:** After 7 days of growth at 25°C in the dark, colonies on MEA reached a diameter of 22.0–25.0 mm. The colony surface appeared dense, with a brown center that gradually faded toward the edge, turning white at the margin. The reverse side

of the colony was dark brown in the center, lightening to white at the margin. Colonies were flat to slightly raised, with a mildly uneven surface, circular in shape, with an entire margin and a wooly to cottony texture. On PDA, colony diameter ranged from 24.0 to 25.0 mm, showing a grayish olive center that faded toward the edge, ending in a white margin. The reverse side was dark olive green in the center, transitioning to white at the margin. Colonies on PDA were also flat to marginally raised, with a mildly uneven surface, circular shape, entire margin, and wooly to cottony texture. Conidiogenous cells were mono- to polyblastic, occurring singly, upright, lateral, and terminal, ranging from cylindrical to irregular in shape, and appeared subhyaline to brown. They were either discrete or integrated, determinate or inconspicuous, and displayed percurrent proliferations with 1–3 conidiogenous loci, measuring  $(5.67\text{--})7.95\text{--}(10.10) \times (3.67\text{--})4.52\text{--}(5.38)\text{ }\mu\text{m}$  ( $n=20$ ). Conidia were globose to oblong, yellowish to dark brown, smooth to verruculose, occurring either solitarily or in short chains, and measured  $(5.92\text{--})6.76\text{--}(7.89) \times (5.86\text{--})6.68\text{--}(7.66)\text{ }\mu\text{m}$  ( $n=20$ ).

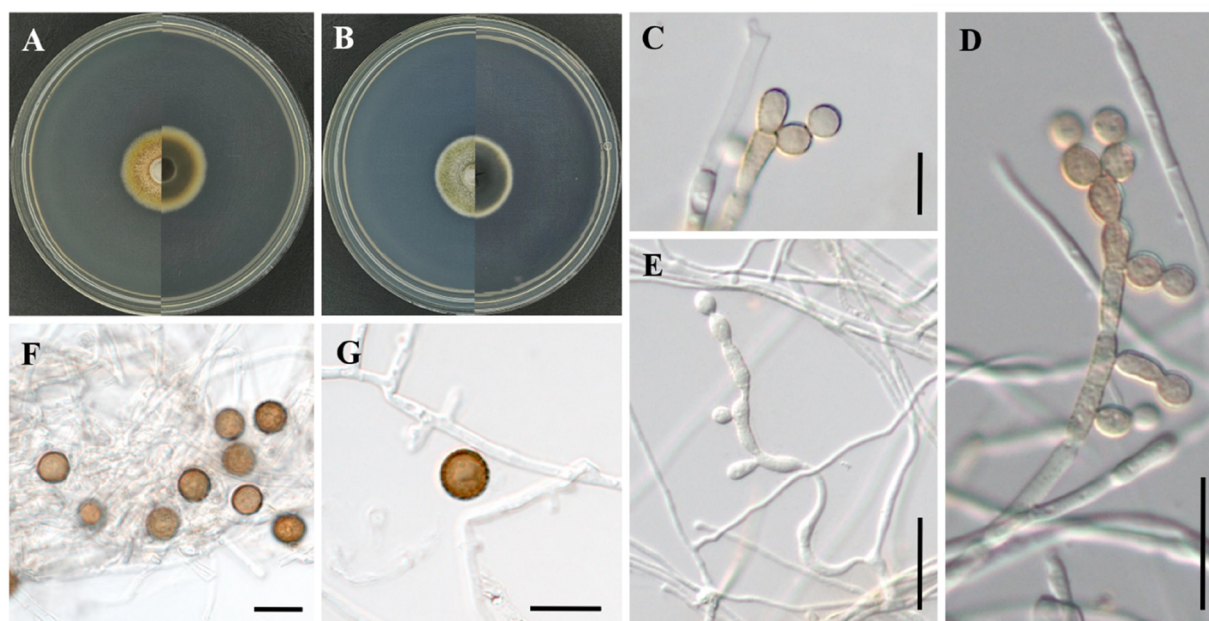
**Specimen examined:** Yeongdeok, Gyeongsangbuk-do, Korea,  $36^{\circ}22'52.644''\text{N}$   $129^{\circ}24'20.52''\text{E}$ , May 9, 2024, *P. chimonanthe* isolated from *D. pacifica*, strain KNUE24P115, NIBRFGC000512624, GenBank No PQ499620 (ITS), PQ499621 (LSU), PQ500557 (SSU), PQ537101 (TEF1).

**Notes:** *P. chimonanthe* was initially reported during the identification of three species of *Periconia* fungi as part of a study isolating several saprobic

**Table 3.** Morphological characteristics of strain KNUE 24S082 and *Neoarthrimum lithocarpicola* CCFCC 54456.

| Characteristics    | <i>Neoarthrimum lithocarpicola</i><br>KNUE 24S146  | <i>Neoarthrimum lithocarpicola</i><br>CCFCC 54456 [35]  |
|--------------------|--|---|
| Colony             | MEA, PDA, 25 °C, 7 days<br><b>MEA</b> , 15.0–30.0 mm, dark grey in center, bright grey to white in margin, reverse side deep yellow in the center, pale yellow in margin, flat, even surface, irregular, filiform margin, cottony texture<br><b>PDA</b> , 14.0–19.00 mm, dark grey in center, bright grey to white in margin, reverse side orange yellow in the center, pale yellow in margin, irregular but nearly circular, entire margin, cottony texture | PDA, 25 °C, 10 days<br>Reaching 60 mm diam, mouse grey to greish-green, flat, spreading, with flocculent aerial mycelium forming concentric rings, edge entire  |
| Asexual morphology | <b>Conidiophores</b> : Cylindrical, septate, branched<br><b>Conidiogenous cells</b> : Erect, blastic, clustered along hyphae, hyaline to pale brown<br><b>Conidia</b> : Brown to dark brown, Ellipsoid, obovoid, acuminate at one end, and polygonal forms, (5.9–)6.7(–7.7) × (3.4–)4.4(–5.0) μm (n=20)  | <b>Conidiophores</b> : Cylindrical, septate, verrucose, flexuous, sometimes reduced to conidiogenous cells<br><b>Conidiogenous cells</b> : Erect, blastic, aggregated in clusters on hyphae, hyaline to pale brown, smooth, globose to subglobose, branched, (4–)5.5–8 × 2.5–3.5(–4) μm ( $\bar{x}$ = 6.6 × 3.1 μm) (n=50)<br><b>Conidia</b> : Brown to dark brown, smooth to finely roughened, subglobose to lenticular, with a longitudinal germ slit, occasionally elongated to ellipsoidal, (5–)6–8(–8.5) × (4.5–)5–5.5(–6) μm ( $\bar{x}$ = 7 × 5.3 μm) (n=50) |

MEA: malt extract agar, PDA: potato dextrose agar.



**Figure 6.** Morphology of *Periconia chimonanthe* KNUE 24S115. A–B Colony after 7 days of growth at 25 °C. The left side shows front view and the right side shows reverse side, on malt extract agar (A) and potato dextrose agar (B). C–G: Microscopic features of conidiogenous and conidia. C: Conidiogenous cells with developing conidia in short chains. D: Polyblastic and variously shaped conidiogenous cells. E: Immature hyaline conidiogenous cells. F–G: Conidia with hyphae. Scale bars: (C, F, G)=10 μm, (D, E)=20 μm.

ascomycetes in China [31]. The holotype was isolated from dead branches of *Chimonanthe praecox* in China. Currently, approximately 40 species are recognized within *Periconia*, which predominantly exist as endophytes and saprobes in plants, though some are known as plant pathogens [31]. *P. chimonanthe* can be differentiated from *P. cortaderiae* by its shorter conidiophores (410–635 × 8.5–12 μm compared to 400–800 × 4–9.4 μm), which are mono- to polyblastic, brown to dark brown, ovoid to subglobose, terminal, proliferative, and possess 1–2 conidiogenous loci. Additionally, *P. chimonanthe* exhibits

larger conidia (7–8 × 6–7 μm compared to 4–6.6 × 4.1–7.1 μm). In contrast, *P. cortaderiae* produces monoblastic conidiogenous cells that are discrete on the stipe, distinguishing it further from *P. chimonanthe* [31, 39] (Table 4).

#### 4. Discussion

This study characterized three previously unrecorded endophytic fungi—*Emericellopsis fuci*, *Neoarthrimum lithocarpicola*, and *Periconia chimonanthe*—isolated from *Dictyopteris pacifica* in Yeongdeok, Korea. The



**Table 4.** Morphological characteristics of strain KNUE 24S115 and *Periconia chimonanthi* KUMCC 20-0266.

| Characteristics    | <i>Periconia chimonanthi</i><br>KNUE 23S115  | <i>Periconia chimonanthi</i><br>KUMCC 20-0266 [31]   |
|--------------------|--|--|
| Colony             | MEA, PDA, 25 °C, 7 days<br><b>MEA</b> , 22.0–25.0 mm, dense, brown in center, gradually fade the edge, white at the margin, reverse side dark brown in the center, white in margin, flat to marginally raised, mildly uneven surface, circular, entire margin, wooly to cottony texture<br><b>PDA</b> , 24.0–25.0 mm, dense, grayish olive at center, gradually fade the edge, white at the margin, reverse side dark olive green in the center, white in margin, flat to marginally raised, mildly uneven surface, circular, entire margin, wooly to cottony texture                                      | PDA, 20–25 °C, normal light, 7 days<br>15 mm, dense, circular, flattened to slightly raised, surface slightly rough, with entire edge, floccose to cottony, radially furrowed at the margin, pale greenish grey at the margin, dark greenish toward the center from above and below; not producing pigmentation on medium  |
| Asexual morphology | <b>Conidiogenous cells</b> : Mono to polyblastic, occurring singly, upright, lateral, and terminal, cylindrical to irregular, subhyaline to brown, discrete or integrated, determinate, or inconspicuous, percurrent proliferations, with 1–3 conidiogenous loci, $(5.67\text{--}7.95\text{--}(10.10)\times(3.67\text{--}4.52\text{--}(5.38))\text{ }\mu\text{m}$ ( $n=20$ )<br><b>Conidia</b> : Globose to oblong, yellowish to dark brown, smooth to verruculose, solitary or in short chains, $(5.92\text{--}6.76\text{--}(7.89)\times(5.86\text{--}6.68\text{--}(7.66))\text{ }\mu\text{m}$ ( $n=20$ ) | <b>Conidiogenous cells</b> : Polyblastic, solitary, erect, lateral and terminal, cylindrical to irregular, luteous to brown, discrete or integrated, determinate, or inconspicuous, percurrent proliferations, with 1–3 conidiogenous loci, $7\text{--}10\times4.5\text{--}6\text{ }\mu\text{m}$ ( $\bar{x}=8.8\times4.9\text{ }\mu\text{m}$ ) ( $n=20$ )<br><b>Conidia</b> : Globose to oblong, or ellipsoidal, subhyaline to brown or dark brown, smooth to verruculose, solitary or in short chains, $6\text{--}8\times6\text{--}8\text{ }\mu\text{m}$ ( $\bar{x}=7.1\times6.9\text{ }\mu\text{m}$ ) ( $n=20$ ) |

MEA: malt extract agar, PDA: potato dextrose agar.

identification was achieved through morphological observation and robust molecular phylogenetic analysis using multiple genetic markers (ITS, LSU, SSU, TEF1, TUB2, and RPB2 sequences). Numerous studies have demonstrated that species within the genus *Emericellopsis* are capable of producing a variety of bioactive metabolites with antimicrobial properties against plant and human pathogens, as well as anticancer effects [40]. Additionally, over 100 bioactive secondary metabolites have been identified from fungi of the genus *Periconia* since 1969, including compounds like Taxol (an anticancer agent) and Piperine (an antimicrobial agent), which has heightened research interest in this genus [41]. These findings suggest that *E. fuci* and *P. chimonanthi* may also possess the capability to produce functional secondary metabolites with potential biotechnological applications. While little is known about the bioactive properties of *N. lithocarpicola*, the discovery that both *P. chimonanthi*, previously known as a terrestrial endophyte, and *N. lithocarpicola* can form symbiotic associations with seaweeds is noteworthy. This expands our understanding of the ecological versatility and adaptability of these fungal species in marine environments.

The isolation of *Emericellopsis fuci*, *Neoarthrimum lithocarpicola*, and *Periconia chimonanthi* from *Dictyopteris pacifica* highlights the diverse fungal communities associated with seaweeds in Korea's unique intertidal zones. *D. pacifica* serves as a valuable host, providing a stable environment that supports the growth and symbiotic relationships of these endophytes, which likely enhance the host's resilience against environmental stressors such as fluctuating salinity and temperature and defend against herbivorous organisms through the production of

secondary metabolites. Furthermore, the successful isolation and identification of these species underscore the potential of Korean coastal ecosystems as reservoirs of marine endophytic fungi with diverse ecological roles and biotechnological potentials, including nutrient cycling, enhancement of host stress tolerance, and defense against pathogens and herbivores, thereby contributing to the overall health and sustainability of their seaweed hosts. This study advances our understanding of fungal diversity associated with native Korean seaweeds, particularly *D. pacifica*, and highlights marine endophytic fungi as promising sources of novel bioactive compounds. Future research should focus on elucidating the specific ecological roles of these fungi and exploring their applications in pharmaceuticals, agriculture, and biotechnology, as well as investigating the biosynthetic pathways responsible for the production of secondary metabolites to discover new natural products with therapeutic and industrial significance.


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