A Destructive Leaf Spot and Blight Caused by *Alternaria kareliniae* sp. nov. on a Sand-Stabilizing Plant, Caspian Sea Karelinia

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Abstract

Leaf spots and stem lesions causing widespread mortality of Caspian Sea karelinia (*Karelinia caspia*) were observed in desert regions of Xinjiang Uyghur Autonomous Region, China. Fifteen samples were collected from five widely distributed counties of Tarim and Junggar Basins in 2016. The pathogen was identified using morphological observations and phylogenetic analyses based on combined partial sequences from seven genes (*Alt a 1*, *ATPase*, *calmodulin*, glyceraldehyde 3-phosphate dehydrogenase, *internal transcribed spacer*, RNA polymerase II, and *translation elongation factor 1*), and placed as a new species: *Alternaria kareliniae* sp. nov. in section *Dianthicola*. The fungus has a small conidium (24.3 to 29.1 to 64.8

(to 75.8) by (9.3 to) 12.4 to 16.5 (to 21.7) μm with a long beak (130 to) 183.9 to 350.4 (to 378.2) μm , as well as four to eight transverse septa, which differs significantly from other species of *Alternaria* section *Dianthicola*. On potato carrot agar, it grew significantly more slowly than others of this section. Pathogenicity tests showed that the fungus could infect leaves and stems of *K. caspia* and cause the same symptoms as those observed in the field. The fungus was reisolated from inoculated leaves and stems of the host. The disease in desert regions appears to be increasing, and it may have future negative implications for desert ecology in these areas. Future research should concentrate on elucidating the disease cycle and disease management alternatives.

The genus *Alternaria* Nees was established in 1816. Since then, more than 1,100 names have been published, and nearly 300 taxa have been accepted (Simmons 2007). Most *Alternaria* spp. are saprobes that are commonly found in soil or on decaying plant tissues (Thomma 2003). Some species are distributed worldwide, infecting a broad range of economically important crops, and are well known as destructive pathogens, such as *Alternaria bataticola*, *A. porri*, *A. solani*, and *A. tomatophila* (Abo-Elyousr et al. 2014; Lopes and Bioiteus 1994; Rotem 1994; Tymon et al. 2016). A few species have been intercepted as quarantine subjects in some countries (Sakoda et al. 2010). Accurate identification of plant-pathogenic fungi and the use of accepted names are essential for the development of effective biosecurity and trade policies as well as a prerequisite for risk assessment (Cai et al. 2011; Hyde et al. 2010). *Alternaria* spp. have been classified predominantly based upon their morphological characteristics, which include conidial morphology, catenulate spore patterns, and characteristics in pure culture. Recent molecular approaches based on multiple gene (e.g., *Alt a 1*, glyceraldehyde 3-phosphate dehydrogenase [*GPDH*], *internal transcribed spacer* [*ITS*], large subunit [*LSU*], RNA polymerase II [*RPB2*], *translation elongation factor 1* [*TEF1*], and β -tubulin [*TUB*]) analyses have been widely used to determine the taxonomic status of species of *Alternaria* (Hong et al. 2005; Lawrence et al. 2012, 2013; Woudenberg et al. 2013). Furthermore, the use of plasma membrane ATPase and calmodulin, and comparisons of type or representative isolates, are recommended for accurate species identification within the genus *Alternaria* (Lawrence et al. 2013, 2016).

Caspian Sea karelinia (*Karelinia caspia* (Pall.) Less. (Asteraceae)) is an ecologically important plant species occurring across the desert ecosystems of Russia, Central Asia, and northwest China (Lin 1999). It exhibits both drought and salt tolerance, which allows it to play a role in reducing wind erosion and desertification (Vonlanthen et al. 2011). Previous studies with this plant have mainly concentrated

on ecology and stress tolerance (Wang et al. 2015; Zhang et al. 2014); however, diseases of this plant are poorly understood.

In late July 2014, a new and serious leaf spot disease was observed in the desert margin zone in northwest Tarim and southern Junggar Basins, Xinjiang Uyghur Autonomous Region of western China. In 2016, due to a significant increase in rainfall compared with previous years, the disease was more severe and caused an epidemic in some areas. Microscopic observations of infected Caspian Sea karelinia plants showed pathogen characteristics typical of the genus *Alternaria*. The objectives of this study were to (i) identify and characterize species of *Alternaria* associated with leaf spot and blight on Caspian Sea karelinia using molecular phylogenetic and morphological approaches, (ii) provide a detailed description of the leaf spot and blight on Caspian Sea karelinia caused by this pathogen, and (iii) assess the pathogenicity of these *Alternaria* isolates to Caspian Sea karelinia.

Materials and Methods

Disease investigation and sample collection. In September 2016, diseased Caspian Sea karelinia plants were surveyed and collected in five severely affected counties (Aksu, Aral, Karamay, Wujiaqu, and Xayar) in Tarim and Junggar Basins of Xinjiang Uyghur Autonomous Region (Fig. 1). These collection sites were distributed in an area covering over 1,000 km², in areas with typically low annual rainfall (<200 mm/year). Each of the five severely affected plot areas was 10 by 10 m, and all asymptomatic, symptomatic, and dead plants were surveyed. Plants were considered infected if they had a leaf spot or blight. With three samples per plot, 15 recently diseased leaves and stems showing typical leaf spot and blight were collected. Samples were transported to the laboratory and processed immediately or stored at 4°C and processed within 24 h of collection.

Fungal isolations. Diseased leaves and stem tissues from necrotic margins were cut into 3- to 5-mm-long pieces and surface sterilized with 75% ethanol for 3 to 5 s and 3% sodium hypochlorite (NaClO) for 2 min. The samples were washed three times with sterile distilled water and blotted dry with sterilized filter paper. The sections were placed onto potato dextrose agar (PDA) supplemented with ampicillin at 100 $\mu\text{g}/\text{ml}$ and incubated at 25°C in darkness for 10 days. Isolates were purified by single-spore culturing and subcultured onto malt extract agar (MEA), oatmeal agar (OA), potato carrot agar (PCA), and V8 juice agar (V8) and incubated at 25°C with a photoperiod of 12 h of light and 12 h of darkness to characterize fungal growth and morphology.

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Morphological studies. Morphological examinations were performed according to descriptions by Simmons (2007). Cultural and conidial morphology were examined after 10 days. Fifty mature conidia and conidiophores mounted in sterile water were randomly selected for size measurements under a compound light microscope (Olympus BX50). The specimens were deposited in the Mycological Herbarium of Tarim University, Aral, China and the Mycological Herbarium of South China Agricultural University, Guangzhou, China, and representative cultures are being maintained in the Culture Collection of Tarim University, Aral, China (CTU).

DNA extraction and polymerase chain reaction amplification. Mycelia were collected from PDA cultures 10 days after incubation and pulverized by the MP Fastprep-24 sample preparation system (MP Biomedicals). Genomic DNA was extracted using the E.Z.N.A.TM Fungal DNA kit (OMEGA Biotek), according to the manufacturer's instructions. The complete ITS region was amplified with the primers V9G (de Hoog and Gerrits van den Ende 1998) and ITS4 (White et al. 1990), the partial *Alt a 1* region with *Alt-for* and *Alt-rev* (Hong et al. 2005), the partial *TEF1* region with EF1-728F and EF1-986R (Carbone and Kohn 1999), the partial *RPB2* region with RPB2-5F2

Fig. 1. Map of the Xinjiang Uyghur Autonomous Region showing the areas where *Alternaria kareliniae* was sampled in this study.

Table 1. Isolates used in this study and their GenBank accession numbers for seven genes^a

Sections, species names	Strains ^b	GenBank accession numbers						
		<i>ITS</i>	<i>GPDH</i>	<i>Alt a 1</i>	<i>RPB2</i>	<i>Calmodulin</i>	<i>TEF1</i>	<i>ATPase</i>
Sect. <i>Cheiranthus</i>								
<i>Alternaria cheiranthi</i>	EGS 41-188; CBS 109384	AF229457	AY278802	AY563290	KC584387	JQ646164	KC584646	JQ671830
<i>A. indefessa</i>	CBS 536.83T; EGS 30-195	KC584234	KC584159	AY563323	KC584458	JQ646165	KC584717	JQ671831
Sect. <i>Dianthicola</i>								
<i>A. dianthicola</i>	CBS 915.96; CBS 116491	JQ693640	KC584113	–	KC584394	JQ646144	KC584653	JQ671810
<i>A. elegans</i>	CBS 109159T; EGS 45.072	NR_136040	KC584114	GQ180092	KC584395	–	KC584654	–
<i>A. kareliniae</i>	CTU C031T	KY945066	KY945063	KY945051	KY945072	KY945057	KY945060	KY945054
	CTU C033	KY945067	KY945064	KY945052	KY945073	KY945058	KY945061	KY945055
	CTU C036	KY945068	KY945065	KY945053	KY945074	KY945059	KY945062	KY945056
<i>A. simsimi</i>	CBS 115265T	NR_136013	KC584137	–	KC584428	–	KC584686	–
Sect. <i>Pseudoulocladium</i>								
<i>A. aspera</i>	CBS 115269T	KC584242	KC584166	KF533899	KC584474	–	KC584734	–
<i>A. chartarum</i>	ATCC 18044; CBS 200.67T	NR_130656	AY278819	AY563319	KC584481	JQ646162	KC584741	JQ671828
<i>A. concatenata</i>	CBS 120006; HSAUPXF 030035	KC584246	AY762950	EU684149	KC584480	–	KC584740	–
<i>A. septospora</i>	CBS 109.38	FJ266489	FJ266500	FJ266515	KC584487	JQ646163	KC584747	JQ671829
Sect. <i>Ulocladioides</i>								
<i>A. atra</i>	ATCC 18040; CBS 195.67T	AF229486	KC584167	–	KC584475	JQ646167	KC584735	JQ671833
<i>A. brassicae-pekinensis</i>	CBS 121493T	KC584244	KC584170	KF533892	KC584478	–	KC584738	–
<i>A. cantlous</i>	CBS 123007T; HSAUP 0572	KC584245	KC584171	FJ041319	KC584479	–	KC584739	–
<i>A. consortialis</i>	CBS 104.31T; CBS 201-67	KC584247	KC584173	FJ266509	KC584482	JQ646173	KC584742	JQ671839
<i>A. cucurbitae</i>	CBS 483.31; EGS 31-021	FJ266483	AY562418	FJ008712	KC584483	JQ646170	KC584743	JQ671836
<i>A. multifforme</i>	CBS 102060T	FJ266486	KC584174	FJ266512	KC584484	JQ646171	KC584744	JQ671837
<i>A. subcucurbitae</i>	CBS 121491T	KC584249	EU855803	EU855807	KC584489	–	KC584749	–
Outgroup								
<i>Stemphylium vesicarium</i>	ATCC 1852; CNU 094002	AF229484	AY278821	AY563275	–	JQ646102	JF331624	JQ671768

^a ITS = internal transcribed spacer, GPDH = glyceraldehyde 3-phosphate dehydrogenase, RPB2 = RNA polymerase II, TEF1 = translation elongation factor 1. Sequences obtained in this study are indicated in bold; – indicates sequence was not available.

^b ATCC = American Type Culture Collection, Manassas, VA; CBS = culture collection of the Centraalbureau voor Schimmelcultures, Fungal Biodiversity Centre, Utrecht, The Netherlands; EGS = personal collection of Dr. E. G. Simmons; CTU = culture collection of Tarim University, Aral, China; HSAUP = Department of Plant Pathology, Shandong Agricultural University, Taian, China; CNU = culture collection of Chungnam National University, Daejeon, Korea; and T = ex-type strain.

(Sung et al. 2007) and fRPB2-7cR (Liu et al. 1999), the partial calmodulin region with CALDF1 and CALDR1 (Lawrence et al. 2013), the partial ATPase region with ATPDF1 and ATPDR1 (Lawrence et al. 2013), and the partial GPDH region with gpd1 and gpd2 (Berbee et al. 1999). The polymerase chain reactions (PCR) were performed in a MyCycler Thermal Cycler (Bio-Rad) in a total volume of 25 μ l containing 2.5 μ l of 10 $\times$ EasyTaq Buffer (Takara Biotech), 50 μ M dNTP, 0.1 μ M each primer, 0.75 U of Taq DNA polymerase, and 1 to 10 ng of genomic DNA. Conditions for PCR amplification consisted of an initial denaturation step of 4 min at 94°C followed by

35 cycles of 45 s at 94°C, 45 s at 56°C, and 60 s at 72°C for ITS; 30 cycles of 60 s at 94°C, 60 s at 56°C, and 45 s at 72°C for GAPDH and Alt a 1; and 30 cycles of 30 s at 94°C, 30 s at 59°C, and 60 s at 72°C for ATPase, calmodulin, and TEF1. The partial *RPB2* gene was obtained by using a touchdown PCR protocol of 5 cycles of 45 s at 94°C, 45 s at 60°C, and 2 min at 72°C; followed by 5 cycles with a 58°C annealing temperature and 30 cycles with a 54°C annealing temperature, as described by Woudenberg et al. (2013); and a final elongation step of 10 min at 72°C. The PCR products were sequenced bidirectionally at the Beijing Genomics Institute BGI, using the same primers

Fig. 2. Symptoms of infection by *Alternaria kareliniae* on *Karelinia caspia* in the field. **A.** Initial symptoms on leaves. **B.** Necrotic spots extending from leaves to stems. **C.** Whole plants withered.

Fig. 3. Maximum-likelihood tree and Bayesian analyses inferred from analysis of combined ITS, GPDH, Alt a 1, RPB2, calmodulin, TEF1, and ATPase sequences of *Alternaria* isolates associated with genus *Karelinia*. Bootstrap values were calculated from 1,000 replications. RaxML bootstrap support values >70 (before the slash marks) and Bayesian posterior probabilities >0.95 (after the slash marks) are shown. The scale bar indicates the number of nucleotide substitutions.

as those used in our PCR procedures. For each gene, consensus sequences were derived from both primer directions using MAFFT.

Phylogenetic analyses. Sequences of the isolates were aligned with published sequences of *Alternaria* spp. retrieved from GenBank (Table 1), using the online version of MAFFT, ver. 7.215 (Kato and Standley 2013). *Stemphylium vesicarium* (Wallr.) E. G. Simmons (culture number ATCC 18521) was used as the outgroup taxon. When a gene sequence was not available for a particular isolate,

the gene for that isolate was replaced by a consensus sequence for that gene constructed from the multiple alignment following Nei and Kumar (2000) and using SequenceMatrix, version 1.7.8 (Vaidya et al. 2011). The concatenated alignment was deposited in TreeBASE (<https://www.treebase.org/>) under accession S20906. Maximum-likelihood (ML) analyses were performed with RAxML 8.0.26 (Stamatakis 2014) by using RAxML GUI v. 1.31 (Silvestro and Michalak 2012) with a rapid bootstrap algorithm, the GTR GAMMAI model,

Fig. 4. Necrotic spots resulting from inoculation of *Alternaria kareliniae* onto *Karelina caspia* leaves, and wound responses on the negative controls. **A**, Negative control inoculation sites showing wound and the absence of necrotic spot development and **B**, inoculation sites showing necrotic spots from isolate CTU C031T; **C**, isolate CTU C033; and **D**, isolate CTU C036.

Fig. 5. Necrotic spots resulting from inoculation of *Alternaria kareliniae* onto *Karelina caspia* stems, and wound responses on the negative controls. **A**, Negative control inoculation sites showing wound and the absence of necrotic spot development and **B**, inoculation sites showing necrotic spot from isolate CTU C031T; **C**, isolate CTU C033; and **D**, isolate CTU C036.

and 1,000 bootstrap replicates. Bayesian interference (BI) analyses were performed as described by Woudenberg et al. (2013) using MrBayes (Ronquist et al. 2012). The phylogenetic trees were checked and edited using FigTREE v1.4.0 (Rambaut 2007).

Pathogenicity tests. Two representative isolates (CTU C031T and CTU C033) from leaf spots and one (CTU C036) from stem lesions of Caspian Sea karelinia were used to inoculate wounded detached leaves and stems of healthy plants to test for pathogenicity, following Pryor and Michailides (2002). Spore suspensions of *Alternaria*

isolates were prepared by flooding PDA cultures with sterile water and adjusting the collected suspension to 10^5 spores/ml using a hemacytometer. Tests were conducted on detached leaves and stems of Caspian Sea karelinia placed in Petri dishes on top of moistened autoclaved filter paper. Each inoculation site of leaf or stem was slightly wounded on the surface with a sterile dissecting needle prior to inoculation. A conidial suspension ($20\ \mu\text{l}$ of 10^5 spores/ml) was placed on each wound site, with three replicates for each treatment. Control leaves and stems were mock inoculated with sterile water. Each test was incubated at $25 \pm 1^\circ\text{C}$ with 80 to 95% relative humidity under a photoperiod of 12 h of light and 12 h of darkness in a controlled-environment chamber.

Results

Disease investigation and fungal isolation. Symptoms of leaf spot and blight on *K. caspia* were found in many desert regions of the Tarim and Junggar Basins. Early symptoms appeared on the mature leaves in late July, in the form of numerous circular, small, dark-brown spots, with a dark halo on the leaf surface surrounded by chlorosis. The infection subsequently progressed to the younger leaves and caused severe necrotic lesions leading to defoliation (Fig. 2A and B). As the disease progressed, lesions occurred on the stems, which turned yellow. Heavy infection resulted in death of the plants (Fig. 2C). From the five regions examined, a 10-by-10-m plot was established in each region representing the highest levels of disease. Disease incidence in these plots ranged from 50 to 90% while mortality ranged from 40 to 70%. Fifteen samples collected from the above plots were subjected to isolation, and all strains showed consistent morphology.

Phylogenetic analyses. Phylogenetic analyses were carried out on three representative strains (CTU C031T, CTU C033, and CTU C036) based on the sequences from seven genes: *Alt a 1*, *ATPase*, *calmodulin*, *GPDH*, *ITS*, *RPB2* and *TEF1* (GenBank accession numbers KY945051 to KY945077). PCR amplification and sequencing of each gene yielded product sizes for these three isolates of 473, 1,195, 722, 580, 491, 865, and 233 bp, respectively. The sequences were then concatenated for phylogenetic analyses. The three sequenced *Alternaria* isolates were compared with 17 other taxa (Table 1).

ML and BI analyses yielded the same tree topology, and a single tree with bootstrap support from ML and BPP (Bayesian posterior probabilities) from BI was generated (Fig. 3). The phylogenetic analysis showed that all isolates from Caspian Sea karelinia clustered together with statistical support of 100% BP (bootstrap proportions) and 1.00 BPP, and formed a strongly defined distinct lineage within *Alternaria* section (sect.) *Dianthicola* separated from *A. simsimi*, *A. elegans*, and *A. dianthicola*.

Pathogenicity tests. The three fungal isolates of *A. kareliniae* inoculated onto detached leaves and stems all gave symptoms of black leaf spot and stem necrosis after 3 to 5 days but no symptoms were observed in mock-inoculated control leaves and stems. Round necrotic spots appeared at the inoculation sites 1 to 2 days after

Fig. 6. Colony characteristics after 10 days of growth on A, potato dextrose agar; B, oatmeal agar; C, malt extract agar; D, potato carrot agar; and E, V8 juice agar.

Fig. 7. Microscopic characteristics of *Alternaria kareliniae*. A, Chlamydospores; B, C, D, and E, conidia with long beaks; and F, conidiophores. Bar = $20\ \mu\text{m}$.

inoculation, and spread along the entire leaves and stems within 5 days. The symptoms were similar to those observed in the field (Figs. 4 and 5). The fungus was easily reisolated from lesions of inoculated leaves and stems, and was identical to the original isolate based on colony morphology and molecular sequence.

Description of the pathogen. Based on the phylogenetic analyses of the combined seven-gene dataset, as well as morphological characteristics, the pathogen collected from *K. caspia* clearly represented a new species, which is described as follows:

Alternaria kareliniae B. Xu & Z. D. Jiang sp. nov. (Figs. 6 and 7)
Mycobank: MB 820927

After 10 days at 25°C, colonies on PCA were consistent with PDA, white to pale gray, 34–48 mm diam. with medium olive zones on the reverse side. Colonies on MEA were similar to V8, gray to light olive, slightly orange-brown on the underside, 24–29 mm diam. On OA, colonies were gray, with sparse aerial mycelium, and 34–37 mm diam. (Fig. 6). Conidiophores arising singly, erect or curved, few branched, pale brown, smooth, round and scarred at the apex, (14.7–) 31.3–70.4 (–105.5) long μm , 3.1–5.8 μm wide with 0–5 septa ($n = 50$). Conidia solitary, few in chains, conidia body (24.3–) 29.1–64.8 (–75.8) $\times$ (9.3–) 12.4–16.5 (–21.7) μm , with 4–8 transverse septa and one to two longitudinal or oblique septa, constricted at the middle transverse septa, the beaks (130–) 183.9–350.4 (–378.2) μm long, 2.1–3.3 μm wide with few septa, some cells bulging strongly in the lower part ($n = 50$) (Fig. 7).

Etymology. The specific epithet refers to the genus of the host plant (*Karelinia caspia* (Pall.) Less.)

Materials studied. In Xinjiang, China, Karamay County, 11 August 2016, (ex-type culture number CTU C031T) and Wujiaqu County, 26 August 2016 (culture number CTU C033) the isolates were derived from leaf spot of *K. caspia*; in Aral County, 15 September 2016, the isolates were derived from stem lesions (culture number CTU C036).

Discussion

This study resulted in the discovery of a previously undescribed *Alternaria* sp. on diseased *K. caspia* which is placed in sect.

Dianthicola. Combined morphological characteristics and phylogenetic analyses confirmed this fungus as a new taxon. The new species was described and named *Alternaria kareliniae*. Currently, *Alternaria* sect. *Dianthicola* contains three species, including *A. simsii*, *A. elegans*, and *A. dianthicola* (Lawrence et al. 2016). *A. dianthicola* with a short beak is obviously different from other species in this section (Ellis and Ellis 1985), including the long-beaked *A. kareliniae*. However, there are only slight morphological differences between *A. kareliniae*, *A. simsii*, and *A. elegans*. (Table 2). *A. kareliniae* has a slower growth rate under the same cultural conditions (PCA) than *A. simsii* or *A. elegans*. In addition, *A. kareliniae* differs from *A. simsii* by having a smaller conidial body and more septa than *A. simsii* (conidial body 45 to 90 by 12 to 18 μm , with 7 to 10 transverse septa) (Simmons 2004). *A. kareliniae* sporulates as solitary or rarely chains of two conidia, while *A. simsii* has chains of 2 to 4 conidia and conidiophores up to 75 to 200 μm (Simmons 2000).

In previous work based on multigene sequence data analysis, phylogenetic lineages within *Alternaria* and allied genera were redefined (Lawrence et al. 2013; Woudenberg et al. 2013). The results showed that the *Alternaria* clade contains 27 sections (internal clades) and eight monotypic lineages (Lawrence et al. 2016). The pathogen *A. kareliniae* belongs to sect. *Dianthicola*, which also consists of *A. simsii*, *A. elegans*, and *A. dianthicola*. In our study, phylogenetic results from the combined dataset of the *GPDH*, *RPB2*, and *TEF1* gene sequences produced topologies concordant with Woudenberg et al. (2013). In the combined dataset of the seven gene sequences, *A. simsii*, *A. elegans*, and *A. dianthicola* and our three representative *A. kareliniae* isolates formed well-supported monophyletic entities (76% BP, 0.99 BPP) (Fig. 3), and this topology was also concordant with Woudenberg et al. (2013).

In previous classifications, plant-pathogenic fungal species were often placed in association with a single host plant species or genus. In more recent classifications of *Alternaria* spp., phylogenetic analyses suggest that these specific host–pathogen associations within different plant families have occurred multiple times throughout the evolution of this taxon (Lawrence et al. 2016). Among *Alternaria* sect. *Dianthicola*, *A. dianthicola*, *A. elegans*, and *A. simsii* are

Table 2. Morphological comparison of *Alternaria kareliniae* with *A. dianthicola*, *A. elegans*, and *A. simsii*

Characteristics		<i>A. kareliniae</i>	<i>A. dianthicola</i>	<i>A. elegans</i>	<i>A. simsii</i>
Cultural	Colony				
Growth on PCA ^a	Growth	Sparse	Abundant	Abundant	Abundant
	Growth rate	34–48 mm after 10 days 4 mm/day	35–40 mm after 5 days 7.5 mm/day	Fast ^b	90 mm after 5 days 18 mm/day
Sporulation	Shape	Solitary or rare chains of two conidia	Chains of 4–5 conidia	Chains of 2–4 conidia	Chains of two or more conidia abundant
Conidiophores	Shape	Simple or branched, a few geniculate extensions	Simple or branched, Geniculate extensions	Short branched, Geniculate extensions	Simple or branched, a few Geniculate extensions
	Size (μm)	(14.7–) 31.3–70.4 (–105.5) by 3.1–5.8	5–150 by 4–6	75–200	Up to 80 by 3.0–5.0
Conidial body	Shape	Variable shape, diamond, obclavate, ovoid or narrow ellipsoid	Obclavate to cylindrical	Ovoid or ellipsoid	Narrowly ellipsoid
	Size (μm)	(24.3–) 29.1–64.8 (–75.8) by (–9.3) 12.4–16.5 (–21.7)	55–130 by 10–16	30–69 by 13–16	45–90 by 12–18
	Transverse septa	4–8	Up to 14	7–10	7–10
	Longitudinal septa	1–2	1–2	1–2	1–2
	Color	Brown with eusepta in darker contrast	Pale olivaceous brown	Moderate brown with darker contrast	Moderate brown with eusepta in darker contrast
	Ornamentation	Lack ornamentation	Lack ornamentation	Lack ornamentation	Lack ornamentation
	Beak				
Beak	Shape	Narrowly filamentous with few septa	Filamentous with septa	Narrowly filamentous with septa	Narrowly filamentous with septa
	Size (μm)	(130–) 183.9–350.4 (–378.2) by 2.1–3.3	25.6–166.3 by 4.2–8.2	225–275 by 2	275–325 by 2.5

^a Potato carrot agar.

^b The growth of *A. elegans* was described by Simmons (2000).

associated with the families Caryophyllaceae, Solanaceae, and Pedaliaceae, respectively (Ellis and Ellis 1985; Simmons 2000, 2004). Most *Alternaria* spp. on species of Asteraceae belong to sect. *Porri* (Simmons 2007; Woudenberg et al. 2014). In this study, *A. kareliniae* was associated with *K. caspia*, which is also in the Asteraceae family. This further demonstrates and confirms that *Alternaria* sect. *Dianthicola* has no strong host plant family specialization or association.

The field observations and in vitro pathogenicity studies showed that *A. kareliniae* is an aggressive pathogen of *K. caspia*, capable of killing the plants within a short period following infection. Widespread mortality of Caspian Sea karelinia has been increasing in frequency with increasing rainfall in recent years, and the disease incidence appears to be increasing. Because *K. caspia* is becoming widely grown in the desert regions of the Tarim and Junggar Basins for preventing local soil erosion, the disease poses a serious threat to soil de-desertification and salt-alkalization control strategies. Studies on pathogen variation, disease progression, pathogenicity on plants in the field, and disease management should be done in the future.

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