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Alternaria telliensis sp. nov., a new species isolated from *Solanaceae* in Algeria

NABAHAT BESSADAT[1,2,3], BRUNO HAMON[2,4], NELLY BATAILLE-SIMONEAU[2,5], KIHAL MABROUK[1,6] & PHILIPPE SIMONEAU[2,7]*

[1]*Applied Microbiology Lab, University Oran1 Ahmed Ben Bella, BP1524 El M'naouer 31000 Oran, Algeria*
[2]*UMR 1345 IRHS, University Angers, INRA, Agrocampus-Ouest, SFR QUASAV, 2 Bld Lavoisier, Angers, France*
[3] *bessadatnabahat@hotmail.com; https://orcid.org/0000-0001-7795-2606*
[4] *bruno.hamon@univ-angers.fr; https://orcid.org/0000-0001-5868-0676*
[5] *nelly.bataille-simoneau@univ-angers.fr; https://orcid.org/0000-0001-6200-4259*
[6] *kihalm@gmail.com; https://orcid.org/0000-0003-2901-373X*
[7] *simoneau@univ-angers.fr; http://orcid.org/0000-0002-3890-9848*
**Corresponding author. Phone: +33677123755*

Abstract

During a biodiversity survey of *Alternaria* associated with leaf spot and blight of *Solanaceae*, a large collection of strains was obtained from samples collected in north-western regions of Algeria in 2013–2018 growing seasons. Amongst these strains, three isolates recovered from tomato and potato had morphological traits different from that usually observed for *Alternaria* species previously reported on *Solanaceae*. Based on analysis of a sequence dataset corresponding to portions of the glyceraldehyde-3-phosphate dehydrogenase (*gpd*), translation elongation factor 1-alpha (*tef1*) and RNA polymerase second largest subunit (*rpb2*) genes along with morphological observations, isolates were identified as a new species in the section *Japonicae*. This novel species, described here as *Alternaria telliensis*, is phylogenetically and morphologically distinct from *A. japonica* and *A. nepalensis* in this section. Pathogenicity tests were performed and isolates were found to be weakly pathogenic to tomato and potato (*Solanaceae*) while highly aggressive on radish, cabbage and turnip (*Brassicaceae*) plants.

Introduction

The fungal genus *Alternaria* (*Pleosporaceae*, *Dothideomycetes*) is distributed worldwide and includes many saprobic and plant pathogen species isolated from a wide range of hosts (Lawrence *et al*. 2016). Some species are known to provoke human health disorders such as allergies and infections in people with compromised immune systems (Bush & Prochnau 2004, Pastor & Guarro 2008, Kustrzeba-Wójcicka *et al*. 2014). In addition, species of this fungal genus are often prolific producers of a variety of toxic compounds (Meena *et al*. 2017). The *Alternaria* genus was discribed firstly by Nees (1816) with *Alternaria tenuis* Nees, as the type species. Later, several re-descriptions and revised criteria of this genus and related genera (for review, see Lawrence *et al.* 2016) resulted in a growing number of new species. The taxonomy of *Alternaria* species was based on the shape, size, and septation of the conidia, as well as sporulation patterns amongst which species-groups concept was introduced (Simmons 1992). The description and addition of more phaeodictyosporic similar taxa based on these critera were summerised by Simmons (2007). However, because of the impact of environmental conditions on morphological characteristics, identification of species remained challenging. On the other hand, molecular studies revealed multiple non-monophyletic lineage within *Alternaria* species clades and complex that did not always correlate with the described species-groups (Lawrence *et al*. 2012, 2014, 2016, Woudenberg *et al*. 2013). In recent studies, both morphological and molecular analyses were used in a complementary manner for classification of *Alternaria* genus which has been divided into 28 sections comprising about 280 accepted species (Woudenberg *et al*. 2013, Lawrence *et al*. 2016, Al Ghafri *et al*. 2019, Marin-Felix *et al*. 2019). Since this re-definition of the genus, the list of *Alternaria* species has been continuously growing after re-discriptions and new discovery (Deng *et al*. 2018, Cai *et al*. 2019, Marin-Felix *et al*. 2019, Poursafar *et al*. 2019, Iturrieta-Gonzalez *et al*. 2020, Nishikawa and Nakashima 2020). For example, *A. japonica* was previously linked to the *A. brassicicola* species-

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group (Pryor & Gilbertson 2000, Pryor & Bigelow 2003, Lawrence *et al.* 2013), but this association was questioned by Hong *et al.* (2005) and *A. japonica* was later clustered in section *Japonicae* (Woudenberg *et al*. 2013). Members of this section were characterized by small conidia arranged in a simple or branched chains (Simmons 2007, Woudenberg *et al*. 2013). It includes two important phytopathogenic species (*A. japonica* and *A. nepalensis*) that cause diseases on many *Brassicaceae* plants such as leaf spot of cabbage, cauliflower, turnip, wild and cultivated rocket (Bassimba *et al*. 2013, Tidwell *et al*. 2014, Siciliano *et al*. 2017), black pod of radish (Chalkley 2020), cabbage seedlings damping-off (Ren *et al*. 2012). These species were also reported as seed-borne pathogens (Simmons 2007, Gilardi *et al*. 2015, Chalkley 2020).

In our prevoius studies, many species were reported to cause leaf spot and blight disease of *Solanaceae* in Algeria (Bessadat *et al*. 2014a, 2014b, 2016, 2017, 2019, Ayad *et al*. 2019), including small spored species: two from section *Alternaria* (*A. alternata, A. arborescens*), two from section *Ulocladioides* (*A. consortialis, A. cantlou*s), and four large spored species from section *Porri* (*A. linariae, A. solani, A. protenta, A. grandis*). However, three isolates recovered during a biodiversity survey of *Alternaria* associated with leaf spot and blight of *Solanaceae* had different morplological traits and did not fit any known species previously described on *Solanaceae*. These isolates were collected from three geographic locations (Relizane, Mascara and Mostaganem) of the algerian north-western region. The main objective of the present work was to identifiy these isolates at the species level.

Materials and methods

Sample collection and strain isolation

During a survey of *Alternaria* infecting *Solanaceae* in 2013–2018, samples of leaves and fruits presenting necrotic symptoms were collected in north-western regions of Algeria. Tissue fragments were collected, surface disinfected by dipping in 2% sodium hypochlorite solution for 2 min and rinsed with sterile distilled water three times. Each sample (0.5 cm^2) was placed on Petri dishes with Potato Carrot Agar (PCA) and incubated at room temperature under diffuse day light. About 850 isolates belonged to the genus *Alternaria*. The strains were purified by monospore culture and stored in 30 % glycerol at -80 °C at the COMIC collection of the SFR QUASAV, Angers (France). Among them, three isolates of NB319 (tomato fruit), DA44 (potato leaf) and NB667 (tomato leaf) with unique morphology were isolated from Relizane, Mascara and Mostaganem, respectively. *A. japonica* reference strain CBS 118390 was used for morphological comparison as wall as pathogenicity assay.

Morphological identification

Isolates were subjected to morphological analysis and compared to the literature available (Simmons 2007). All isolates were grown on PCA at room temperature (20 ± 2 °C) under natural day light/dark cycle for 7–12 days. For examination of microscopic details, mycelia were prepared in pure lactic acid without added dye, observed with a Optika microscope (Italy) and photographed. Dimensions were based on observing 150 conidia and 100 conidiophores per isolate. Macro-morphological traits (i. e. colour, aspect and diameter of the colonies) were assessed on PCA, Potato Dextrose Agar (PDA), Oatmeal Agar (OA) and Malt Extract Agar (MEA). The effect of temperature on growth was studied at different temperatures (4 °C, 15 °C, 20 °C, 25 °C, 35 °C and 40 °C). Petri dishes (90 mm) containing 15 ml of each media (PCA, PDA, MEA, OA) were inoculated with 5 mm mycelial discs of 15 day-old culture. Colony diameter was mesured after 7 days of incubation. The type strain (NB667) has also been deposited in the Westerdijk Fungal Biodiversity Institute (Utrecht, The Netherlands) and referenced CBS 145643.

DNA extraction, PCR amplification and nucleotide sequencing

Total genomic DNA was extracted from fresh cultures after seven days at 25 °C using a mini-prep procedure described by Goodwin and Lee (1993) and then stored at -20 °C. The internal transcribed spacer of nuclear ribosomal DNA (*ITS rDNA*) was amplified with the universal primers ITS1 and ITS4 (White *et al*. 1990). Portions of the glyceraldehyde-3-phosphate dehydrogenase (*gpd*), translation elongation factor 1-alpha (*tef1*), RNA polymerase second largest subunit (*rpb2*), plasma membrane ATPase (*ATPase*), actin (*act*), calmodulin (*cmd*) and major *Alternaria* allergen (*Alt a1*) were amplified with primers pairs gpd1/gpd2 (Berbee *et al*. 1999), EF1-728F/EF1-986R (Carbone & Kohn 1999), RPB2–

5F2/fRPB2–7cR (Liu *et al*. 1999, Sung *et al*. 2007), ATPDF1/ATPDR1, ACTDF1/ACTDR1, CALDF1/CALDR1 (Lawrence *et al*. 2013) and Alt-for/Alt-rev (Hong *et al*. 2005), respectively. Amplifications were carried out in 50 µL volumes, with 75 mM Tris-HCl pH 9.0, 20 mM $(NH_4)_2SO_4$, 0.01 % (w/v) Tween 20, 1.5 mM $MgCl_2$, 200 µM each deoxyribonucleotide triphosphate, 1 unit of thermostable DNA polymerase (GoTaq, Promega) and 400 nM of each relevant oligonucleotide primer. Amplification conditions were the same as described in the references provided above. The resulting PCR products were sequenced by GATC Lab (Germany). DNA sequences were deposited in GenBank with the accession numbers provided in Table 1.

TABLE 1. Strains included in the phylogenetic study and GenBank accession numbers.

Section	Species [a]	Strain	GenBank accession No.			
			ITS	***gpd***	***rpb2***	***tef1***
Brassicicola	*Alternaria brassicicola*	CBS 118689	JX499031	KC584103	KC584383	KC584642
	A. conoidea	CBS 132.89	AF348226	FJ348227	KC584452	KC584711
	A. septorioides (T)	CBS 106.41	KC584216	KC584136	KC584427	KC584685
Cheiranthus	*A. cheiranthi*	CBS 10984	AF229457	KC584107	KC584387	KC584646
	A. indefessa (T)	CBS 536.83	KC584234	KC584159	KC584458	KC584717
Dianthicola	*A. dianthicola*	CBS 116491	KC584194	KC584113	KC584394	KC584653
	A. elegans (T)	CBS 109159	KC584195	KC584114	KC584395	KC584654
	A. simsmii (T)	CBS 115265	JF780937	KC584137	KC584428	KC584686
Japonicae	*A. japonica*	CBS 118390	KC584201	KC584121	KC584405	KC584663
	A. nepalensis (T)	CBS 118700	KC584207	KC584126	KC584414	KC584672
	A. telliensis	**DA44**	**MT013034**	**MK904522**	**MK904537**	**MK904549**
	A. telliensis	**NB319**	**MT013033**	**MK904521**	**MK904535**	**MK904548**
	***A. telliensis* (T)**	**NB667=CBS 145643**	**MT013035**	**MK904523**	**MK904536**	**MK904550**
Panax	*A. calycipyricola* (T)	CBS 121545	KC584186	KC584104	KC584384	KC584643
	A. eryngii	CBS 121339	JQ693661	AY562416	KC584397	KC584656
	A. panax	CBS 482.81	KC584209	KC584128	KC584417	KC584675
	A. photistica (T)	CBS 212.86	KC584212	KC584131	KC584420	KC584678
Pseudoulocladium	*A. aspera* (T)	CBS 115269	KC584242	KC584166	KC584474	KC584734
	A. chartarum (T)	CBS 200.67	AF229488	KC584172	KC584481	KC584741
	A. concatenata (T)	CBS 120006	KC584246	AY762950	KC584480	KC584740
	A. septospora	CBS 109.38	FJ266489	FJ266500	KC584487	KC584747
Teretispora	*A. leucanthemi* (T)	CBS 421.65	KC584240	KC584164	KC584472	KC584732
	A. leucanthemi	CBS 422.65	KC584241	KC584165	KC584473	KC584733
Ulocladioides	*Alternaria sp.*	CBS 198.67	AF229487	KC584169	KC584477	KC584737
	A. brassicae-pekinensis (T)	CBS 121493	KC584244	KC584170	KC584478	KC584738
	A. cucurbitae	CBS 483.81	FJ266483	AY562418	KC584483	KC584743
	A. heterospora (T)	CBS 123376	KC584248	KC584176	KC584488	KC584748
	A. thalictrigena (T)	CBS 121712	EU040211	KC584144	KC584436	KC584694
/	*Stemphylium vesicarium* (T)	CBS 191.86	KC584239	AF443884	KC584471	KC584731

[a] T: Ex-type strains. Newly generated sequences and new species are indicated in bold.

Phylogenetic analyses

DNA sequences were concatenated and aligned through ClustalW algorithm and refined manually using MEGA 7 (Kumar *et al*. 2016). Phylogenetic analysis was performed using the maximum likelihood (ML) and Bayesian inference (BI) approaches under IQTree v.1.6.11 (Nguyen *et al*. 2015) and MrBayes v.3.2.1 (Ronquist & Huelsenbeck 2003), respectively. The best-fit evolutionary models for each dataset calculated by ModelFinder (Kalyaanamoorthy *et al*. 2017) under the Bayesian Information Criterion (BIC) selection procedure were TN+F+G (*gpd*) and TNe + G (*tef1* and *rpb2*). The ML analysis was carried out with 1000 ultrafast bootstrap replicates and only values above 70% were considered significant. BI were performed to estimate the posterior probabilities (PP) of tree topologies based on the Markov Chain Monte Carlo (MCMC) analysis with four chains, 1M generations, sampled every 1000 generations. Burn-in was set to 25% and only PP values above 0.95 were considered significant.

Pathogenicity assay

Pathogenicity of two strains (NB667 and DA44) was assessed on two *Solanaceae* species (tomato var. Saint Pierre and potato var. Désirée) and three *Brassicaceae* species (radish var. Redondo Rojo Punta Blanca, cabbage var. capitata alba, turnip var. Croissy). Disinfected seeds and tubers were grown after germination in plastic pots containing sterilized soil: 3/4 potting soil: 1/4 sand mixture. Two month-old plants were inoculated as previously described in Bessadat *et al*. (2017). Conidia suspensions, prepared in sterile distilled water containing 0.01% Tween 80 from 7–14 day-old cultures and adjusted to 10^5 conidia/mL, were sprayed onto plants using manual pressure sprayer. Three replicates were performed for each test and plants treated with sterile distilled water were used as negaive controls. Positive controls viz: NB436 (*A. grandis*) (Bessadat *et al*. 2019) and CBS 118390 (*A. japonica*) were used for comparison on *Solanaceae* and *Brassicaceae*, respectively. After inoculation, plants were covered with plastic bags to maintain high humidity. The plastic bags were removed from plants after 48 h and symptom development was monitored by visual ratings for three weeks to estimate the disease progress. The percentage of leaf necrotic area (l. n. a) was calculated 7, 14 and 21 days after inoculation (DAI) and isolates were rated as weakly pathogenic (0 -29% l. n. a), moderately (30 -60% l. n. a) and highly pathogenic (up to 60% l. n. a).

Results

Molecular phylogenetic analysis

An initial BLASTn analysis of *gpd*, *rpb2* and *tef1* sequences from isolates NB319, NB667 and DA44 in GenBank revealed 95–98% identity with corresponding sequences from *A. japonicae* CBS 118390. Surprisingly, highest scores with strains belonging to section *Ulocladium* were obtained for the ITS locus. For more accurate positioning of these isolates within the *Alternaria* genus, combined *gpd-rpb2-tef1* sequences were aligned with those of 25 other *Alternaria* strains representing 8 sections and one monophyletic lineage. One *Stemphylium* strain (*S. vesicarium* CBS 191.86) was used as outgroup (Table 1). The combined aligned dataset had a total length of 1593 bp (*gpd*: 537 bp, *rpb2*: 841 bp, *tef1*: 215 bp), of which 320 bp were phylogenetically informative (*gpd*: 95 bp, *rpb2*: 165 bp, *tef1*: 60 bp). The topology of the trees inferred by the two phylogenetic methods (ML and BI) was similar. The ML phylogenetic tree with high bootstrap support (99%) and posterior probability values (1.0) confirmed the present isolates belonging to the section *Japonicae* (Fig. 1). In addition, they clearly represented a separate clade supported with 100% bootstrap values and 1.0 posterior probability from the two-described species in this section (*A. japonica* and *A. nepalensis*). It should be considered as members of a new species.

Taxonomy

Alternaria telliensis N. Bessadat, D. Ayad and P. Simoneau ***sp. nov.*** (Fig. 2)
MycoBank: MB 830798

Etymology: name refers to Algerian Tell that corresponds to the northern part of the country where the fungus was isolated.

Specimen examined: Algeria, Mostaganem, Hassi Mamache, isolated from *Lycopersicum esculentum* leaf, Longitude: 0°4′25″W, Latitude: 35°51′29″N, 01 August 2018, (holotype NB 667 = CBS 145643), GenBank accession numbers, MT013035, ITS; MK904523, *gpd*; MK904536, *rpb2*; MK904550, *tef1*; MK913532, *ATPase*; MK904553, *cmd*; MK940319, *act*; MK940315, *alt a1*, collected by Nabahat Bessadat.

Additional specimen examined: Algeria, Relizane, isolated from *Lycopersicum esculentum* leaf, 13 September 2013, isolate NB319 (isotype), GenBank accession numbers, MT013033, ITS; MK904521, *gpd*; MK904535, *rpb2*; MK904548, *tef1*; MK913530, *ATPase*; MK904551, *cmd*; MK940317, *act*; MK940313, *alt a1*, collected by Nabahat Bessadat.

Algeria, Mascara, isolated from *Solanum tuberosum* leaf, 15 september 2014, isolate DA44 (isotype), GenBank accession numbers: MK013034, ITS; MK904522**,** *gpd*; MK904537, *rpb2*; MK904549, *tef1*; MK913531, *ATPase*; MK904552, *cmd*; MK940318, *act*; MK940314, *alt a1*, collected by Djida Ayad.

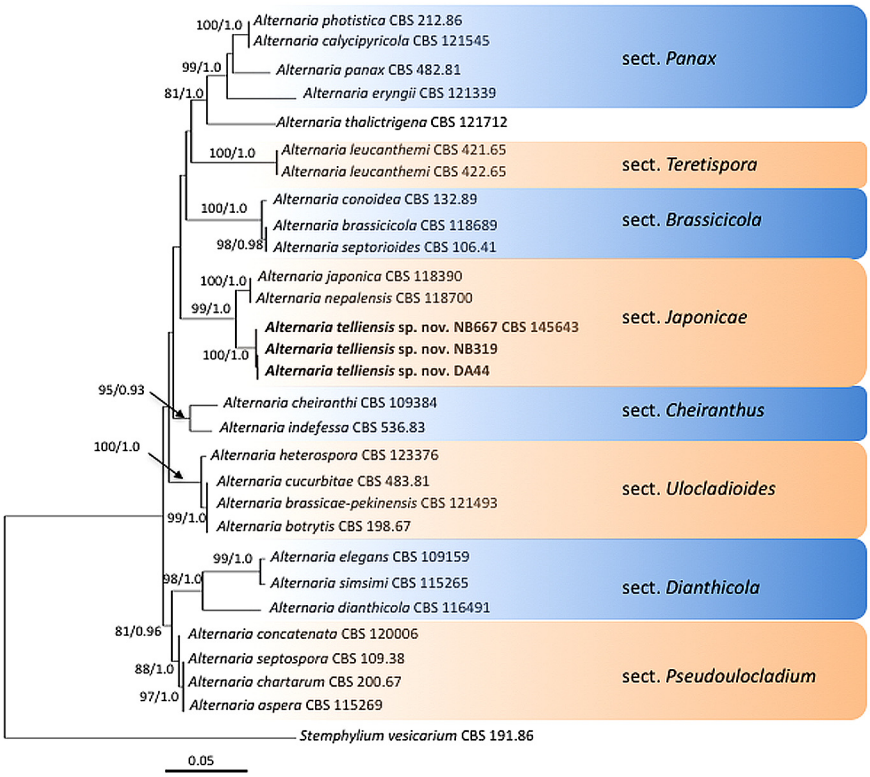

FIGURE1. Maximum Likelihood (ML) tree constructed with *gpd*, *rpb2*, and *tef1* sequences from 28 *Alternaria* strains representatives of eight sections and one monophyletic lineage. The phylogenetic tree was rooted with *Stemphylium vesicarium*. Bootstrap support values for ML greater than 70% and Bayesian posterior probabilities greater than 0.95 are given. Isolates belonging to the newly described species are indicated in bold.

Description: Colonies on PCA are loosely cottony without concentric rings of growth or sporulation (Fig. 2a). Sporulating structures develop directly from surface hyphae in the central area of the colony. Large number of primary conidiophores (45–82(–141) µm long × (5–8(–10) µm wide), with 1–14 transverse septa produced from the aerial portion of the colony 7–14 days after incubation. Conidia are mainly short ovoid, beakless with a rounded apical cell. They are often in short chains of 2–4 (5) conidia (Fig. 2d) or remain solitary (Fig. 2e). Chain formation is by means of apical and lateral secondary conidiophores, arising from terminal cells as well as from any other cell of the conidia body. The initial conidium on a primary conidiophore gives rise to 1–3 secondary conidiophores from which solitary conidia or chains of 2 conidia develop. Secondary conidiophores are simple or branched, septate (1–11 transverse septa), and variable in size (25–55(–116) µm long × 4–8 µm wide). Young conidia almost always are ovoid with 1–3 transverse septa and (15–22(–33) × 11–16(–20) µm in size (Fig. 2f). A high percentage of the spore population matures in culture as ellipsoid, obclavate or broadly ovoid conspicuously constricted at most of the transverse septa; size range is 23–35(–53) × 17–28 µm with 1–4 transverse septa and 1–4 longitudinal septa. Occasional production of asymmetric conidia due to asynchronous division and growth of individual cells yielding an infinity of three-dimensional morphologies (Fig. 2g). These conidia were mainly observed on cabbage leaves (Fig. 2h–i). Conidia are

brown in colour becoming darker with punctuate walls at maturity. Outer walls and all septa become relatively thick and darkly pigmented. Abundant micro-chlamydospores and cellular knots are formed within or on the surface agar substrate upon aging (Fig. 2j–k), giving the colony a dark powdery appearance in center.

FIGURE 2. *Alternaria telliensis sp. nov.* Colony after 7 days on PCA (a), PDA (b) and MEA (c), sporulation pattern on PCA at 14 days (d), conidiophores and mature solitary conidia (e), conidiogenous cells and young conidia (f), asymetric conidia (g), conidia and conidiophores on cabbage leaves (h–i), micro-chlamydospores and cellular knots in hypha surface (j–k). Bars = 50 µm.

Isolates had similar morphology on PDA and developed grey colonies formed by a cottony poorly sporulating mycelium with regular margins (58.1 ± 1.0 mm diameter after 7 days at 25°C). *A. japonica* reference strain (CBS 118390) developped small olivaceous to dull green velvety colonies with irregular margins (37.2 ± 0.9 mm diameter after 7 days at 25°C). The effect of temperature on growth was determined by colony diameter method by using PCA, PDA, MEA and OA medium. The fungus grew best on PCA and PDA at 20–25 °C. Minimum temperatature growth was recorded at 4°C and 35°C. No growth was obseved at 40 °C on all media (Table 3). *A. telliensis sp. nov.* isolates are distinguishable and separable at higher magnifications from *A. japonica* reference strain CBS 118390 (Table 2).

Conidia of *A. nepalensis* and *A. japonica* are long ellipsoid or long ovoid with a rounded apical cell but without a distinct apical beak. Length of mature conidia is up to 35 µm in *A. telliensis* compared to other species of this section which are longer (up to 60 µm). They clearly represent different sporulation patterns and conidial morphology under recommended conditions of Simmons (2007). A hight percetage of *A. telliensis* conidia remain solitary in aerial hyphae tips. The two other species of this section produce abundant short chains of 2–4 conidia, occasionally branched in *A. japonica*. Both *A. telliensis* and *A. japonica* produce micro-chlamydospores in culture unlike *A. nepalensis* where no chlamydospores were observed (Simmons 2007).

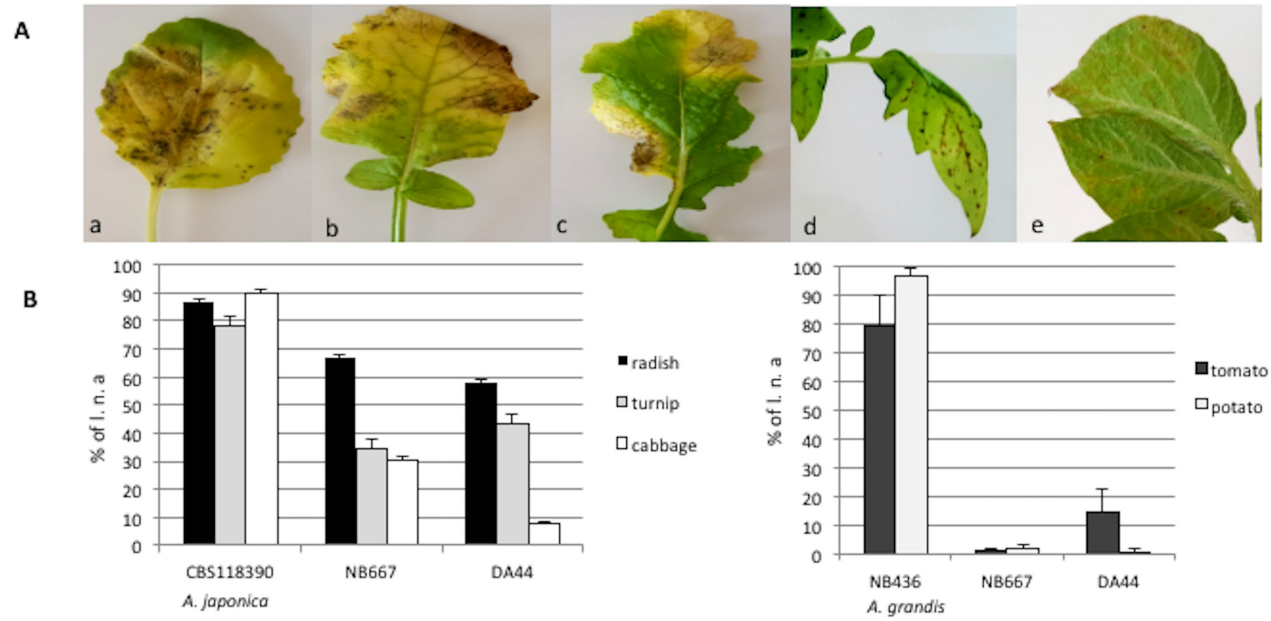

FIGURE 3. A Symptoms on leaves of cabbage (**a**), turnip (**b**), radish (**c**), tomato (**d**) and potato (**e**) at 21DAI with *Alternaria telliensis sp. nov.* (NB667). **B** Percentage and mean of leaf necrotic area (l.n.a) recorded at 21 DAI on radish, turnip, cabbage, tomato and potato leaves inoculated by isolates NB667 and DA44. The *Alternaria japonica* type strain CBS 118390 and *A. grandis* NB436 were included for comparison on *Brassicacea* and *Solanaceae* plants, respectively.

TABLE 2. Comparison of the conidial morphology of *Alternaria telliensis sp. nov.* with species of section *Japonicae* on PCA medium.

Species	Conidia			Conidiophores (µm)	Chlamydo-spores	Reference
	Max. size (µm)	mean size (µm)	Septa			
A. telliensis	35–53 × 17–28	15–33 × 11–20	1–6	45–141 × 3–10	++	Present study
A. japonica	60–109 × 15–23	43–58 × 18–23	5–9	27–108 × 5–7	+++	Present study
A. japonica	80–100 × 20–30	55–70 × 18–22	5–10	55–60 × 5–6	+++	Simmons, 2007
A. nepalensis	60–95 × 15–21	30–60×5–6	1–4	80–125 × 4–5	-	Simmons, 2007

Pathogenicity assay

The pathogenicity of *A. telliensis sp. nov.* was first evaluated on their host plants (tomato and potato) using whole plant inoculation method. The two tested strains (NB667, DA44) were able to provoke lesions on all inoculated plants and showed almost similar results (Fig. 3). Small necrotic spots (1–2 mm) were observed on tomato and potato basal leaves in the first week after inoculation. Lesions size did not expand in the second and third week after inoculaton and did not exceeded 15% of leaf necrotic area (l.n.a) at 21 DAI. No symptoms were observed in control plants while 79% (tomato) to 96% (potato) leaf surfaces were necrotic at 21 DAI with *A. grandis* NB436 used as positive control .

TABLE 3. Culturual characters and temperature effect on *Alternaria telliensis* sp. nov. (NB667) growth after 7 days of incubation on PCA, PDA, MEA and OA.

Medium	Colony Type	colour	Margin	Colour on the reverse side of Petri plates	Sporulation at 25°C	Colony diameter at different temperatures (mm)						
						4°C	15°C	20°C	25°C	30°C	35°C	40°C
PCA	Velvety, fluffy	Greyish-green	White regular	Light brown with grey center	++	10.3 ± 0.6	40.8 ± 1,0	68.8 ± 1.5	77.1 ± 1.9	43.6 ± 1.4	-	-
PDA	Cottony, dense	Greyish-green with a white surface	White irregular	Dark grey with light grey margin	++	12.9 ± 0.3	42.8 ± 1.2	59.8 ± 1.6	58.1 ± 1.0	37.0 ± 0.8	6.0 ± 0.8	-
OA	Cottony, dense	Light green with greyish surface	Regular with grayish rim	Brown with dark center	+	11.9 ± 0.6	31.4 ± 1.1	4.,4 ± 1.7	53.3 ± 0.5	36.8 ± 2.5	6.0 ± 1.4	-
MEA	Cottony, compact	Greyish-green	Regular with white rim	Brown with light grey margin	+	13.1 ± 0.3	38.4 ± 0.5	56.3 ± 6.7	53.4 ± 0.8	38.0 ± 1.6	6.8 ± 0.9	-

*Poor (+) or moderate (++) fungal sporulation.

To assess the host range of *A. telliensis sp. nov.*, three species from *Brassicaceae* (radish, cabbage and turnip) were inoculated with DA44, NB667 and compared to a positive control *A. japonica* (CBS 118390). Highest severity was recorded on *Raphanus sativus* (62% of l.n.a). Isolate DA44 was only weakly aggressive on *Brassica oleracea* plants (7% of l.n.a). Isolates were both moderatly pathogenic toward *Brassica rapa* L (39% of l.n.a). Irrespective of the inoculated plant species, disease severity was always higher with the inoculation of *A. japonica* (radish 87%, cabbage 80%, turnip 78 % of l.n.a). At 21 DAI, inoculated leaves were detached and used to re-isolate the fungus. After 1 week of incubation on PCA plates at room temperature, the fungus was identified as *A. telliensis* by microscopic observations which fulfilled Koch's postulates.

Discussion

Many new *Alternaria* species have been reported in recent years based on DNA sequences analyses, which has dramatically increased the number of accepted species (Marin-Felix et al. 2019) with section *Porri* comprising more than 60 species (Cai *et al*. 2019). Only two species, *A. japonica* and *A. nepalensis*, have been included into section *Japonicae.* In the present study, three strains obtained from *Solanaceae* were assigned to this section on the basis of multi-locus sequence analysis at *gpd, rpb2* and *tef1* loci that were formerly used for redefining the genus (Woudenberg *et al.* 2013). These strains formed a separate clade within the section and could be easily distinguished from *A. japonica* based on morphological characters and were assigned to a new species, *A. telliensis sp. nov.*. The three strains were isolated from ripe tomato fruit and from leaves of potato and tomato cultivated in three different regions of north-western part of Algeria. Species within section *Japonicae* (*A. japonica, A. nepalensis*) occur mainly on *Brassicaceae* (Woudenberg *et al*. 2013). However, there are reports of strains isolated from moth bean and groundnut seeds (Rathod & Chavan 2010), sesame seeds (Nayyar *et al*. 2017) but also tomato seeds (Khulbe & Sati 1987) and identified as *A. japonica* based on morphological characters. Our results show that *A. telliensis sp. nov.* was able to infect their original hosts albeit with low aggressiveness. Besides, severe symptoms on radish and turnip suggesting that *A. telliensis sp. nov.* may be a potential threat for *Brassicaceae.*

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