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## *Bimuria omanensis* sp. nov. (Didymosphaeriaceae, Pleosporales) from Oman

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### Abstract

*Bimuria* is a monotypic genus in Didymosphaeriaceae, typified by *B. novae-zelandiae* collected in terrestrial habitats from New Zealand soil. In our study, *Bimuria omanensis*, a novel species was isolated from unidentified decaying leaves in Oman. The phylogenetic placement of *B. omanensis* is resolved based on maximum likelihood, maximum parsimony and Bayesian analyses of combined LSU, ITS and TEF1- $\alpha$  sequence data of Didymosphaeriaceae. The placement of *Bimuria omanensis* as a distinct species, is confirmed based on phylogeny. This is the first record of an asexual morph in *Bimuria* and first record of a *Bimuria* species from Oman. The relationship of this taxon with other phylogenetically closely related Didymosphaeriaceae species is shown.

**Keywords:** 1 new species, Dothideomycetes, first record, morphology, phylogeny

### Introduction

The family Didymosphaeriaceae was introduced by Munk (1953) and is typified by *Didymosphaeria*. Generally, species in family Didymosphaeriaceae are characterized by uni-septate ascospores with trabeculate pseudoparaphyses (Aptroot 1995, Liew *et al.* 2000, Ariyawansa *et al.* 2014a, b, Wanasinghe *et al.* 2016). They occur as saprobes, endophytes or pathogens associated with various plant substrates worldwide (Ariyawansa *et al.* 2014b, Liu *et al.* 2015, Tennakoon *et al.* 2016, Wanasinghe *et al.* 2016) and are rarely parasitic on other fungi (Hyde *et al.* 2013, Ariyawansa *et al.* 2014b). In previous classifications, the placement of Didymosphaeriaceae was doubtful. von Arx & Müller (1975) considered the family as a synonym of Pleosporaceae while Lumbsch & Huhndorf (2007) assigned Didymosphaeriaceae to Montagnulaceae. However, Ariyawansa *et al.* (2014b) provided a well-resolved phylogenetic analysis and morphological comparison for Didymosphaeriaceae. Therefore, Montagnulaceae was synonymized under Didymosphaeriaceae (Ariyawansa *et al.* 2014a, Feng *et al.* 2019). Previously, there were several studies on Didymosphaeriaceae and the most recent studies are those of Wanasinghe *et al.* (2016, 2018), Tibpromma *et al.* (2018)

and Phookamsak *et al.* (2019). Wijayawardene *et al.* (2018) accepted 26 genera in Didymosphaeriaceae. Currently, there are 32 genera accounted into this family (Hongnanan *et al.* 2020).

The genus *Bimuria* belongs in Didymosphaeriaceae although, it has been introduced by Hawksworth *et al.* (1979) and placed in Pleosporaceae. Hawksworth *et al.* (1979) suggested a close affinity of *Bimuria* to *Montagnula* species based on morphological characters such as shape of ascomata, asci type and trabeculate pseudoparaphyses (sensu Liew *et al.* 2000). Further, they mentioned that these genera can be separated based on the carbonaceous peridium of *Montagnula*, while ascospore formed as a mass outside of the ostiole of *Bimuria* (Hawksworth *et al.* 1979, Ariyawansa *et al.* 2014a). Barr (1987) transferred *Bimuria* to Phaeosphaeriaceae and Lumbsch & Huhndorf (2007) placed it in Melanommataceae. Multi-gene phylogenetic analysis (Schoch *et al.* 2009) confirmed that *Bimuria* is a well-resolved genus in Montagnulaceae. Ranghoo & Hyde (1999) reported that *Bimuria novae-zelandiae* closely resembles *Ascomauritiana lignicola* in its ascospores characteristics. *Ascomauritiana lignicola* however, differs to *B. novae-zelandiae* by having 4-spored unitunicate asci, while *B. novae-zelandiae* has 2-spored bitunicate asci (Hawksworth *et al.* 1979, Ranghoo & Hyde 1999, Ariyawansa *et al.* 2014a).

*Bimuria* is presently monotypic (Index Fungorum 2020). Previously, *Bimuria novae-zelandiae* was recorded from a terrestrial habitat as a saprobe from soil in New Zealand (Hawksworth *et al.* 1979). In our study, a novel saprobic fungus, *Bimuria omanensis* is introduced from unidentified decaying leaves in Oman. Hawksworth *et al.* (1979) suggested that ascospore release and the large, thick-walled ascospores may be an adaptation for survival in soil-borne habitats. This study is the first record of the asexual morph of *Bimuria* and first record of *Bimuria* species in Oman. Illustrations with comprehensive morphological descriptions and rDNA analysis based on ITS, LSU, and TEF1- $\alpha$  sequence data are provided for the novel species.

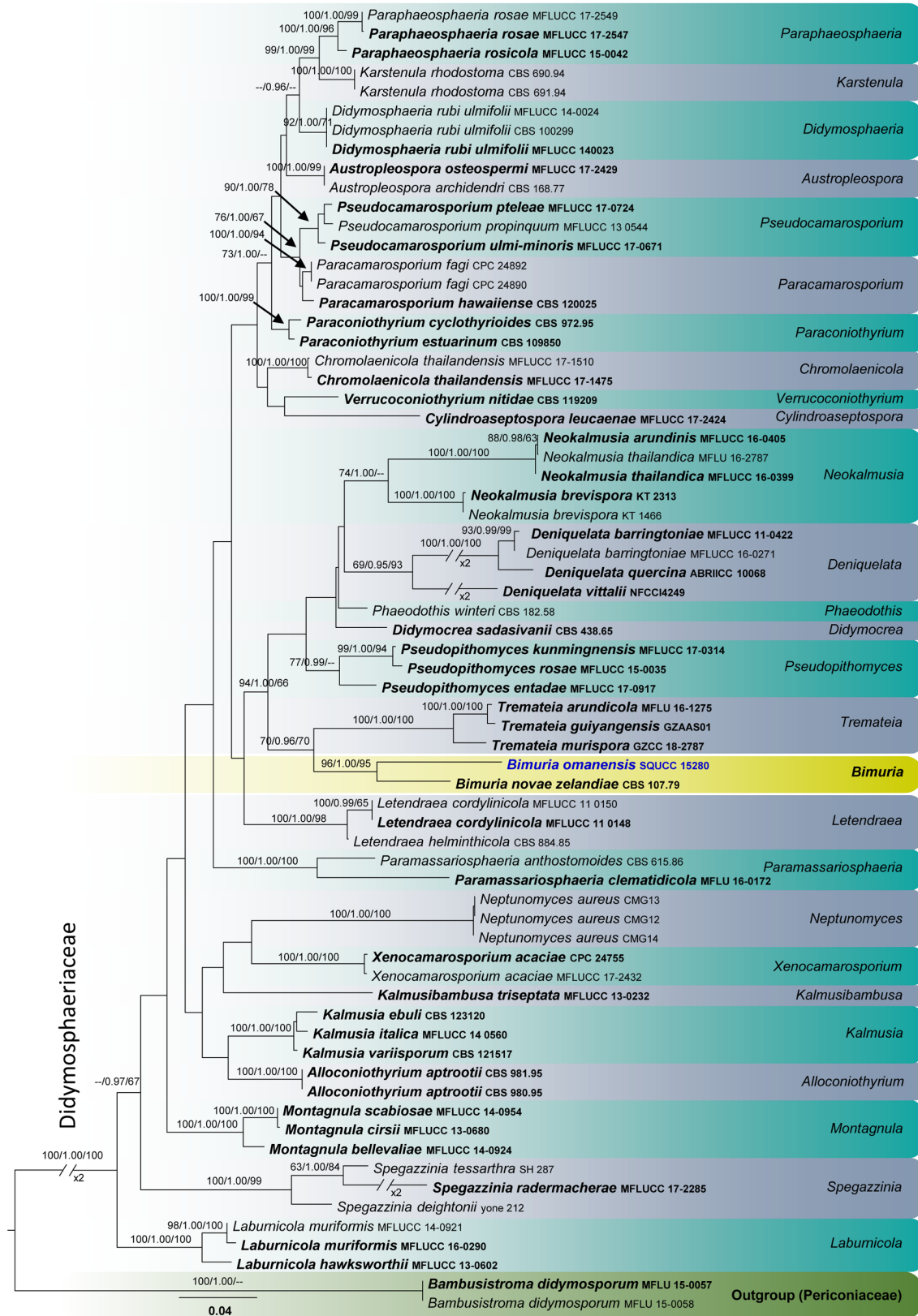
## Materials and methods

### *Sample collection, morphological studies and specimen deposition*

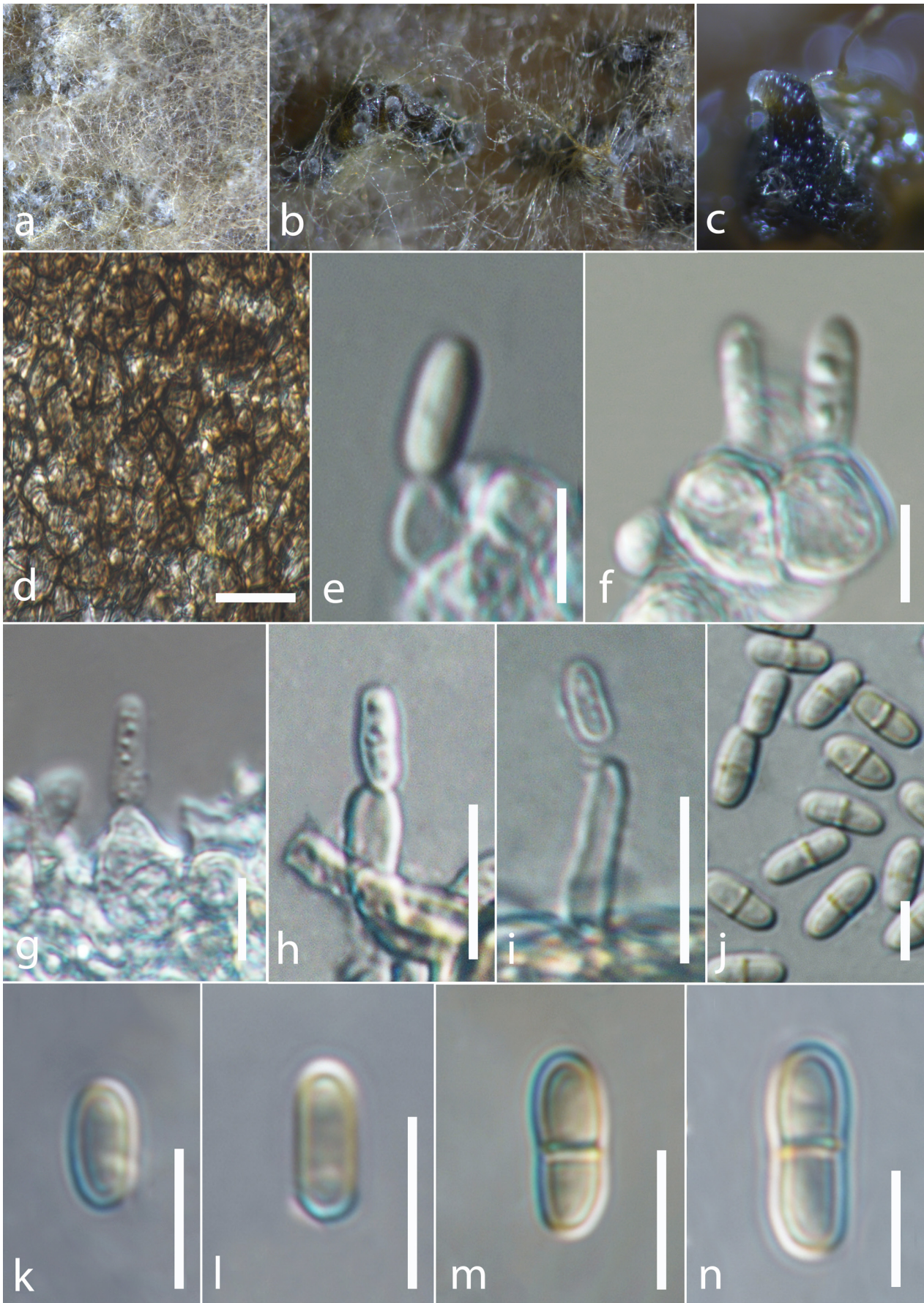
The specimens were collected from a dead leaves of an unknown host plant in Oman (July, 2016). The samples were taken to the laboratory in a zip lock bag. Single spore isolation was carried out as described by Chomnunti *et al.* (2014). Germinated spores were aseptically transferred in to small potato dextrose agar (PDA) plates. The plates were incubated at 25 °C for 10 to 15 days for pure cultures. Colonies were observed weekly. Photographs of enlarged structures in culture were taken by using a Carl Zeiss Stemi 508 stereo microscope. Micro morphological characters were examined and photographed using a Nikon Eclipse Ni-U microscope. All structures (peridium cells, conidiogenous cells and conidia) were processed for photographs by using the water mounted clean glass slides. The measurements of photomicrograph structures were measured using NIS-element D software. Images used for figures were processed with Adobe Photoshop CS6 Extended version 13.0.1 software (Adobe Systems, USA). After two weeks, the well grown cultures were used for DNA extraction. The holotype material is deposited at Sultan Qaboos University Herbarium and the living culture is deposited at Sultan Qaboos University Culture Collection. Facesoffungi and Index Fungorum numbers were registered (Jayasiri *et al.* 2015, Index Fungorum 2020).

### *DNA extraction, PCR amplification and sequencing*

Genomic DNA was extracted from the scraped fresh fungal mycelium grown on PDA for 2 weeks at 25 °C by using a modified protocol in Al-Sadi *et al.* (2012). DNA to be used as template for PCR was stored at 4 °C for use in regular work and duplicated at -20 °C for long-term storage. DNA sequence data were obtained from the sequences of three loci, internal transcribed spacers (ITS), large subunit (LSU) and translation elongation factor-1 alpha (TEF1- $\alpha$ ) by amplifying primer pair ITS5/ ITS4, LR0R/ LR5 and EF1 983F/EF1-2218R respectively (Vilgalys & Hester 1990, White *et al.* 1990, Rehner & Buckley 2005). Amplifications were carried out in an Applied Biosystems ProFlex PCR System (Life Technologies, USA) with the following profile: an initial denaturation at 94 °C for 3 min, followed by 40 cycles of denaturation at 94 °C for 45 s, annealing at 56 °C for 50 s and extension at 72 °C for 1 min, and a final extension at 72 °C for 10 min. Sequencing of the PCR amplicons were conducted using the same primers used for the amplification reactions. The PCR products were verified by staining with ethidium bromide on 1 % agarose electrophoresis gels. DNA sequences were deposited in GenBank.



**FIGURE 1.** RAxML tree based on analysis of a combined dataset of LSU, ITS and TEF1- $\alpha$  partial sequence data. Bootstrap support values for ML and MP equal to or greater than 60 %, Bayesian posterior probabilities (PP) equal to or greater than 0.95 are defined as ML/PP/MP above the nodes. Genera, where known are indicated with coloured blocks. The new isolate is in blue. The type strains are in bold. The scale bar represents the expected number of nucleotide substitutions per site. *Bambusistroma didymosporum* (MFLU 15-0057, MFLU 15-0058) is used as the outgroup taxon.



**FIGURE 2.** *Bimuria omanensis* (SQU H-115, Holotype) **a.** Hyphae. **b–c.** Sporulating conidiomata in culture media **d.** Conidioma wall. **e–i.** Conidiogenous cells. **j–n.** Conidia. Scale bars: d = 20  $\mu$ m, h–i = 10  $\mu$ m, e–g, j–n = 5  $\mu$ m.

**TABLE 1.** Taxa used for molecular study and their GenBank numbers.

Species	Strain /Voucher*	GenBank accessions		
		ITS	LSU	TEF1- $\alpha$
<i>Alloconiothyrium aptrootii</i>	CBS 980.95 <sup>T</sup>	JX496121	JX496234	-
<i>Alloconiothyrium aptrootii</i>	CBS 981.95 <sup>T</sup>	JX496122	JX496235	-
<i>Austropleospora archidendri</i>	CBS 168.77	JX496049	JX496162	-
<i>Austropleospora osteospermi</i>	MFLUCC 17-2429 <sup>T</sup>	MK347757	MK347974	MK360044
<i>Bambusistroma didymosporum</i>	MFLU 15-0057 <sup>T</sup>	KP761733	KP761730	KP761727
<i>Bambusistroma didymosporum</i>	MFLU 15-0058	KP761734	KP761731	KP761728
<b><i>Bimuria omanensis</i></b>	<b>SQUCC 15280<sup>T</sup></b>	<b>MT274326</b>	<b>MT271820</b>	<b>MT279046</b>
<i>Bimuria novae zelandiae</i>	CBS 107.79 <sup>T</sup>	MH861181	AY016356	DQ471087
<i>Chromolaenicola thailandensis</i>	MFLUCC 17-1510	MN325018	MN325006	MN336551
<i>Chromolaenicola thailandensis</i>	MFLUCC 17-1475 <sup>T</sup>	MN325019	MN325007	MN335652
<i>Cylindroaseptospora leucaenae</i>	MFLUCC 17-2424 <sup>T</sup>	NR_163333	NG_066310	MK360047
<i>Deniquelata barringtoniae</i>	MFLUCC 11-0422 <sup>T</sup>	NR_111779	NG_042696	-
<i>Deniquelata barringtoniae</i>	MFLUCC 16-0271	MH275059	MH260291	MH412766
<i>Deniquelata quercina</i>	ABRIICC 10068 <sup>T</sup>	MH316153	MH316157	-
<i>Deniquelata vittalii</i>	NFCCI4249 <sup>T</sup>	MF406218	MF182395	MF182398
<i>Didymocrea sadasivani</i>	CBS 438.65 <sup>T</sup>	MH858658	DQ384103	-
<i>Didymosphaeria rubi ulmifolii</i>	CBS 100299	MH862698	JX496124	-
<i>Didymosphaeria rubi ulmifolii</i>	MFLUCC 14-0023 <sup>T</sup>	KJ436586	KJ436586	-
<i>Didymosphaeria rubi ulmifolii</i>	MFLUCC 14-0024	-	KJ436585	-
<i>Kalmusia ebuli</i>	CBS 123120 <sup>T</sup>	KF796674	JN644073	-
<i>Kalmusia italica</i>	MFLUCC 14-0560 <sup>T</sup>	KP325440	KP325441	-
<i>Kalmusia variisporum</i>	CBS 121517 <sup>T</sup>	NR_145165	JX496143	-
<i>Kalmusibambusa triseptata</i>	MFLUCC 13-0232 <sup>T</sup>	KY682697	KY682695	-
<i>Karstenula rhodostoma</i>	CBS 690.94	-	GU301821	GU349067
<i>Karstenula rhodostoma</i>	CBS 691.94	LC014559	AB807531	AB808506
<i>Laburnicola hawksworthii</i>	MFLUCC 13-0602 <sup>T</sup>	KU743194	KU743195	-
<i>Laburnicola muriformis</i>	MFLUCC 16-0290 <sup>T</sup>	KU743197	KU743198	KU743213
<i>Laburnicola muriformis</i>	MFLUCC 14-0921	KU743200	KU743201	-
<i>Letendreaea cordylinicola</i>	MFLUCC 11-0150	KM213996	KM213999	-
<i>Letendreaea cordylinicola</i>	MFLUCC 11-0148 <sup>T</sup>	NR_154118	NG_059530	-
<i>Letendreaea helminthicola</i>	CBS 884.85	MK404145	AY016362	MK404174
<i>Montagnula bellevaliae</i>	MFLUCC 14-0924 <sup>T</sup>	KT443906	KT443902	KX949743
<i>Montagnula cirsii</i>	MFLUCC 13-0680 <sup>T</sup>	KX274242	KX274249	KX284707
<i>Montagnula scabiosae</i>	MFLUCC 14-0954 <sup>T</sup>	KT443907	KT443903	-
<i>Neokalmusia arundinis</i>	MFLUCC 16-0405 <sup>T</sup>	KY706142	KY706132	KY706145
<i>Neokalmusia brevispora</i>	KT 2313 <sup>T</sup>	LC014574	AB524601	AB539113
<i>Neokalmusia brevispora</i>	KT 1466	LC014573	AB524600	AB539112
<i>Neokalmusia thailandica</i>	MFLU 16-2787 <sup>T</sup>	NR_154255	NG_059792	-
<i>Neokalmusia thailandica</i>	MFLUCC 16-0399	KY706141	KY706131	-
<i>Neptunomyces aureus</i>	CMG12	MK912121	-	MK948000
<i>Neptunomyces aureus</i>	CMG13	MK912122	-	MK948001
<i>Neptunomyces aureus</i>	CMG14	MK912123	-	MK948002
<i>Paracamarosporium fagi</i>	CPC 24890	KR611886	KR611904	-
<i>Paramassariosphaeria anthostomoides</i>	CBS 615.86	MH862005	GU205223	-

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TABLE 1 (Continued)

Species	Strain /Voucher*	GenBank accessions		
		ITS	LSU	TEF1- $\alpha$
<i>Paraphaeosphaeria rosae</i>	MFLUCC 17-2547 <sup>T</sup>	MG828935	MG829044	MG829222
<i>Paraphaeosphaeria rosae</i>	MFLUCC 17-2549	MG828937	MG829046	MG829223
<i>Paraphaeosphaeria rosicola</i>	MFLUCC 15-0042 <sup>T</sup>	NR_157528	MG829047	-
<i>Phaeodothis winteri</i>	CBS 182.58	-	GU301857	-
<i>Pseudocamarosporium propinquum</i>	MFLUCC 13 0544	KJ747049	KJ813280	-
<i>Pseudocamarosporium pteleae</i>	MFLUCC 17-0724 <sup>T</sup>	NR_157536	MG829061	MG829233
<i>Pseudocamarosporium ulmi-minoris</i>	MFLUCC 17-0671 <sup>T</sup>	NR_157537	MG829062	-
<i>Pseudopithomyces entadae</i>	MFLUCC 17-0917 <sup>T</sup>	-	NG_066305	MK360083
<i>Pseudopithomyces kunmingensis</i>	MFLUCC 17-0314 <sup>T</sup>	MF173607	MF173605	-
<i>Pseudopithomyces rosae</i>	MFLUCC 15-0035 <sup>T</sup>	MG828953	MG829064	-
<i>Spegazzinia deightonii</i>	yone 212	-	AB807582	AB808558
<i>Spegazzinia radermacherae</i>	MFLUCC 17-2285 <sup>T</sup>	MK347740	MK347957	MK360088
<i>Spegazzinia tessartha</i>	SH 287	JQ673429	AB807584	AB808560
<i>Tremateia arundicola</i>	MFLU 16-1275 <sup>T</sup>	KX274241	KX274248	KX284706
<i>Tremateia guiyangensis</i>	GZAAS01 <sup>T</sup>	KX274240	KX274247	KX284705
<i>Tremateia murispora</i>	GZCC 18-2787 <sup>T</sup>	NR_165916	MK972751	MK986482
<i>Verrucoconiothyrium nitidae</i>	CBS 119209	EU552112	EU552112	-
<i>Xenocamarosporium acaciae</i>	CPC 24755 T	NR_137982	NG_058163	-
<i>Xenocamarosporium acaciae</i>	MFLUCC 17-2432	MK347766	MK347983	MK360093

Newly generated sequences are indicated in bold. \* Type strains are marked with superscript <sup>T</sup> and “-” sequences are not available.

Abbreviations: **AFTOL**—Assembling the Fungal Tree of Life; **CDA**—Canadian Department of Agriculture, Ottawa, Canada; **CPC**—Cultures of Pedro Crous, housed at Westerdijk Fungal Biodiversity Institute; **CBS**—Centraalbureau voor Schimmelcultures, Utrecht, The Netherlands; **CMG**—M. Gonçalves, **GZAAS**—Guizhou Academy of Agricultural Sciences, Guiyang, China; **KT**—Kazuaki Tanaka; **MFLUCC**—Mae Fah Luang University Culture Collection, Chiang Rai, Thailand; **MFLU**—Mae Fah Luang University Herbarium, Chiang Rai, Thailand; **SH**—S. Hatakeyama; **SQUCC**—Sultan Qaboos University Culture Collection.; **UTHSC**—University of Tennessee Health Science Center, Memphis, USA; **yone**—H. Yonezawa.

### Molecular data analyses

Lasergene SeqMan Pro v.7 was used to obtain consensus sequences from sequences generated from forward and reverse primers. Contig sequences were checked with BLAST searches to recognize high similarity indices and the related literature (Wanasinghe *et al.* 2018, Phookamsak *et al.* 2019) were followed (TABLE 1). The single and multiple (LSU, ITS and TEF1- $\alpha$ ) alignments of all reference sequences, were automatically generated with MAFFT v. 7 (<http://mafft.cbrc.jp/alignment/server/index.html>, Katoh & Standley 2013, Katoh *et al.* 2019). BioEdit v. 7.0.5.2 software was used when manual improvement is needed (Hall 1999). The terminal ends and ambiguous regions of the alignment were deleted.

Phylogenetic analyses of both individual and combined datasets were based on maximum-likelihood (ML), maximum parsimony (MP) and Bayesian criteria (BI). Both RAxML and Bayesian analyses were run on the CIPRES Science Gateway portal (Miller *et al.* 2012). ML bootstrap values equal or greater than 60 % are given above each node (first value, FIGURE 1). MrModeltest v. 2.3 (Nylander 2004) was run under the AIC (Akaike Information Criterion) implemented in PAUP v. 4.0b10. The evaluated best-fit model was GTR + I + G for each locus in both ML and BI. ML analyses for the data-sets were performed with RAxML-HPC2 on XSEDE v. 8.2.10 (Stamatakis 2014) using a GTR+GAMMA substitution model with 1000 bootstrap iterations. The BI analysis was computed with MrBayes v. 3.2.6 (Ronquist *et al.* 2012). Six simultaneous Markov chains were run for 2,000,000 generations, and trees were sampled every 100th generation, ending the run automatically when standard deviation of split frequencies dropped below 0.01. BI posterior probabilities (BYPP) greater than 0.95 are given above each node as the second value

(FIGURE 1). MP was carried with the heuristic search option in PAUP (Phylogenetic Analysis Using Parsimony) v. 4.0b10 with the following parameter settings: characters unordered with equal weight, random taxon addition, branch swapping with tree bisection-reconnection (TBR) algorithm, branches collapsing if the maximum branch length was zero. Alignment gaps were treated as missing characters in the analysis of the combined data set, where they occurred in relatively conserved regions. Trees were inferred using the heuristic search option with 1000 random sequence additions, with maxtrees set at 1000. Descriptive tree statistics for parsimony; Tree Length (TL), Consistency Index (CI), Retention Index (RI), Relative Consistency Index (RC) and Homoplasy Index (HI) were calculated for generated trees. The Kishino-Hasegawa tests (Kishino & Hasegawa 1989) were performed in order to determine whether trees were significantly different. MP bootstrap values equal or greater than 60 % are given above each node (third value, FIGURE 1). Phylograms were visualized with FigTree v1.4.0 program (Rambaut 2012) and reorganized in Microsoft power point (2016). The finalized alignment and tree were deposited in TreeBASE (submission ID: 26053).

## Results

### *Phylogenetic analyses*

The combined LSU, ITS and TEF1- $\alpha$  alignment was used to construct the final phylogenetic analyses of ML, MP and BI (FIGURE 1). Topologies of trees (under ML, MP and BI criteria) recovered for each gene dataset were visually compared and the overall tree topology was congruent to those obtained from the combined dataset. The RAxML analysis of the combined dataset yielded a best scoring tree (FIGURE 1) with a final ML optimization likelihood value of -16230.039098. The matrix had 945 distinct alignment patterns, with 30.68 % proportion of gaps and completely undetermined characters in this alignment. Parameters for the GTR+I+G model of the combined LSU, ITS and TEF1- $\alpha$  were as follows: Estimated base frequencies were as follows: A = 0.22673, C = 0.268772, G = 0.279874, T = 0.224625; substitution rates AC = 1.515326, AG = 2.346647, AT = 1.538961, CG = 1.166802, CT = 6.724986, GT = 1.000; proportion of invariable sites I = 0.473655; gamma distribution shape parameter  $\alpha$  = 0.472834. The maximum parsimonious dataset for the combined gene sequences consisted of 2440 characters, of which 1627 were constant, 642 (26.31 %) parsimony-informative and 171 parsimony-uninformative. The parsimony analysis of the data matrix resulted in the maximum of four equally most parsimonious trees with a length of 2730 steps (CI = 0.457, RI = 0.683, RC = 0.312, HI = 0.543) in the first tree. The BI analysis ran 1450000 generations before the average standard deviation for split frequencies reached below 0.01 (0.009899). The alignment contained a total of 1201 unique site patterns. After discarding the first 20 % of generations, 11601 trees remained from which 50 % consensus trees and posterior probabilities (BYPP) were calculated.

Newly generated sequences from our new isolate, *Bimuria omanensis* (SQUCC 15280), showed an affinity to *B. novae-zelandiae* (CBS 107.79) in the combined phylogeny (FIGURE 1) and this relationship received 96% ML, 95% MP and 1.00 BI support. *Bimuria* and *Tremateia* are closely related in the phylogenetic analysis with 70% ML, 70% MP and 0.96 BI support (FIGURE 1). In the combined sequence analysis, with the exception of *Cylindroaseptospora*, *Didymocrea*, *Kalmusibambusa*, *Neokalmusia*, *Phaeodothis* and *Verrucoconiothyrium*, all other genera had high bootstrap support (FIGURE 1).

## Taxonomy

*Bimuria* D. Hawksw., Chea & Sheridan, N.Z. Jl Bot. 17(3): 267 (1979) **amend.**

*Saprobic* in terrestrial habitats. **Sexual morph:** See Hawksworth *et al.* (1979) and Ariyawansa *et al.* (2014a). **Asexual morph:** Coelomycetous. *Conidiomata* pycnidial, arise on mycelia as black spore mass, aggregated clusters are scattered, irregular and superficial to semi-immersed. *Conidiomatal wall* composed of thick walled, pale to dark brown cells of *textura angularis*. *Conidiogenous cells* enteroblastic, phialidic, ampulliform or short cylindrical, determinate, sometimes cylindrical, elongated neck, rough and hyaline. *Conidia* oblong to cylindrical, 1-septate, smooth and thin-walled, pale brown to hyaline.

Type species:—*Bimuria novae-zelandiae* D. Hawksw., Chea & Sheridan, N.Z. Jl Bot. 17(3): 268 (1979)

Notes:—Hawksworth *et al.* (1979) introduced *Bimuria* and it was placed in Pleosporaceae based on its sexual morphology. Based on phylogenetic analysis of SSU, LSU, RPB2 and TEF1- $\alpha$  sequence data, Schoch *et al.* (2009) and Ariyawansa *et al.* (2014a) confirmed that the *Bimuria novae-zelandiae* (CBS 107.79) should be placed in Montagnulaceae (= Didymosphaeriaceae) and related to *Tremateia*. In this current study, we observed that our novel strain (SQUCC 15280) clusters with *Bimuria novae-zelandiae* with strong bootstrap support in our phylogenetic analyses (Fig. 1). Therefore, we conclude that it is appropriate to consider our isolate as a species in *Bimuria*. *Bimuria* was only known from its sexual morph and we amend *Bimuria* in order to accommodate its coelomycetous asexual morph from our novel taxonomic account.

*Bimuria omanensis* Wijesinghe, Wanas., Al-Sadi, K.D. Hyde & Maharachch., *sp. nov.*

*Index Fungorum number*: IF557436; *Facesoffungi number*: FoF 07928; FIGURE 2.

**Etymology**:—Name reflects the county Oman, from where the species was isolated.

**Holotype**:—SQU H-115

**Asexual morph**: Coelomycetous. *Conidiomata* pycnidial, arise on mycelia as black spore mass, aggregated clusters are scattered, irregular and superficial to semi-immersed. *Conidiomatal wall* composed of thick-walled, pale to dark brown cells of *textura angularis*. *Conidiogenous cells* 8–9  $\times$  7–8  $\mu\text{m}$  ( $\bar{x}$  = 8.36  $\times$  7.6  $\mu\text{m}$ , n=10), enteroblastic, phialidic, ampulliform or short cylindrical, determinate, sometimes cylindrical, with elongate neck, rough and hyaline. *Conidia* 7–10  $\times$  3–4.5  $\mu\text{m}$  ( $\bar{x}$  = 8.84  $\times$  3.94  $\mu\text{m}$ , n = 25) oblong to cylindrical, 1- septate, smooth and thin walled, hyaline to pale brown.

**Culture characteristics**:—Colonies on PDA reaching 60 mm diam. after 14 days at 24 °C, dark grey to brown in upper surface.

**Known distribution**:—Oman (this study)

**Material examined**:—OMAN, The Jebel Akhdar, Dakhiliyah Governorat, on decaying leaves of unidentified plant, July 2016, SSN Maharachchikumbura OM09 (SQU H-115, holotype), ex-type living culture SQUCC 15280.

**Gene sequence data**: ITS (MT274326), LSU (MT271820), TEF-1 $\alpha$  (MT279046)

**Notes**:—In our DNA sequence analysis, *Bimuria novae-zelandiae* (CBS 107.79) and *B. omanensis* (SQUCC 15280) are monophyletic with strong bootstrap support (Fig. 1). Morphological comparison between these taxa are currently impossible as *B. novae-zelandiae* is known from its sexual morph and only the asexual morph is known for *B. omanensis*. Comparison of the 570 ITS (+5.8S) nucleotides of these strains reveals 70 (12.3%) nucleotide differences. This could be due to ITS polymorphisms (Stadler *et al.* 2020) and it is not surprising that these strains appear to belong to the same species. A comparison of the 852 nucleotides across the TEF-1 $\alpha$  region revealed 32 bp (3.75%) differences between these strains suggesting these are distinct species (Jeewon & Hyde 2016). *Bimuria novae-zelandiae* was isolated from soil of a barley field in New Zealand (Hawksworth *et al.* 1979). *Bimuria omanensis* was collected from a decaying leaf of a desert shrub in Ad Dakhiliyah Governorate, Oman. Therefore, based on the molecular data and habitat differences, we conclude that these two taxa are distinct species. *Didymosporina aceris*, *Gordonomyces mucovaginus* and *Lichenodiplis lecanorae* share similar conidial morphology to *Bimuria omanensis* (Wijayawardene *et al.* 2016). Phylogenetically *Gordonomyces mucovaginus* and *Lichenodiplis lecanorae* are not closely related to *Bimuria omanensis* (data not shown) and sequence data are unavailable for *Didymosporina aceris*.

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