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Sexual morph of *Alternaria scirpivora* (*Alternaria* section *Nimbya*) from Iran

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Abstract: Species of the *Alternaria* section *Nimbya* are plant pathogens or saprophytes associated mainly with the plants in the *Cyperaceae* and *Juncaceae* families. Until now, five species from this section have been reported in Iran, but their sexual morphs have not been identified. In the present study, the sexual morph of *Alternaria scirpivora* was induced under laboratory conditions on PCA medium containing autoclave sterilized culms of *Scirpus acutus* incubated at 23–25 ºC for 90 days. Mature ascomata were formed after 2–3 months, containing asci and ascospores. The morphological characteristics of the sexual and asexual morphs are described and the phylogenetic relationships of *A. scirpivora* with closely related species are discussed. This is the first report of the sexual stage of this species in Iran and Asia.

Keywords: *Alternaria*, Ascomata, Homothallism, Morphology, Molecular phylogeny.

INTRODUCTION

The genus *Alternaria* was established by Nees (1816), with *A. tenuis* Nees as the type species (syn.: *A. alternata* (Fr.) Keissl.), and is currently recognized as a well-known genus within the family *Pleosporaceae*, *Pleosporales*, *Dothideomycetes* (Woudenberg et al. 2013, Lawrence et al. 2016). *Alternaria* is characterized by the production of mononematous, macro- or micronematous, simple or branched conidiophores, integrated or discrete, mono- to poly-tretic conidiogenous cells, solitary or catenate, straight or curved, phragmo- or dictyo-septate, smooth or verrucose and median brown to dark brown conidia with a rounded or narrowly-beaked tip (Woudenberg et al. 2013, 2014, 2015, Li et al. 2023). *Alternaria* species have diverse lifestyles, functioning as destructive plant pathogens, epiphytes, saprophytes, or endophytes, with a worldwide distribution. They have been reported from plants in both terrestrial and aquatic environments, as well as in the air. Some species have emerged as animal or human pathogens, causing skin infections, allergic sinusitis, keratitis, and facial osteomyelitis, mainly in immunocompromised patients (Lawrence et al. 2012, 2013, 2016, Woudenberg et al. 2013, 2014, 2015, Li et al. 2023). The taxonomy of the genus *Alternaria* is complex and has undergone major revisions over time, particularly through the use of molecular techniques. Simmons (1992, 2007) used two morphological criteria, three-dimensional sporulation patterns, and conidial morphology, of the isolates under standard culture conditions (PCA culture medium, under white fluorescent light with 8/16 h light/dark cycle, at 23–25 ºC, and without humidity control) to differentiate species boundaries and identified nearly 280 species, grouped in three sections (small-, medium- and large-spored *Alternaria*). Woudenberg et al. (2013) revised the classification of *Alternaria* through a multi-gene phylogenetic analysis, resulting in the elevation of 24 internal clades within the *Alternaria* complex to sections and the synonymization of 13 generic names, including both asexual and sexual morphs, under the *Alternaria*. At present, 29 sections, each typified by a type species, and seven monotypic lineages have been identified within the genus *Alternaria* (Grum-Grzhimaylo et al. 2015, Lawrence et al. 2016, Al Ghafri et al. 2019, Gannibal et al. 2022). *Alternaria* section *Nimbya* is characterized by simple, short to moderately long conidiophores with sympodial proliferations, solitary or short chains of narrowly elongate obclavate conidia with disto- and eu-septa, broad octagonal to rounded cell lumina, broadly octagonal to rounded and short to long tapered beaks (Woudenberg et al. 2013, Gannibal 2018, Ahmadpour 2019, Ahmadpour et al. 2021). Species in the section *Nimbya* are identified as saprophytes or plant pathogens, causing leaf lesions on plants in the families of *Cyperaceae* and *Juncaceae*

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(Simmons 1989, Johnson et al. 2002, Zhao & Zhang 2005, Woudenberg et al. 2013, Gannibal 2018, Ahmadpour 2019, Ahmadpour et al. 2021). So far, five species have been reported in this section in Iran (Ahmadpour 2019, Ahmadpour et al. 2021), but, there is no report on their sexual morphs yet. In this study, the sexual morph of *Alternaria scirpivora* was produced in laboratory conditions. Therefore, we have illustrated and described both the sexual and asexual morphs from this species.

MATERIALS AND METHODS

Fungal isolates and Morphological observations

Fungal isolates were obtained from the culms of *Scirpus acutus* (*Cyperaceae*, *Poales*) showing grey to brown lesions in Miyandoab, Urmia, and Shahindezh Counties in West Azarbaijan province, Iran (Ahmadpour et al. 2021). The isolates were purified by single spore or hyphal tip methods on 2% water agar (2% WA) and potato dextrose agar (PDA; 39 g/L, Merck, Darmstadt, Germany) culture media. Morphological observations were made from fungal strains grown on potato carrot agar (PCA) (20 g white potato, 20 g carrot, 20 g agar, 1 liter of distilled water) medium, incubated at 23–25 ºC under cool white fluorescent light with an 8/16 h light/dark cycle for 5–7 days (Simmons 1989). Ascomata formation was induced by inoculating PCA medium with a 6 cm segment of autoclave-sterilized host plant culms, and incubated at 23–25 ºC for 60–90 days (Johnson et al. 2002). Characteristics of the colonies such as color, pattern, and diameter were observed and recorded, and colony color was assessed using Rayner's (1970) color charts. Fungal structures were measured (20–50 measurements per structure) and photographed using an Olympus AX70 microscope from slide mounts prepared with a clear lactic acid solution. The identified isolates were stored as pure cultures in the fungal culture collections of the Iranian Research Institute of Plant Protection (IRAN) and Urmia University (FCCUU) (Table 1).

DNA extraction, PCR amplification, sequencing and Phylogenetic analyses

Total DNA extraction, amplification of the internal transcribed spacer region of rDNA (ITS–rDNA), parts of the glyceraldehyde-3-phosphate dehydrogenase (*GAPDH*), RNA polymerase second largest subunit (*RPB2*), translation elongation factor 1-alpha (*TEF1*), and *Alternaria* major allergen (*Alt a 1*) genes, as well as polymerase chain reaction (PCR) mixtures, and thermal cycling conditions were done according to the method described by Ahmadpour et al. (2021). Multi-locus phylogenetic analyses were conducted on the combined dataset of the five genes/regions (ITS–rDNA + *GAPDH* + *RPB2* + *TEF1* + *Alt a 1*). DNA sequences from the type or representative strains of the *Alternaria* sections *Crivellia*, *Embellisia*, *Nimbya*, and *Phragmosporae* were retrieved from the GenBank database (Table 1) and used in the analyses. The sequences were aligned using the online alignment tool, MAFFT version 7 (https://mafft.cbrc.jp/alignment/server/) (Katoh et al. 2019). The best-fit substitution models were determined with the Akaike Information Criterion (AIC) in MrModeltest 2.3 (Nylander 2004). The maximum likelihood (ML), maximum parsimony (MP), and Bayesian (BI) analyses were performed using the CIPRES Science Gateway portal (accessible at https://www.phylo.org/) (Miller et al. 2012). The analyses utilized the following tools: RAxML-HPC BlackBox v. 8.2.12, which employed the GTR + GAMMA model with 1,000 bootstrap iterations (Stamatakis 2014); PAUP on ACCESS v. 4.a168, which applied a heuristic search with 1000 bootstrap replicates and branch swapping using the tree-bisection-reconnection (TBR) algorithm (Swofford 2002); and MrBayes 3.2.2 on XSEDE, which implemented the Markov Chain Monte Carlo (MCMC) method with four chains, 1 million generations, a sampling frequency of 1,000, and a 25% burn-in phase (Ronquist et al. 2012). *Stemphylium botryosum* (CBS 714.68) and *S. vesicarium* (CBS 191.86) were used as the outgroup taxa (Woudenberg et al. 2013, Ahmadpour et al. 2021). The resultant phylogenetic trees were visualized in FigTree v. 1.4.4 (Rambaut 2019) and edited in graphic design software, Adobe Illustrator® CC 2021.

RESULTS

Molecular phylogenetic analyses

The combined of DNA sequence data from the five gene regions resulted in a dataset with 2578 characters (nucleotide + gaps) (556 for ITS–rDNA, 547 for *GAPDH*, 803 for *RPB2*, 221 for *TEF1*, and 451 for *Alt a 1*) in which 1692 sites were constant, 886 variable, 115 parsimony-uninformative, and 771 parsimony-informative. The nucleotide substitution models GTR+I+G, HKY+G, SYM+I+G, K80+I, and HKY+G were identified by MrModeltest 2.3 for ITS–rDNA, *GAPDH*, *RPB2*, *TEF1*, and *Alt a 1* datasets, respectively. The combined dataset analysis using RAxML generated a best-scoring tree, with the final

Table 1. GenBank and culture collection accession numbers of isolates used in this study. Newly generated sequences are shown in bold.

Species name	Section	Collection no.	Country	Host/substrate	GenBank accession numbers					Reference
					ITS-rDNA	*GAPDH*	*TEF-1*	*RPB2*	*Alt a 1*	
A. caricicola	*Nimbya*	IRAN 3418C^{T}	Iran	*Carex* sp.	MK508871	MK505392	MT187265	MT187279	MT187233	Ahmadpour, 2019, Ahmadpour et al. 2021
A. caricicola	*Nimbya*	FCCUU 1371	Iran	*Carex* sp.	MK508872	MK505393	MT187266	MT187280	MT187234	Ahmadpour, 2019, Ahmadpour et al. 2021
A. caricicola	*Nimbya*	FCCUU 1373	Iran	*Carex* sp.	MW192223	MW206386	MW206385	MW206384	MW206383	Ahmadpour et al. 2021
A. caricis	*Nimbya*	CBS 480.90^{T}	USA	*Carex hoodii*	AY278839	AY278826	KC584726	KC584467	AY563321	Woudenberg et al. 2013
A.chlamydosporigena	*Embellisia*	CBS 341.71	USA	Air	KC584231	KC584156	KC584710	KC584451	-	Woudenberg et al. 2013
A.chlamydospora	*Phragmosporae*	CBS 491.72^{T}	Egypt	Soil	KC584189	KC584108	KC584647	KC584388	-	Woudenberg et al. 2013
A. cypericola	*Nimbya*	IRAN 3423C^{T}	Iran	*Cyperus* sp.	MT176120	MT187250	MT187262	MT187276	MT187235	Ahmadpour et al. 2021
A. cypericola	*Nimbya*	IRAN 3424C	Iran	*Cyperus* sp.	MT176122	MT187251	MT187263	MT187277	MT187236	Ahmadpour et al. 2021
A. cypericola	*Nimbya*	IRAN 3425C	Iran	*Cyperus* sp.	MT176121	MT187252	MT187264	MT187278	MT187237	Ahmadpour et al. 2021
A. didymospora	*Phragmosporae*	CBS 766.79	Adriatic Sea	Seawater	FJ357312	FJ357300	KC584714	KC584455	-	Woudenberg et al. 2013
A. embellisia	*Embellisia*	CBS 339.71	USA	*Allium sativum*	KC584230	KC584155	KC584708	KC584449	-	Woudenberg et al. 2013
A. heyranica	*Nimbya*	IRAN 3516C^{T}	Iran	*Carex* sp.	MT176114	MT187244	MT187256	MT187270	MT187238	Ahmadpour et al. 2021
A. heyranica	*Nimbya*	FCCUU 1382	Iran	*Carex* sp.	MT176115	MT187245	MT187257	MT187271	MT187239	Ahmadpour et al. 2021
A. junci-acuti	*Nimbya*	IRAN 3508C	Iran	*Juncus acutus*	MT176111	MT187241	MT187253	MT187267	MT187230	Ahmadpour et al. 2021
A. junci-acuti	*Nimbya*	IRAN 3518C	Iran	*Juncus acutus*	MT176112	MT187242	MT187254	MT187268	MT187232	Ahmadpour et al. 2021
A. junci-acuti	*Nimbya*	IRAN 3512C^{T}	Iran	*Juncus acutus*	MT176113	MT187243	MT187255	MT187269	MT187231	Ahmadpour et al. 2021
A. limaciformis	*Phragmosporae*	CBS 481.81^{T}	UK	Soil	KC584203	KC584123	KC584665	KC584407	JQ646368	Lawrence et al. 2013, Woudenberg et al. 2013
A. papavericola	*Crivellia*	CBS 116606^{T}	USA	*Papaver somniferum*	FJ357310	FJ357298	KC584705	KC584446	JN383501	Lawrence et al. 2012, Woudenberg et al. 2013
A. penicillata	*Crivellia*	CBS 116608^{T}	Austria	*Papaver rhoeas*	FJ357311	FJ357299	KC584698	KC584440	JN383502	Lawrence et al. 2012, Woudenberg et al. 2013
A. penicillata	*Crivellia*	CBS 116607	Austria	*Papaver rhoeas*	KC584229	KC584153	KC584706	KC584447	-	Woudenberg et al. 2013
A. phragmospora	*Phragmosporae*	CBS 274.70^{T}	Netherlands	Soil	JN383493	JN383474	KC584721	KC584462	JN383509	Lawrence et al. 2012, Woudenberg et al. 2013
A. scirpicola	*Nimbya*	CBS 481.90	UK	*Scirpus* sp.	KC584237	KC584163	KC584728	KC584469	-	Woudenberg et al. 2013
A. scirpinfestans	*Nimbya*	EGS 49-185	USA	*Scirpus acutus*	JN383499	JN383480	-	-	JN383514	Lawrence et al. 2012
A. scirpivora	*Nimbya*	EGS 50-021	USA	*Scirpus acutus*	JN383500	JN383481	-	-	JN383515	Lawrence et al. 2012
A. scirpivora	***Nimbya***	**IRAN 3513C**	**Iran**	***Scirpus acutus***	**MT176116**	**MT187247**	**MT187259**	**MT187273**	**-**	Ahmadpour et al. 2021
A. scirpivora	***Nimbya***	**IRAN 3514C**	**Iran**	***Scirpus acutus***	**MT176117**	**MT187246**	**MT187258**	**MT187272**	**-**	Ahmadpour et al.2021
A. scirpivora	***Nimbya***	**IRAN 3421C**	**Iran**	***Scirpus acutus***	**MT176118**	**MT187248**	**MT187260**	**MT187274**	**MT187240**	Ahmadpour et al.2021
A. scirpivora	***Nimbya***	**IRAN 3419C**	**Iran**	***Scirpus acutus***	**MT176119**	**MT187249**	**MT187261**	**MT187275**	**-**	Ahmadpour et al. 2021
A. tellustris	*Embellisia*	CBS 538.83^{T}	USA	Soil	FJ357316	AY562419	KC584724	KC584465	AY563325	Lawrence et al. 2012, Woudenberg et al.2013
Stemphylium botryosum	-	CBS 714.68^{T}	Canada	*Medicago sativa*	KC584238	AF443881	KC584729	AF107804	-	Woudenberg et al. 2013
S. vesicarium	-	CBS 191.86^{T}	India	*Medicago sativa*	KC584239	AF443884	KC584731	KC584471	-	Woudenberg et al. 2013

ML optimization likelihood value of –12224.081762. The phylogenetic trees resulting from the maximum likelihood, maximum parsimony, and Bayesian analyses displayed similar topologies and the phylogenetic tree from ML analysis was used to represent the phylogeny of the new isolates (Fig. 1). The phylogram revealed that the four studied isolates clustered well in *Alternaria* section *Nimbya* with the representative strain of *Alternaria scirpivora* (EGS 50-021) with high statistical support values (ML/MP/BI= 100/100/1.0). (Fig. 1). Therefore, our isolates were identified as *A. scirpivora* and confirmed morphological identification.

Taxonomy and morphology

Alternaria scirpivora (E.G. Simmons & D.A. Johnson) Woudenb. & Crous, Stud Mycol 75:198. 2013. Fig. 2 and Fig. 3

Synonymy:

≡*Nimbya scirpivora* E.G. Simmons & D.A. Johnson, in Johnson, Simmons, Miller & Stewart, Mycotaxon 84: 424 (2002).

≡*Macrospora scirpivora* E.G. Simmons & D.A. Johnson, in Johnson, Simmons, Miller & Stewart, Mycotaxon 84: 422 (2002).

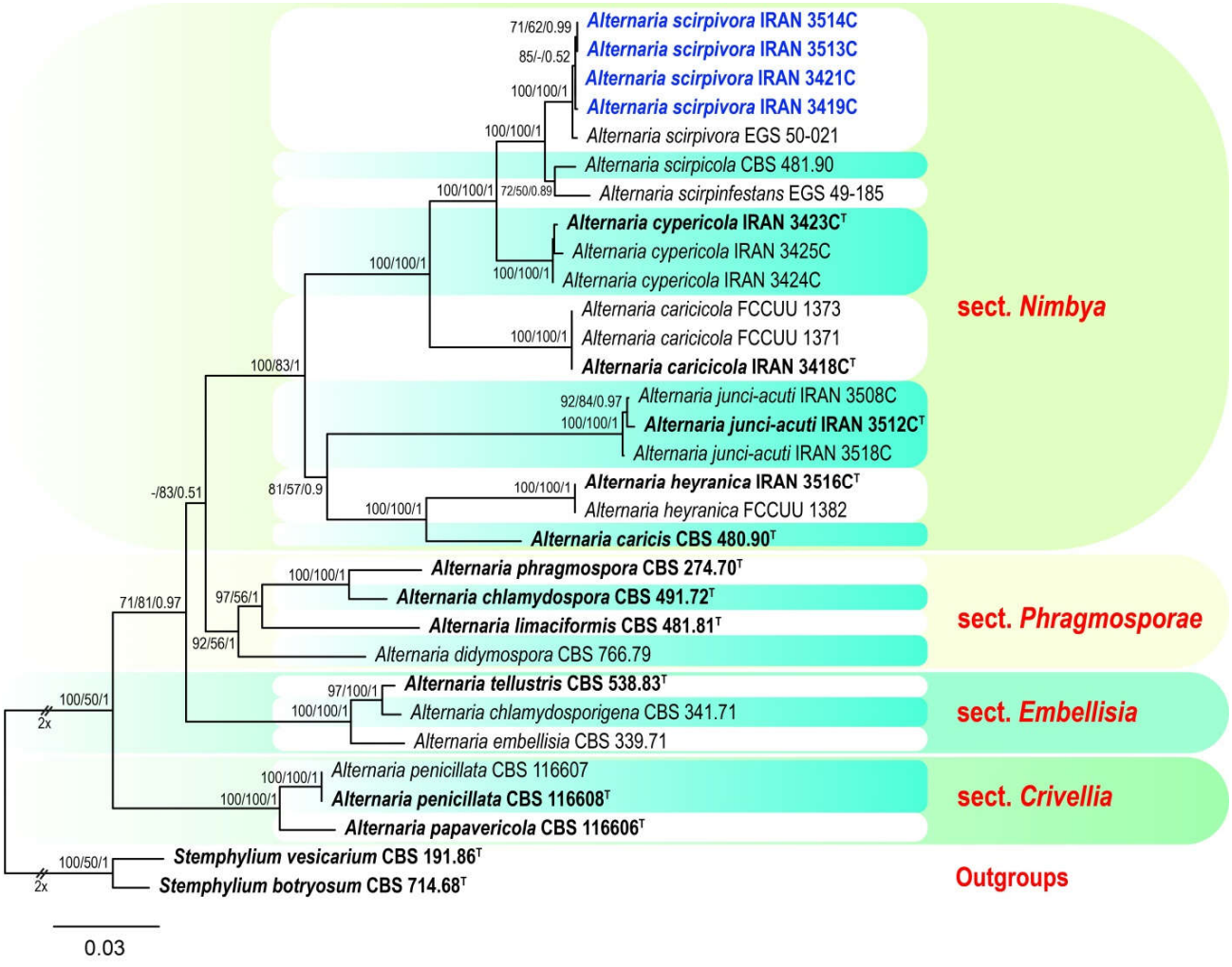

Fig. 1 Phylogenetic tree inferred from Maximum Likelihood (ML) of the combined dataset of ITS, *GAPDH*, *RPB2*, *TEF1*, and *Alt a 1* of *Alternaria* species. The maximum likelihood and maximum parsimony (MP) bootstrap support (MLBS/MPBS) values ⩾50% and Bayesian posterior probabilities (BIPP) ⩾0.50 are given at the nodes. The tree was rooted to *Stemphylium botryosum* strain CBS 714.68 and S. *vesicarium* strain CBS 191.86, and newly identified strains are in blue boldface. The scale bar indicates the number of nucleotide substitutions. ^{T} indicates ex-type strains.

Description – Sexual morph: Ascomata formed on sterilized *Scirpus acutus* stem sections placed on PCA medium, are solitary, scattered to occasionally aggregate, semi-immersed to immersed, becoming erumpent at maturity, globose to subglobose, dark brown to black, ostiolate, 280–500 × 250–450 µm ($\bar{x}$ = 350 × 300 µm, n = 20). Peridium 23–50 µm wide, composed of outer and inner layers, the outer layer consists of heavily pigmented thick-walled cells of textura angularis, and the inner layer consists of light brown to hyaline thick-walled cells of textura angularis. Subhymenium 2–3 µm wide, cellular, septate, branched, dense pseudoparaphyses. Asci are broadly ellipsoid to clavate (when immature), equilateral or musiform, bitunicate, fissitunicate, with short pedicel, commonly 8-spored, 105–125 × 37–57 µm ($\bar{x}$ = 116 × 48 µm, n = 20). Ascospores bi-seriate, partially overlapping, mostly ellipsoid to fusiform, with rounded ends, muriform, with 3–5 transverse septa, 2–4 longitudinal septa, and 0–1 oblique septa, strongly constricted at the median septum, and moderate in other septa, subhyaline to pale brown, 40–46 × 15–21 µm ($\bar{x}$ = 42 × 17 µm, n = 50). Asexual morph on PCA medium: Hyphae branched, septate, light brown, smooth, 2–4 µm wide. Conidiophores macronematous, solitary, straight or slightly curved, simple, septate, light brown to brown, with a single conidiogenous locus or 1–3 geniculate with 1–3 conidiogenous loci, 20–50 × 5–6 µm. Conidia mostly in chains of 2–10 conidia, rarely solitary, straight or slightly curved, obclavate to narrow ovoid or narrow ellipsoid, conidial bodies 30–60 × 5–8 µm ($\bar{x}$ = 45 × 7 µm, n = 50), light brown to brown, surface smooth, 3–10 (mostly 6–8) transverse distosepta, 1–2 transverse eusepta and without longitudinal or oblique septa. Intercalary conidia, typically with 1 to 3 cells, often form during chain development. These conidia produce a secondary spore without forming a distinct secondary conidiophore, except for an apical pore. The cell lumina are distinctly delimited and rectangular, rounded, hexagonal, or encompass the entire volume of the cell. Beaks filiform, septate (2–5 septa), hyaline to light brown, 10–50 × 2–4 µm, occasionally swollen at the tip. Chlamydospores were not observed.

Culture characteristics – Colonies on PCA reaching 60–70 mm diam. after 7 days at 25 °C, flat, entire, dark green to olivaceous brown with sparse, felty, some white to grey aerial mycelium. Sporulation is abundant on PCA from the erect conidiophores that arise directly from the surface or the aerial hyphae. Ascomata formed on PCA containing culms of host plant after 2–3 months at 25 ºC.

Specimen examined – IRAN, West Azarbaijan province, Miyandoab, on infected culms of *Scirpus acutus* (*Cyperaceae*), 20 Sept. 2017, A. Ahmadpour, isolate IRAN 3421C; Shahindezh, on infected culms of *S. acutus*, 20 Sep. 2017, A. Ahmadpour, isolate IRAN 3419C; Urmia, on infected culms of *S. acutus*, 10 Jul. 2019, A. Poursafar, isolates IRAN 3513C and IRAN 3514C (Table 1).

Habitat and distribution – From the culms of *Scirpus acutus* (*Cyperaceae*), USA and Iran (Johnson et al. 2002, Zhao & Zhang 2005, Ahmadpour et al. 2021, Farr et al. 2024, this study).

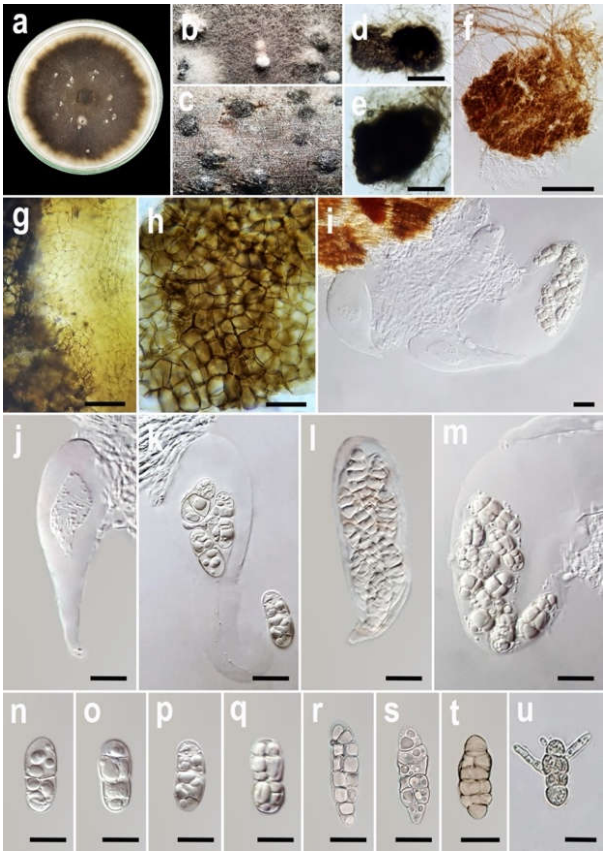

Fig. 2 Sexual morph of *Alternaria scirpivora* (IRAN 3421C). **a–f.** Ascomata on PCA medium containing culms of host plant after two months. **g–h.** Peridium. **i–m.** Asci. **n–t.** Mature and immature ascospores. **u.** Germinated ascospore. Scale bars: **d–f** = 100 µm, **g–u** = 20 µm.

Notes – *Alternaria scirpivora* was originally described from the culm lesions of *S. acutus* from the Pacific Northwest and Minnesota, USA (Johnson et al. 2002). This species is phylogenetically related to *A. scirpicola* and *A. scirpinfestans* (Fig. 1) but can be distinguished by its sporulation pattern, size of conidia, and the number of transverse pseudosepta (Johnson et al. 2002, Ahmadpour et al. 2021). *Alternaria scirpivora* has been identified as a homothallic fungus, producing numerous sexual forms on PCA medium with host culms within 21–23 days (Johnson et al. 2002). According to the phylogenetic tree, the four studied isolates were clustered well with *A. scirpivora* in a distinct subclade, supported by 100% ML/MP bootstrap values and a 1.0 BI posterior probability (Fig. 1). *Alternaria scirpivora* and *A. scirpinfestans* are

pathogens of *Scirpus* spp. (*S. acutus* and *S. validus*) (Johnson et al. 2002, Ahmadpour et al. 2021).

Fig. 3 Asexual morph of *Alternaria scirpivora* (IRAN 3421C). **a–b.** Symptoms on *Scirpus acutus*. **c–e.** Colony on PCA after five days. d. Sporulation pattern on PCA medium. **e–l.** Conidiophores and conidia. **m.** Conidia. Scale bars: **e–m** = 20 µm.

DISCUSSION

In this study, the sexual morph of *Alternaria scirpivora* was produced under laboratory conditions using a PCA culture medium supplemented with autoclave sterilized culms of *Scirpus acutus* after 2–3 months at 23–25 ºC. Immature ascocarps were produced in the PCA culture medium without the presence of host tissues; however, no asci or ascospores were observed. The sexual morph of species in the section *Nimbya* has not been documented in Iran or other Asian countries. Attempts by Ahmadpour et al. (2021) to induce the formation of sexual morphs were unsuccessful. The morphological characteristics of the sexual morph produced in this study are almost similar to those reported by Johnson et al. (2002). In their study, the mature ascomata were formed easily on sterilized stem sections of either *Scirpus acutus* or *S. validus* placed on 2% water agar medium at 15–23 ºC within 21–23 days. Of the three studied isolates, EGS 50-021 produced ascomata more easily and more abundantly compared to the EGS 50-008 and EGS 50-020 isolates. Variation in the timing of ascomata formation in the current study may be attributed to the genetic nature of the isolates, the type of culture medium utilized, and the incubation conditions applied.

Sexual morphs of species in *Nimbya* section were identified as *Macrospora* Fuckel (Simmons 1989, Johnson et al. 2002, Woudenberg et al. 2013). *Macrospora* species are morphologically similar to *Allewia* (the teleomorph of *Embellisia* species), *Lewia* Barr & Simmons (the teleomorph of *Alternaria* species) *Pleospora* Rabenh. Ex Ces. & De Not. (the teleomorph of *Stemphylium* species) and *Pyrenophora* Fr. (the teleomorph of *Drechslera* species) (Simmons 1989, Johnson et al. 2002, Woudenberg et al. 2013). Nevertheless, the genera *Allewia*, *Lewia*, *Macrospora*, and their asexual morphs have been synonymized under the *Alternaria* genus (Woudenberg et al. 2013). So far, sexual morphs have been described in eight sections of *Alternaria viz. Alternaria*, *Crivellia*, *Embellisioides*, *Eureka*, *Infectoriae*, *Nimbya*, *Omanensis* and *Panax* (Kwaśna & Kosiak 2003, Simmons 2007, Al Ghafri et al. 2019, Hashemlou et al. 2020). Of the ten species formally described in the section *Nimbya* (Woudenberg et al. 2013, Gannibal 2018, Ahmadpour 2019, Ahmadpour et al. 2021), the sexual morph has been observed and described in three species: *A. scirpicola*, *A. scirpinfestans*, and *A. scirpivora* (Simmons 1989, Johnson et al. 2002, this study). Sexual development offers several advantages to fungal species. It serves as a key source of genetic variation through recombination during meiosis and crossing over, which enhances the fitness of fungal species and improves their adaptation to changing environments. Additionally, it helps repair genetic damage via recombination, masks lethal mutations and prevents their fixation in the genome. Moreover, the production of highly resistant sexual structures or spores provides an extra benefit in unfavorable environmental conditions (Dyer & O'Gorman 2012, Ellena et al. 2020). In filamentous ascomycetes, sexual reproduction is governed by a single mating type locus (*MAT*), which consists of two distinct DNA sequences, called the *MAT1-1* and *MAT1-2* idiomorphs (Turgeon 1998, Wang et al. 2017). Successful sexual reproduction depends on the expression of genes from both idiomorphs, allowing mating systems to be categorized based on the genetic content of the *MAT* locus. Fungi have two main sex-determination systems: homothallism and heterothallism. Homothallic fungi are self-fertile and can undergo sexual reproduction from a single spore culture, whereas heterothallic fungi are self-sterile and require a compatible partner for reproduction (Wilson et al. 2015, Sun et al. 2019). In this study, the isolates developed mature ascomata with asci and ascospores from single spore cultures, indicating that they are

homothallic, consistent with observations from all *Alternaria* species whose sexual forms have been documented so far. Since species from the section *Nimbya* have predominantly been found on plants in the *Cyperaceae* and *Juncaceae* families, and given the widespread distribution of these plants in different regions of Iran as well as their significant diversity, it is expected that new and diverse species from this section will be discovered and described. Further studies are required to explore the diversity of *Alternaria* species in the section *Nimbya* along with their sexual morphs, in relation to these plants across various geographic locations in Iran.

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شکل جنسی *Alternaria scirpivora* (*Alternaria* section *Nimbya*) از ایران

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چکیده: گونه‌های بخش *Nimbya* از جنس *Alternaria* پوده‌رست یا بیمارگرهای گیاهی عمدتاً مرتبط با گیاهان تیره‌های اویارسلام و سازو می‌باشند. تا کنون، پنج گونه از این بخش در ایران گزارش شده ولی شکل جنسی آنها مشخص نگردیده است. در مطالعه حاضر، شکل جنسی گونه *Alternaria scirpivora* در شرایط آزمایشگاهی روی محیط کشت PCA حاوی ساقه‌های سترون شده گیاه *Scirpus acutus* که به مدت ۹۰ روز در دمای ۲۵-۲۳ درجه سلسیوس نگهداری شده بودند، تحریک شد. آسکوماتای بالغ حاوی آسک‌ها و آسکوسپورها پس از گذشت ۳-۲ ماه تشکیل شدند. ویژگی‌های ریخت‌شناختی شکل‌های جنسی و غیرجنسی توصیف شدند و روابط تبارشناختی *A. scirpivora* با گونه‌های با خویشاوندی نزدیک بحث گردید. این اولین گزارش از شکل جنسی گونه مذکور در ایران و آسیا می‌باشد.

کلمات کلیدی: *Alternaria*، آسکوما، هموتالیسم، ریخت‌شناسی، تبارشناسی مولکولی.

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