Journal of Plant Diseases and Protection
https://doi.org/10.1007/s41348-022-00582-y

SHORT COMMUNICATION

New report of *Biscogniauxia rosacearum* as a pathogen on almond trees in Iran

Mahboobeh Sohrabi[1] · Hamid Mohammadi[1] · Josep Armengol[2] · Maela León[2]

Received: 20 July 2021 / Accepted: 24 January 2022

Abstract

Biscogniauxia species are known as fungal trunk pathogens on various tree species in the world. During a survey of trunk diseases of fruit trees conducted in Iran, a branch dieback was observed on almond trees in Kerman province (in the South-east of Iran). Evaluation of symptomatic branches revealed wood discoloration in cross-sections and the presence of fruiting bodies of an ascomycete fungus on their bark. A total of 31 fungal isolates were obtained, 13 isolates recovered from necrotic wood tissues and 18 isolates from fruiting bodies. These isolates were subjected to morphological analysis as well as sequencing analysis of the partial ITS-rDNA and beta tubulin gene sequences. These fungal isolates were identified as *Biscogniauxia rosacearum*. Results of the pathogenicity tests showed that this species is pathogenic on almond shoots. Based on our knowledge, this is the first report of this species on almond trees in Iran and in the world.

Keywords Beta tubulin · Canker · ITS-rDNA · *Prunus dulcis* · Trunk diseases

Introduction

Almond (*Prunus dulcis* (Mill.) Rchb.) is one of the most important nut crops in the family Rosaceae. Iran is known as a center of origin for almond domestication and one of the most important almond producing countries in the world. This country counts for a total production of 128,000 t which represents the third largest production (FAO 2021). Previous studies have shown that almond and other *Prunus* species can be infected by many fungal trunk pathogens worldwide (Damm et al. 2007; Gramaje et al. 2012; Sohrabi et al. 2020). Among them, some species of the genus *Biscogniauxia* Kuntze are known as causal agents of charcoal cankers in various *Prunus* species (Ju et al. 1998; Hilton 2000; Raimondo et al. 2016; Zibarova and Kout 2017). This genus is included into the family Xylariaceae (Ascomycota), and it has been reported from different countries in America (Rogers et al. 2008), Africa (Linnakoski et al. 2012), Asia (Whalley et al. 2012; Rostamian et al. 2016) and Europe (Ragazzi et al. 2012).

Biscogniauxia mediterranea (De Not.) O. Kuntze is one of the most well-known species of this genus, which has been reported as an important fungal trunk pathogen in various tree hosts, including oak, worldwide (Jurc and Ogris 2006; Henriques et al. 2016). This species, along with *B. granmoi* (Fr.) Pouzar and *B. pruni* Granmo, Laessøe & Scheuer, have been previously reported from some *Prunus* species (Ju et al. 1998; Hilton 2000; Zibarova and Kout 2017).

Based on available references, some *Biscogniauxia* species have also been reported affecting various tree species in Iran. These include *B. mediterranea* on *Amygdalus scoparia*, *Quercus brantii, Q. castaneifolia* and *Zelkova carpinifolia*, *B. anceps* (Sacc.) J.D.Rogers, Y.M.Ju & Cand. on *Diospyros lotus*, *Mespilus germanica*, *Parrotia persica* and *Q. castaneifolia*, *B. plana* on *Diospyros lotus* and *Q. castaneifolia* and *B. capnodes* on *Q. castaneifolia* (Mirabolfathy et al. 2011, 2013; Raei et al. 2014; Rostamian et al. 2016; Safaee et al. 2017).

During a study conducted by Raimondo et al. (2016), a new species of *Biscogniauxia,* namely *B. rosacearum* A. Carlucci & M. L., was described from rosaceous hosts and reported as the causal agent of charcoal canker of pear, plum and quince trees in Southern Italy. This fungus has also been

✉ Hamid Mohammadi
hmohammadi@uk.ac.ir

[1] Department of Plant Protection, Faculty of Agriculture, Shahid Bahonar University of Kerman, Kerman, Iran

[2] Instituto Agroforestal Mediterráneo, Universitat Politècnica de València, Camino de Vera S/N, 46022 Valencia, Spain

Published online: 13 February 2022

reported affecting *Q. castaneifolia* and *Vitis vinifera* in Iran (Rostamian et al. 2016; Bahmani et al. 2021).

In 2018, 31 isolates of a *Biscogniauxia* species were obtained from necrotic wood tissues as well as fruiting bodies found on the bark of almond branches showing dieback symptoms in Iran. We found no previous reports of *Biscogniauxia* species on almond trees. Therefore, the aims of this study were to identify these isolates by using morphological and molecular techniques and to verify their pathogenicity to almond.

Materials and methods

Sampling and fungal isolation

During a survey of fungal trunk diseases conducted on almond orchards in Kerman province (Southeast of Iran) in 2018, a branch dieback was observed on almond trees, with the presence of numerous ascomycetous fruiting bodies on the bark of some affected branches. Samples were collected from symptomatic branches as well as fruiting bodies found on the bark of these branches. Fungal isolations were performed from collected samples on potato dextrose agar plates (PDA, Merck, Germany) and incubated at 25 °C in the dark. Prior to morphological and molecular studies, fungal colonies were purified using single spore or hyphal tip methods for isolates recovered from fruiting bodies, or for isolates recovered from necrotic wood tissues, respectively. All cultures were maintained in the culture collection of the Department of Plant Protection at the Shahid Bahonar Univesity of Kerman, Kerman.

Morphological and molecular identification and phylogeny

Isolates were tentatively identified based on ascomata (sexual morph) and fungal colonies (asexual morph) morphology. Microscopic characters of perithecia, asci and ascospores were determined directly from fruiting bodies produced on branches and placing them on water slides. Fungal colonies were grown on MEA (20 g malt extract; 15 g agar; Oxoid, UK), PDA (Biokar diagnostics, France) and oatmeal agar (OA; 60 g oatmeal; Fluca analytical, USA, 12.5 g agar; Oxoid, UK), incubated at 25 °C in the dark for 21 d (Ju and Rugers 1996; Raimondo et al. 2016). The main microscopic structures were mounted on glass microscope slides, and for each structure, 30 measurements were made using a microscope (BH2, Olympus Optical, Tokyo, Japan).

Total genomic DNA was extracted from mycelium of 6 representative pure cultures, 3 obtained from fruiting bodies and 3 from necrotic wood tissues (Table 1) using CTAB method (Doyle and Doyle 1990). PCR amplifications were performed in a 25 µl reaction volume containing 1 µl of template DNA (50 ng/µl), 2.5 µl PCR buffer (10x), 2.5 µl MgCl2 (25 mM), 2.5 µl dNTP (8 mM), 1 µl of each primer (10 µM) and 0.2 µl Horse-PowerTaq DNA polymerase (5 u/µl) (Canvax Biotech, Cordoba, Spain). For sequencing, partial sequences of the internal transcribed spacer 1 and 2 including the intervening 5.8S nrDNA gene (ITS-rDNA) and a part of β-tubulin gene (BT) were amplified using primer sets ITS1/ITS4 (White et al. 1990) and oligonucleotide primers T1 (O'Donnell and Cigelnik 1997) and Bt2b (Glass and Donaldson 1995), respectively. PCR reactions were performed in an Applied Biosystems Veriti 96-well Thermal Cycler (Massachusetts, USA). The program applied for amplification was as follows: 1 cycle of 3 min at 94 °C, 35 cycles of 30 s at 94 °C, 30 s at 55 °C, 60 s at 72 °C; 1 cycle of final extension for 10 min at 72 °C. The PCR product was separated by electrophoresis on a 1.0% agarose gel and visualized by staining with REALSAFE Nucleic Acid Staining Solution (Durviz S.L., Valencia, Spain). PCR products were purified and sequenced by Macrogen (Madrid, Spain) and Bioneer Corporation (Daejeon, South Korea). All sequences run through the BLAST (Basic Local Alignment Search Tool, http://blast.ncbi.nlm.nih.gov/Blast.cgi) to determine their basic identity.

For phylogenetic analyses, *Biscogniauxia* spp. reference sequences were retrieved form GenBank based on the information provided by Raimondo et al. (2016) for the description of the new species *B. rosacearum*. These sequences are representative of the different clades of their phylogenetic analysis (Table 1). Individual loci sequences obtained in this study and those references retrieved from GenBank were aligned using default settings of ClustalW algorithm (Thompson et al. 1994) included within MEGAX software package (Kumar et al. 2018). For multilocus analyses, the single locus alignments were concatenated using SequenceMatrix 1.8 (Vaidya et al. 2011). Phylogenetic analyses for each locus and concatenated datasets were based on Maximum Parsimony (MP), Maximum Likelihood (ML) and Bayesian inference (BI). The MP and ML analyses were conducted in MEGAX (Kumar et al. 2018). Measures calculated for parsimony included tree length (TL), consistency index (CI), retention index (RI) and rescaled consistency index (RC). For ML, the most suitable substitution model selected was HKY + G, which had the lowest Bayesian Information Criterion score in the models feature of MEGA X. In both cases, the robustness of the topology was evaluated by 1000 bootstrap replications (Felsenstein 1985). Bayesian analyses were performed using MrBayes v 3.2 (Ronquist et al. 2012) on the CIPRES Science Gateway V 3.3 (Miller et al. 2010). The best-fitting model of nucleotide evolution for each partition was determined by MrModeltest 2.3 (Nylander 2004) using the Akaike Information Criterion (AIC). Four simultaneous analyses were run for 100 million

Table 1 Origins, host and GenBank accession numbers of the *Biscogniauxia* strains used in phylogenetic analyses (Iranian isolates are shown in bold type)

Isolates		Host	Origin	GenBank accession number	
Species	Code			ITS	BT
Annulohypoxylon cohaerens	YMJ 310	*Fagus* sp.	France	EF026140	AY951655
Biscogniauxia anceps	YMJ 123	*Corylus avellana*	France	EF026132	AY951671
Biscogniauxia arima	YMJ 122[x]	Wood	Mexico	EF026150	AY951672
Biscogniauxia atropunctata	YMJ 128	Wood	United States	JX507799	AY951673
Biscogniauxia cylindrispora	YMJ 89092701[y]	Bark of *Cinnamomum*	Taiwan	EF026133	AY951679
Biscogniauxia formosana	YMJ 8903220[y]	Bark of *Quercus* sp.	Taiwan	JX507802	AY951680
Biscogniauxia mediterranea	YMJ 147	Corticated wood of *Fagus* sp.	France	EF026134	AY951684
	Bx63	*Quercus pubescens*	Italy	KT253501	KT253535
Biscogniauxia rosacearum	CBS 141046[z]	*Prunus domestica*	Italy	KT253493	KT253527
	CBS 141,002	*Cydonia oblonga*	Italy	KT253490	KT253524
	Bx1	*Pyrus communis*	Italy	KT253495	KT253529
	IRNBS7✹	***Prunus dulcis***	**Iran**	**MW452323**	**MW456673**
	IRNBS11✹	***Prunus dulcis***	**Iran**	**MW452325**	**MW456674**
	IRNBS68✹	***Prunus dulcis***	**Iran**	**MW452324**	**MW456675**
	IRNHM-KIR1◆	***Prunus dulcis***	**Iran**	**MZ190889**	**MZ198114**
	IRNHM-KIR2◆	***Prunus dulcis***	**Iran**	**MZ190890**	**MZ198115**
	IRNHM-KIR3◆	***Prunus dulcis***	**Iran**	**MZ190891**	**MZ198116**
Hypoxylon rubiginosum	YMJ 24	Wood of *Fraxinus*	United Kingdom	EF026143	AY951751

[x]Isotype strains

[y]Holotype strains

[z]Ex-type strains

✹ = Isolates recovered from fruiting bodies

◆ = Isolates recovered from necrotic wood tissues

generations, sampling every 1,000, with four Markov Chain Monte Carlo (MCMC) chains. The first 25% of saved trees were discarded and posterior probabilities determined from the remaining trees.

Pathogenicity test

Three isolates obtained from fruiting bodies were selected to conduct pathogenicity tests on detached woody shoots of almond trees (12–15-years-old) under controlled conditions (Table 2). Shoots were cut into 30–35-cm-pieces (2.5 cm in diameter). The outer bark of them was surface-sterilized with 70% ethanol. For each isolate, six shoots were inoculated with a mycelial plug (4 mm in diameter) taken from the margin of a 16-day-old fungal colony on PDA, and six shoots were also inoculated with non-colonized, sterile agar plugs as control treatments. All inoculated areas were protected by moist cotton and covered with Parafilm

Table 2 Mean lesion length and re-isolation frequencies of three representative *Biscogniauxia rosacearum* isolates inoculated onto almond tree shoots in the pathogenicity test

Isolates		Mean lesion length (mm)			Re-isolation frequency (%)
Species	Code (IRN)	Up	Down	Total	
Biscogniauxia rosacearum	IRNBS68	107.00 **A**	85.50 **A**	192.50 **A**	85.7
	IRNBS11	78.50 **B**	77.83 **AB**	156.33 **AB**	76.2
	IRNBS7	62.33 **B**	55.83 **B**	118.17 **B**	57.1
Control	–	3.17 **C**	4.33 **C**	7.50 **C**	0
LSD ($P < 0.05$)	–	27.114	28.121	51.744	–

Different letters in bold face indicate significant differences only within a column at $P < 0.05$

(Pechiney Plastic Packaging, USA). The base of inoculated and control shoots were inserted into plastic containers, filled with 1500 ml distilled water and then kept at 25 ± 2 °C. This experiment was arranged in a completely randomized design. After 40 days of incubation, all inoculated shoots were collected, and the length of wood discoloration was measured from the point of inoculation. Re-isolation of fungal isolates was made from the margins of necrotic lesions as described previously to fulfill Koch's postulates, and fungal identity was verified based on colony morphology and microscopic structures. The significance of differences in mean lesion lengths were determined by one-way analysis of variance (Proc ANOVA) using SAS v. 9.1 (SAS Institute, Cary, North Carolina, USA), and the LSD test was used for comparison of treatment means at $P < 0.05$.

Results

Morphological characterization

In this study, 31 fungal isolates were recovered from necrotic wood tissues (13 isolates) and fruiting bodies (18 isolates). The asexual morph produced a *Nodulisporium*-like colony, as defined by Ju and Rogers (1996). Colonies reached a radius of 39.5 mm in 7 d at 25 °C. Colonies on MEA were floccose with aerial mycelium and entire margin, olive gray, after 14 d; colonies on PDA were olive with aerial mycelium and entire margin, after 14 d, and on OA, they were gray to pale olive with aerial mycelium and entire margin, after 14 d. Conidiophores were hyaline to brownish, smooth to finely roughened, dichotomously branched, with two or three, and sometimes more, conidiogenous cells on each terminus. Conidia were hyaline, smooth, obovoid to clavate, 5.25-µm-long and 2.10-µm-wide (Fig. 1). Perithecia were dark brown, 190–285 µm in diameter and

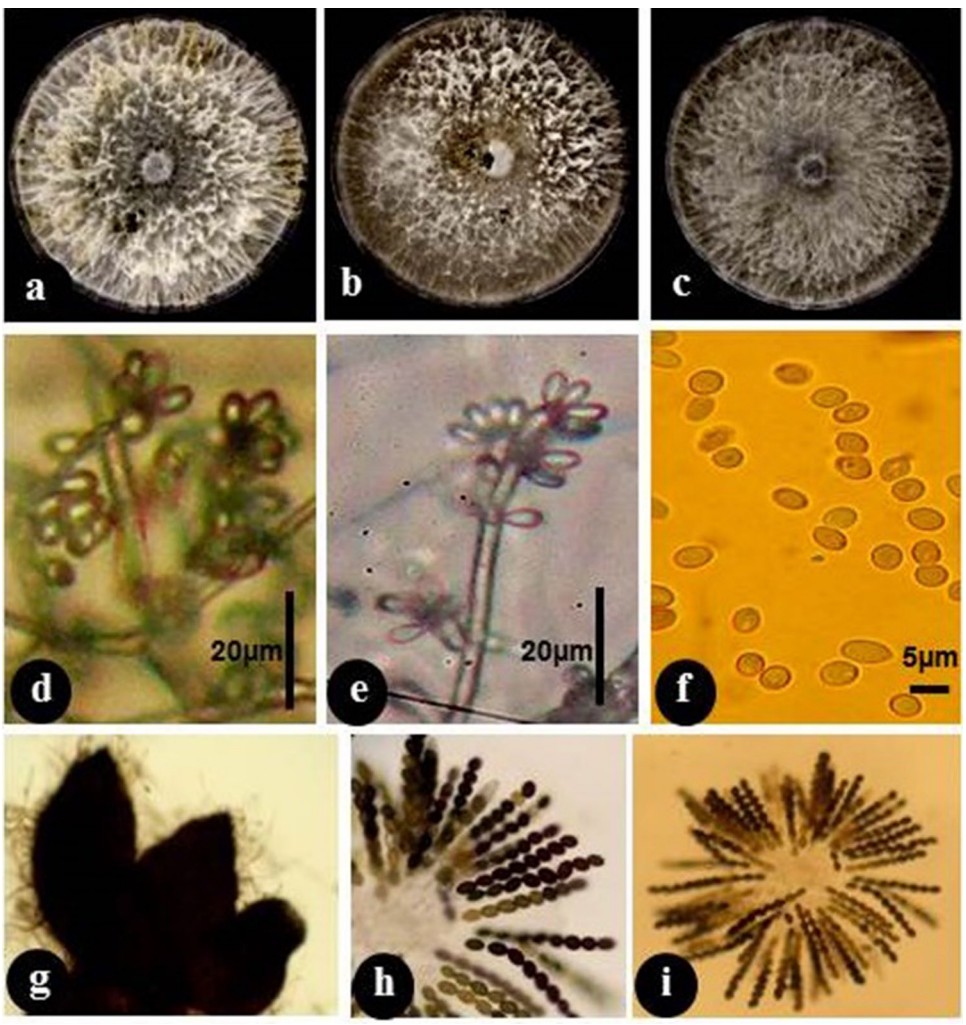

Fig. 1 *Biscogniauxia rosacearum*, isolate IRNBS11. **a** Colony on PDA, **b** MEA, **c** OA after 14 days. **d, e** Branched conidiophores and phialides, **f** Conidia, **g** Ascocarp, **h, i** asci and ascospores. Scale bar = 20 µm, f = 5 µm

680–890 µm in high, asci: cylindrical, 85–105-µm-long and 5.5–8.9-µm-wide, ascospores: brown to dark brown, unicellular, smooth, 7.9–9.8-µm-long and 4.3–5.2-µm-wide.

Molecular characterization

For phylogenetic analyses, the ITS and BT sequences were obtained for six representative isolates and aligned with 10 reference sequences and two out-groups. Phylogenies resulting from the individual locus were compared visually, and no differences were detected between them, indicating that both datasets could be combined. The combined alignment consisted of 1725 characters including gaps (ITS: 725 and BT: 1000). Of these, 749 were constant and 444 parsimony informative. Maximum parsimony analysis resulted in seven equally most parsimonious trees (TL = 1595, CI = 0.662; RI = 0.651, RC = 0.431). The topology of the trees identified by all phylogenetic analysis of concatenated dataset (BI, ML and MP) were identical, therefore only the BI tree is presented with BI posterior probability scores and ML and MP bootstrap support values at the nodes. The Iranian isolates clustered with the reference isolate of *B. rosacearum* (Fig. 2). Based on the morphology and cultural characteristics and molecular data, the isolates were identified as *B. rosacearum*.

Pathogenicity test

All inoculated isolates produced wood discoloration upward and downward from the point of inoculation after 40 days of incubation (Fig. 3). The isolate IRNBS68 was the most aggressive, causing the longest (192.5 mm) wood lesions, although there was no significant difference between length of lesions produced by this isolate and the isolate IRNBS11 (156.3 mm). The isolate IRNBS7 gave the shortest lesions (118.2 mm) on detached shoots of this host. On detached shoots, wood lesion lengths caused by all fungal isolates were statistically different to those observed on the non-inoculated controls. The mean of upward and downward wood discoloration lengths caused by the three isolates were significantly larger than in control treatments. The highest percentage recovery was obtained for IRNBS68 (85.7%), the lowest for IRNBS7 (57.1%). *Biscogniauxia rosacearum*

Fig. 3 Pathogenicity tests of *Biscogniauxia rosacearum* isolates inoculated onto detached shoots of almond trees 35 days after inoculation: **1** Control (the arrow shows the point of inoculation), **2** IRNBS68, **3** IRNBS11, **4** IRNBS7

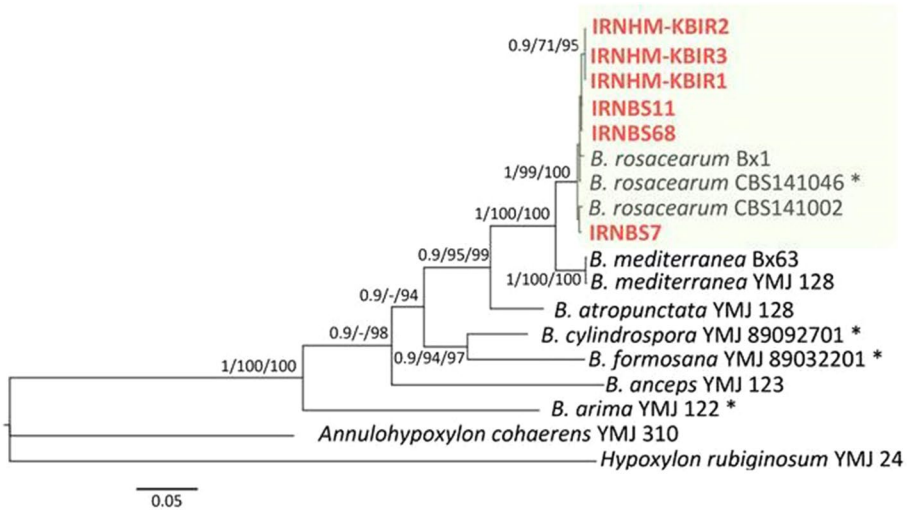

Fig. 2 Phylogenetic tree based on Bayesian Inference analyses of combined ITS and BT sequence data. *Annulohypoxylon cohaerens* (YMJ 310) and *Hypoxylon rubiginosum* (YMJ 24) were used as outgroup, and Iranian isolates obtained in this study are shown in red color. Support values [Bayesian inference (BI) posterior probabilities/maximum likelihood (ML) bootstrap/maximum parsimony (MP) bootstrap] are given at the nodes. Bootstrap values less than 70% are indicated with "- ". Type strains are indicated with an asterisk. Scale bar shows expected changes per site

was not re-isolated from the small lesions observed on the control shoots.

Discussion

Based on morphological characteristics and molecular analysis, all isolates of the *Biscogniauxia* species obtained from almond trees were identified as *B. rosacearum*. The genus *Biscogniauxia* as a Xylariaceous pyrenomycete was monographed by Ju et al. (1998). Species of this genus are endophytes or secondary pathogens that can infect various tree species under stress conditions (Raimondo et al. 2016). Some *Biscogniauxia* species are known to cause charcoal canker, trip canker, wood decay and decline of forest and fruit trees (Hendry et al. 1998; Granata and Sidoti 2004; Raimondo et al. 2016). *Biscogniauxia mediterranea* as the most well-known pathogen species in this genus has a worldwide distribution and causes charcoal canker disease on *Quercus* L. species and other forest and ornamental trees (Jurc and Ogris 2006; Ragazzi et al. 2012; Henriques et al. 2016). Previous studies have also shown that some *Biscogniauxia* species are able to infect *Prunus* species. These include of *B. granmoi* on *Prunus padus* in Austria (Raimondo et al. 2016) and Czech Republic (Zibarova and Kout 2017), *B. pruni* on *Prunus padus* in Austria and Latvia (Ju et al. 1998) and *B. mediterranea* on *Prunus emarginata* in Canada and British Columbia (Hilton 2000). *Biscogniauxia rosacearum* is a new species of *Biscogniauxia,* which was first described from rosaceous hosts and reported as the causal agent of charcoal canker of *Pyrus communis*, *Prunus domestica* and *Cydonia oblonga* in Southern Italy (Raimondo et al. 2016). This fungus has also been isolated from insect gallery systems in oak trees in Italy (Pinna et al. 2019) and olive trees in South Africa (Spies et al. 2020). More recently, this pathogen has also been reported from *Q. castaneifolia* and grapevines in Iran (Rostamian et al. 2016; Bahmani et al. 2021). Based on our knowledge, this is the first report of *B. rosacearum* on almond trees in Iran and in the world.

Results of the pathogenicity tests on almond shoots showed that all isolates of *B. rosacearum* inoculated on almond shoots were pathogenic on this host. Our results are consistent with findings of previous pathogenicity studies on pear, plum and quince trees in Southern Italy and grapevine in Iran, where inoculation of this fungus on wood stems showed that this fungus can be considered as a pathogen on these hosts (Raimondo et al. 2016; Bahmani et al. 2021). Six isolates out of total isolates obtained during this study were selected for molecular studies. These isolates had generally the same morphological features and grouped together in the phylogenetic tree, being the same species. Of them, 3 isolates were used for pathogenicity tests, which showed variation in their pathogenicity. Bahmani et al. (2021) also found variable symptoms when they inoculated *B. rosacearum* isolates on grapevines, including yellowing, necrosis of leaves, internal wedge-shape necrosis of the vascular tissue and brown lesions which expanded upward and downward of the inoculation point on the stems. Differences observed among isolates in the pathogenicity tests could also be related with their genetic variability observed in the phylogenetic analyses of such as, for instance, on the isolate IRNBS7. Henriques et al. (2014) also reported relevant genetic variability among isolates of *B. mediterranea* obtained from the same stroma on cork oak trees. Our study confirmed that almond trees can also be considered as a woody host for *B. rosacearum*. Therefore, further studies are needed to determine the importance of this fungus on almond and other fruit trees in Iran.

Acknowledgements This research was conducted at Shahid Bahonar University of Kerman, Kerman, Iran.

Authors' contribution Conceptualization: MS and HM; Methodology: MS, HM, JA and ML; Formal analysis and investigation: MS; Writing—original draft preparation: MS; Writing—review and editing: MS, HM, JA and ML; Supervision: HM.

Funding The first author is financially supported by the Iranian Ministry of Science, Research and Technology (MSRT) just for her 4 months stay in Spain as a part of her PhD project. However, no funding was received from MSRT for this study by the authors.

Declarations

Conflict of interest All authors certify that they have no affiliations with or involvement in any organization or entity with any financial interest or non-financial interest in the subject matter or materials discussed in this manuscript.

References

Bahmani Z, Abdollahzadeh J, Amini J, Evidente A (2021) *Biscogniauxia rosacearum* the charcoal canker agent as a pathogen associated with grapevine trunk diseases in Zagros region of Iran. Sci Rep 11:14098

Damm U, Crous PW, Fourie PH (2007) Botryosphaeriaceae as potential pathogens of *Prunus* species in South Africa, with descriptions of *Diplodia africana* and *Lasiodiplodia plurivora* sp. nov. Mycologia 99:664–680

Doyle JJ, Doyle JL (1990) Isolation of plant DNA from fresh tissue. Focus 12:13–15

FAOSTAT (2021) Food and agriculture organization of the United Nations. http://www.fao.org/faostat/es/#dat

Felsenstein J (1985) Confidence limits on phylogenies: an approach using the bootstrap. Evolution 39:783–791

Glass NL, Donaldson GC (1995) Development of primer sets designed for use with the PCR to amplify conserved genes from filamentous ascomycetes. Appl Environ Microbiol 61:1323–1330

Gramaje D, Agusti-Brisach C, Perez-Sierra A, Moralejo E, Olmo D, Mostert L, Damm U, Armengol J (2012) Fungal trunk pathogens associated with wood decay of almond trees on Mallorca (Spain). Persoonia 28:1–13

Granata G, Sidoti A (2004) *Biscogniauxia nummularia*: pathogenic agent of a beech decline. For Pathol 34:363–367
Hendry SJ, Lonsdale D, Boddy L (1998) Strip-cankering of beech (*Fagus sylvatica*): pathology and distribution of symptomatic trees. New Phytol 140:549–565
Henriques J, Nóbrega F, Sousa E, Lima A (2014) Diversity of *Biscogniauxia mediterranea* within single stromata on cork oak. J Mycol article ID 324349:5p
Henriques J, Nóbrega F, Sousa E, Lima A (2016) Analysis of the genetic diversity and phylogenetic relationships of *Biscogniauxia mediterranea* isolates associated with cork oak. Phytoparasitica 44:19–34
Hilton SEd (2000) Canadian plant disease survey. Agric Agri-Food Can 80:151
Ju YM, Rogers JD (1996) A revision of the genus *Hypoxylon*. American Phytopathological Society, St. Paul, MN
Ju YM, Rogers JD, San Martin F, Granmo A (1998) The genus *Biscogniauxia*. Mycotaxon 66:1–98
Jurc D, Ogris N (2006) First reported outbreak of charcoal disease caused by *Biscogniauxia mediterranea* on Turkey oak in Slovenia. Plant Pathol 55:299–299
Kumar S, Stecher G, Li M, Knyaz Ch, Tamura K (2018) MEGA X: molecular evolutionary genetics analysis across computing platforms. Mol Biol Evol 35:1547–1549
Linnakoski R, Puhakka-Tarvainen H, Pappinen A (2012) Endophytic fungi isolated from *Khaya anthotheca* in Ghana. Fungal Ecol 5:298–308
Miller MA, Pfeiffer W, Schwartz T (2010) Creating the CIPRES Science Gateway for inference of large phylogenetic trees. In Proceedings of the Gateway Computing Environments Workshop (GCE), New Orleans, LA: IEEE (pp 1–8)
Mirabolfathy M, Groenewald JZ, Crous PW (2011) The Occurrence of charcoal disease caused by *Biscogniauxia mediterranea* on chestnut- leaved oak (*Quercus castaneifolia*) in the Golestan forests of Iran. Plant Dis 95:876
Mirabolphathy M (2013) Outbreak of charcoal disease on *Quercus* spp. and *Zelkova carpinifolia* trees in forest of Zagros and Alborz Mountains in Iran. Iran J Plant Pathol 49:257–263
Nylander JAA (2004) MrAIC.pl. Program distributed by the editor. Evolutionary Biology Centre, Uppsala University
O'Donnell K, Cigelnik E (1997) Two divergent intragenomic rDNA ITS2 types within a monophyletic lineage of the fungus *Fusarium* are non orthologous. Mol Phylogenet Evol 7:103–116
Pinna C, Linaldeddu BT, Deiana V, Maddau L, Montecchio L, Lentini A (2019) Plant pathogenic fungi associated with *Coraebus florentinus* (*Coleoptera*: *Buprestidae*) attacks in declining oak forests. Forests 10:1–12
Raei S, Khodaparast SA, Abbasi M (2014) More records of xylariaceous fungi from North of Iran. Rostaniha 15:110–121
Ragazzi A, Ginetti B, Moricca S (2012) First report of *Biscogniauxia mediterranea* on English ash in Italy. Plant Dis 96:1694
Raimondo ML, Lops F, Carlucci A (2016) Charcoal canker of pear, plum, and quince trees caused by *Biscogniauxia rosacearum* sp. nov. in Southern Italy. Plant Dis 100:1813–1822
Rogers JD, Vasilyeva LN, Hay FO (2008) New *Xylariaceae* from Hawaii and Texas (USA). Sydowia 60:277–286
Ronquist F, Teslenko M, van der Mark P, Ayres DL, Darling A, Höhna S, Huelsenbeck JP (2012) MrBayes 3.2: efficient Bayesian phylogenetic inference and model choice across a large model space. Syst Biol 61:539–542
Rostamian M, Kavosi MR, Bazgir E, Babanezhad M (2016) First report of *Biscogniauxia mediterranea* causing canker on wild almond (*Amygdalus scoparia*). Australas Plant Dis Notes 11:30
Safaee D, Khodaparast SA, Mirabolfathy M, Mousanejad SA (2017) Multiplex PCR-based technique for identification of *Biscogniauxia mediterranea* and *Obolarina persica* causing charcoal disease of oak trees in Zagros forests. Forest Pathol 47:e12330. https://doi.org/10.1111/efp.12330
Sohrabi M, Mohammadi H, León M, Armengol J, Banihashemi Z (2020) Fungal pathogens associated with branch and trunk cankers of nut crops in Iran. Eur J Plant Pathol 157:327–351
Spies CFJ, Mostert L, Carlucci A, Moyo P, van Jaarsveld WJ, Plessis IL, van Dyk M, Halleen F (2020) Dieback and decline pathogens of olive trees in South Africa. Persoonia 45:196–220
Thompson JD, Higgins DG, Gibson TJ (1994) Clustral W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. Nucleic Acids Res 22:4673–4680
Vaidya GD, Lohman J, Meier R (2011) Sequence Matrix: concatenation software for the fast assembly of multi-gene datasets with character set and codon information. Cladistics 27:171–180
Whalley AJS, Phosri C, Ruchikachorn N, Sihanonth P, Sangvichien E, Suwannasai N, Thienhirun S, Whalley MA (2012) Interesting or rare Xylariaceae from Thailand. Rajabhat J Sci Human Soc Sci 13:9–19
White TJ, Bruns T, Lee S, Taylor J (1990) Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sninsky JJ, White TJ (eds) PCR protocols, a guide to methods and applications. Academic Press, San Diego, CA, USA, pp 315–322
Zibarova L, Kout J (2017) Xylariaceous pyrenomycetes from Bohemia: species of *Biscogniauxia* and *Hypoxylon* new to the Czech Republic, and notes on the other rare species. Czech Mycol 69:77–108