

1 **Outbreak of *Phytophthora cinnamomi* causing severe decline of avocado trees in southern Turkey**

2

3 By İ. Kurbetli¹, G. Sülü¹, M. Aydoğdu¹, S. Woodward² and S. Bayram¹

4

5 1: Plant Health Department, Bati Akdeniz Agricultural Research Institute, Antalya, Turkey

6 2: University of Aberdeen, School of Biological Sciences, St. Machar Drive, Aberdeen AB24 3UU, Scotland, UK

7

8

8 **Summary**

9

10 Since the summer of 2017, severe decline symptoms have been observed on 10- to 25-year-old
11 avocado trees in almost all commercial orchards planted in the Mediterranean coastal region of
12 Turkey. Young, newly planted trees in infected orchards were also affected by the disease. Affected
13 trees showed wilting, leaf discoloration, defoliation and severe dieback. Some trees were
14 completely desiccated. Although fine roots of symptomatic trees usually were decayed, reddish
15 brown cankers also occurred on taproots and lateral roots, of heavily infected trees. The pathogens
16 were isolated from necrotic root and soil samples of symptomatic trees, using selective medium and
17 soil baiting, and were identified based on morphological features and DNA sequences. One isolate
18 each of *Phytophthora cryptogea* and *P. palmivora* were identified, while all other isolates were *P.*
19 *cinnamomi*. In addition, a subcortical fan-shaped mycelium, characteristic of *Armillaria* spp. was
20 observed in the crown of a symptomatic tree and identified as *Armillaria gallica* by DNA
21 sequences. Pathogenicity of *Phytophthora* isolates was tested by stem inoculation on avocado
22 seedlings. Two months after inoculation, canker lesions developed on stems of seedlings inoculated
23 by any of the three *Phytophthora* spp.. In contrast, collenchyma callus formed over the wound
24 points on control plants over the same time period. This is the first report of *P. cinnamomi*, *P.*
25 *cryptogea*, *P. palmivora* and *A. gallica* causing root rot of avocado trees in Turkey. In addition, *P.*
26 *cryptogea* and *A. gallica* are reported for the first time associated with disease on this host. Due to
27 the severe symptoms and widespread occurrence, *P. cinnamomi* should be considered a potential
28 threat to avocado cultivation and natural ecosystems of this region of Turkey.

29

30

30 **1 Introduction**

31 Avocado (*Persea americana* Miller) is a tree species native in Guatemala, Central America and
32 Mexico (Henaó et al., 2017). It is a significant and nutritious fruit crop grown in both the tropical
33 and subtropical regions in many parts of the world (Menge et al., 2012). World production of
34 avocados in 2017 was estimated at approx. 6 million tonnes with the highest production in Mexico.
35 other important avocado producing countries include Dominican Republic, Peru, Indonesia,
36 Columbia, Brazil, Kenya, Venezuela, Chile, the United States (California), China and Guatemala

37 (FAOSTAT, 2019). Avocado production is currently increasing in Turkey, such that export of this
38 fruit have begun.

39 *Phytophthora cinnamomi* is the most important Oomycota species damaging forest trees and is also
40 destructive in woody ornamentals and orchard crops including avocado (Robin et al. 2012).
41 Avocado root rot caused by *P. cinnamomi* has long been known as the main disease of this crop
42 throughout the world (Zentmyer, 1980; Erwin & Ribeiro, 1996). The pathogen has reduced
43 production in many areas of the world, becoming the major limiting factor in production in
44 Australia, California, Mexico, South Africa and Spain (Ploetz et al., 2002; Perez-Jimenez 2008). In
45 Mexico, the pathogen has been present in all the main avocado production areas with incidence
46 varying between 5 and 90%, depending on the region (Perez-Jimenez 2008). California avocado
47 groves were affected by the disease with incidence of 60 – 70%, and estimated annual losses of over
48 \$30 million (Coffey, 1992; Erwin & Ribeiro, 1996). In Eastern Australia, the pathogen is wide-
49 spread, seriously affecting avocado production (Pegg et al., 1987). In South Africa, the number of
50 infected orchards increased rapidly once the crop was introduced, and by the early 1970s, the
51 disease was estimated to affect 20% of all trees (Milne & Chamberlain, 1971). In the Andalusian
52 region of Spain, 40% of avocado orchards were invaded by *P. cinnamomi* (Perez-Jimenez et al.
53 2005). Approximately half of the avocado orchards established in the Canary Islands of Spain are
54 affected by the disease (Rodriguez-Padron et al. 2018). In Israel, the pathogen was first isolated
55 from avocado in 1982, and the number of orchards infested over subsequent years increased (Perez-
56 Jimenez 2008).

57 Apart from *P. cinnamomi*, several other *Phytophthora* species, including *P. cactorum*, *P. citricola*,
58 *P. citrophthora*, *P. heveae*, *P. nicotianae* and *P. palmivora*, have been reported worldwide affecting
59 avocado trees (Erwin & Ribeiro, 1996). Recently, *P. citricola* isolates known to cause avocado
60 trunk canker, were renamed *P. menzei* (Hong et al., 2009). More recently, avocado orchards were
61 surveyed in the Canary Islands and *Phytophthora* species, including *P. cinnamomi*, *P. multivora*, *P.*
62 *niederhauserii*, *P. nicotianae*, and *P. palmivora* were obtained from the roots of symptomatic trees
63 or orchard soils (Rodriguez-Padron et al., 2018). The most frequently isolated species was *P.*
64 *cinnamomi*, whereas the most virulent species was *P. niederhauserii* in that study.

65 In periodic observations since the summer 2017, decline symptoms due to root rot were observed on
66 10- to 25-year-old avocado trees in commercial orchards in Mediterranean coastal parts of Turkey,
67 where almost all avocados in the country are produced. The aim of the work reported here was to
68 identify the causal agents of the root rot and decline symptoms on avocados, including evaluations
69 of pathogenicity in controlled environment tests.

70

2 Materials and Methods

Sampling and isolations

Commercial avocado orchards of Turkey are located in the Alanya and Gazipaşa districts of Antalya province, and the Anamur district of Mersin province in southern Turkey (Fig. 1). This region, located between 36° and 37° north, has a sub-tropical climate and almost all avocado production in Turkey is centered there. Avocado orchards in this region were surveyed for plants with symptoms of root rot in 2018 to 2019. During the surveys, wilting, leaf discoloration, defoliation and severe dieback symptoms, sometimes resulting in tree mortality, were frequently observed in the orchards (Fig. 2). Fine roots of symptomatic trees were rotted. Reddish brown cankers formed on taproots and lateral roots of some heavily infected trees. Root and soil samples were collected from the roots of a single symptomatic tree in each of 23 different orchards. In addition, fan-shaped mycelium under the bark of the lower stem of a declined tree was observed in a single orchard (Fig. 2). Wood tissue including the mycelium was also sampled from that tree.

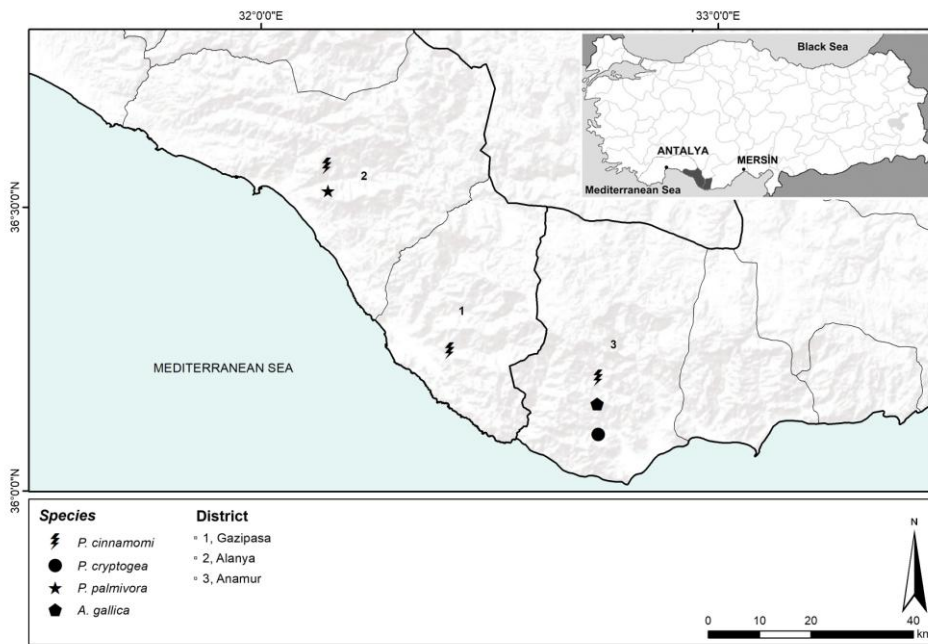
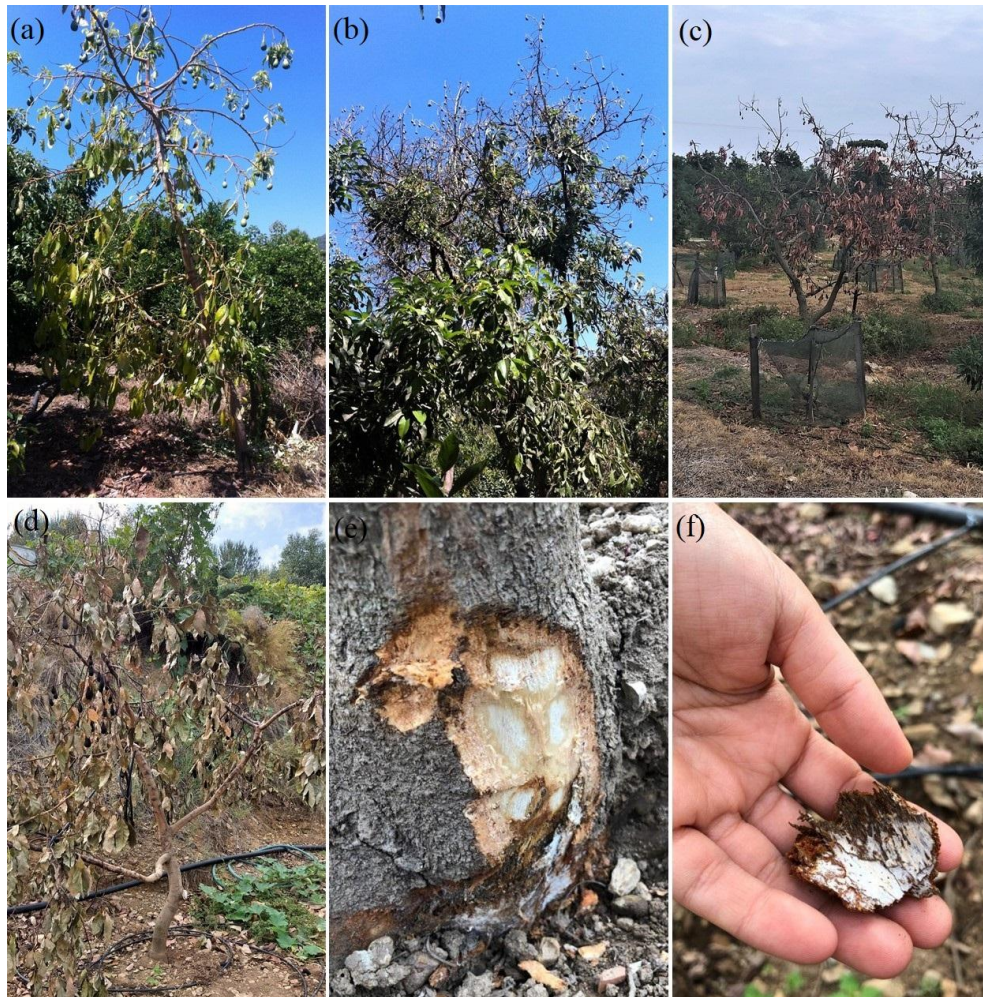


Figure 1. Distribution of *Phytophthora* spp. Alanya and Gazipaşa districts of Antalya province, and Anamur district of Mersin province.



88

89 Figure 2. Wilting and defoliation (a); dieback (b); and decline (c) symptoms of *Phytophthora* spp; decline symptom (d) and fan shaped mycelium of
 90 *Armillaria* sp. (e-f) on avocado trees.

91 Necrotic roots of symptomatic avocado trees were washed in running tap water and air-dried. Small
 92 pieces of tissue, 3 – 5 mm³ were excised from the lesion margins from taproots, fine roots and stem
 93 bases and placed onto PARP semi selective medium (Jeffers & Martin, 1986) without further
 94 surface sterilisation (Fig. 3). Cultures were incubated at 20–22°C in the dark and any hyphal growth
 95 emerging examined after 2–3 days under a light microscope. Single hyphal tips of emerging
 96 colonies with coenocytic hyphae having wide branching angles were excised and transferred to
 97 carrot juice agar (CA: 200 ml boiled carrot juice, 800 ml distilled water, 20 g agar) or V8 juice agar
 98 (200 ml clarified V8 juice, 800 ml distilled water, 20 g agar) to obtain pure cultures.

99 Soil samples were taken 1–1.5 m from stem bases at a depth of 10–30 cm beneath the soil organic
 100 horizon. Soil was not used in isolation attempts when at least one isolate of *Phytophthora* sp. was
 101 obtained from the root or collar tissues of a tree. Approximately 500 mL of each soil sample was
 102 flooded with distilled water. The organic material floating on the water surface was removed with
 103 cheesecloth. Young leaves of avocado cv. Topa Topa were floated on the water as baits and the
 104 traps maintained at room temperature. Leaves on which dark or brownish lesions appeared after 3–5

105 days at 22–24°C were cut into small segments (5 mm²) and placed onto PARP medium (Fig. 3).
106 Cultures were incubated and subcultured as described above.
107



108
109 Figure 3. Necrotic root tissues obtained from avocado trees (a-b); *Phytophthora* isolations from root pieces (c) and leaflets in bait (d).
110

111 *Identification of Phytophthora spp. and Armillaria sp.*

112 Isolates of *Phytophthora* spp. were identified on the basis of morphological characteristics and by
113 molecular analysis of the ITS region of the rDNA. Morphological identification was based on
114 colony morphology, and microscopic structures such as hyphal swellings, chlamydozoospores and
115 morphological features of sporangia (Erwin & Ribeiro, 1996; Gallegly & Hong, 2008). When half
116 the surface of CA or V8 juice agar in Petri dishes was covered by a colony, 5-mm-diameter agar
117 disks were cut from the growing edge were placed in 6-cm-diameter Petri dishes previously flooded
118 with 7 ml rain water to induce the formation of sporangia. Colony morphology was described on
119 CA, CMA, malt extract agar (MEA), potato dextrose agar (PDA) and V8A. The ability of the
120 isolates to grow at 35°C was determined on CA and PDA after incubation for 5 days.

121 Morphological identification was confirmed by ITS sequences of rDNA. Internal transcribed spacer
122 (ITS) regions of ribosomal DNA (rDNA) of 6 isolates of *Phytophthora* spp. were amplified using
123 the universal primer pairs ITS-6 (5'-GAAGGTGAAGTCGTAACAAGG-3') and ITS-4 (5'-
124 TCCTCCGCTTATTGATATGC-3'); for the single isolate of *Armillaria* sp., ITS-1 (5'-
125 TCCGTAGGTGAACCTGCGG-3') and ITS-4 (5'-TCCTCCGCTTATTGATATGC-3') were used.
126 In addition, the translational elongation factor 1- α (EF 1- α) gene region of an isolate of *Armillaria*
127 sp. was amplified using primers EF595F (5'-CGTGACTTCATCAAGAACATG-3') and EF1160R

128 (5'-CCGATCTTG TAGACGTCCTG-3') (Maphosa et al., 2006). Mycelium of *Armillaria* sp. used
129 in this work was taken directly from wood tissues.

130 PCR products were separated in 2% agarose gels, stained with safe DNA dye and visualized under
131 UV light. Sequence analysis was carried out by GENOKS (Ankara, Turkey). Sequences were
132 subjected to BLAST searches on GenBank (<http://www.ncbi.nlm.nih.gov>) to find the closest
133 matches.

134 For phylogenetic analysis, the sequences generated in this study (ITS) were supplemented with
135 additional sequences of *P. cinnamomi*, *P. cryptogea*, *P. palmivora* and *A. gallica* obtained from
136 GenBank (Table 1). Evolutionary history of the isolates was inferred using the Neighbor-Joining
137 method (Saitou & Nei, 1987). Evolutionary distances were computed using the Maximum
138 Composite Likelihood method (Tamura et al., 2004) and presented in units of the number of base
139 substitutions per site. Evolutionary analyses were conducted in MEGA7 (Kumar et al., 2016).

140 Table 1. Accession numbers of isolates obtained from GenBank used in construction of phylogenetic trees.

| Species | Culture accession No. | GenBank accession |
|---------------------|-----------------------|-------------------|
| <i>P. cinnamomi</i> | Pc06Bb | MN960453 |
| <i>P. cinnamomi</i> | PN2035 | MH236250 |
| <i>P. cinnamomi</i> | 508435 | MN135945 |
| <i>P. cinnamomi</i> | TARI 94006 | GU111594 |
| <i>P. cinnamomi</i> | AR-1 | MH777152 |
| <i>P. cinnamomi</i> | 133 | KP070662 |
| <i>P. cinnamomi</i> | 1873 | KU961906 |
| <i>P. cinnamomi</i> | CMW33386 | GU799635 |
| <i>P. cryptogea</i> | Carrai7II | KP070719 |
| <i>P. cryptogea</i> | 9 | MG712918 |
| <i>P. cryptogea</i> | PC01 | MH401205 |
| <i>P. cryptogea</i> | 1072 | EU200283 |
| <i>P. palmivora</i> | CPR22 | KU308392 |
| <i>P. palmivora</i> | PPG8 | KY475630 |
| <i>P. palmivora</i> | Pp43-Wera-leaf | KP183963 |
| <i>P. palmivora</i> | 176PC | HQ237479 |

141

142 Pathogenicity tests

143 Pathogenicity of four isolates of *P. cinnamomi* and one isolate each of *P. cryptogea* and *P.*
144 *palmivora* was tested by stem inoculation on avocado seedlings. Five one-year-old avocado
145 seedlings cv. Topa Topa for each isolate were inoculated with 4-mm agar plugs from five-day-old
146 cultures grown on V8A. Five seedlings were inoculated with sterile agar plugs as controls. All
147 inoculations were sealed with moist, autoclaved cotton wool and wrapped in aluminum foil to
148 prevent drying. Plants were kept in a greenhouse at 18–20±1°C and watered as required. Four
149 weeks after inoculation, the outer bark was removed and canker lesions measured. Symptomatic
150 tissue was excised from the margin of the canker and plated onto PARP to re-isolate the pathogen.
151 Canker lengths on stems were subjected to analysis of variance (ANOVA), based on a completely
152 randomized design, and means separated using the Tukey Test.

153

154

155

3 Results

156 *Isolation and identification*

157 Nineteen *Phytophthora* isolates were obtained from necrotic root tissues of 11 symptomatic trees
 158 and 8 soil samples. All but 4 orchards surveyed were found to be infested with *Phytophthora* spp.
 159 (82.6%). Based on cultural, morphological and molecular characteristics, isolates were identified as
 160 *P. cinnamomi* (17 isolates), *P. cryptogea* (1) and *P. palmivora* (1) (Table 2; Fig. 4). *Armillaria*
 161 *gallica* was isolated from the single symptomatic tree in an orchard in Anamur district of Mersin
 162 province, which had a mycelial sheath under the bark of the lower stem; *P. cinnamomi* was isolated
 163 from the soil in this same orchard. ITS (MT229426) and EF 1- α (MT239054) sequences of the *A.*
 164 *gallica* isolate, and r DNA ITS sequences of 6 *Phytophthora* spp. isolates (MH219917–MH219918,
 165 and MN833031–MN833034) were loaded into GenBank. The phylogenetic tree obtained using ITS
 166 sequences of *Phytophthora* spp. obtained in the present study and sequences deposited in GenBank
 167 is shown in Figure 5.

168

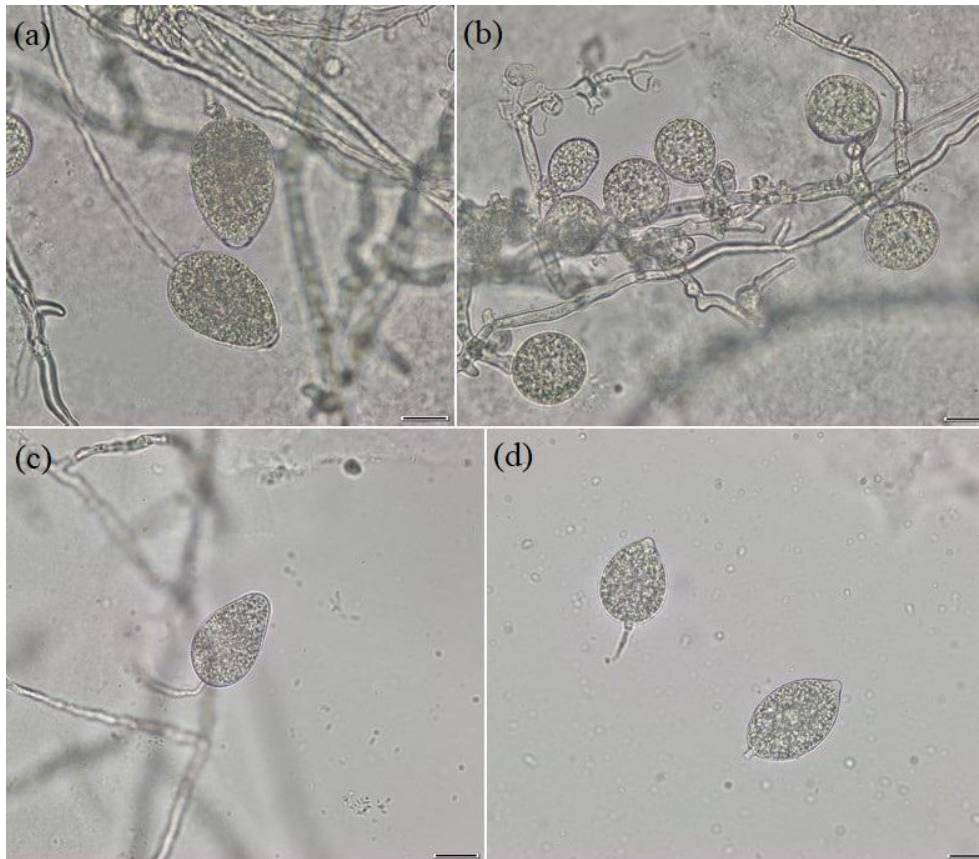
Table 2. Morphological and cultural characteristics of *Phytophthora* isolates obtained from avocado orchards in Turkey.

| | <i>P. cinnamomi</i> | <i>P. cryptogea</i> | <i>P. palmivora</i> |
|------------------------------------|---|--|---------------------------------|
| Sexuality | Heterothallic | Heterothallic | Heterothallic |
| Sporangia | | | |
| Shape | Mostly ovoid, rarely ellipsoid and obpyriform | Usually ovoid and obpyriform, rarely ellipsoid | Mostly ovoid, rarely obpyriform |
| Papillae | No | No | Yes |
| Total range (μ m) | 44.3–82.5 x 32.4–45.7 | 42.8–62.0 x 28.3–38.7 | 38.8–57.9 x 28.5–42.4 |
| Length/breadth mean (μ m) | 52.3 x 36.6 | 50.9 x 32.4 | 48.1 x 33.5 |
| Length/breadth ratio | 1.43 | 1.57 | 1.44 |
| Internal proliferation and nesting | No | Yes | No |
| Caducity | Non-caducous | Non-caducous | Caducous with short pedicel |
| Chlamydospore | Yes | No | Yes |
| Maximum temperature growth | <35°C | <35°C | <35°C |
| Colony pattern | Rosette on PDA | Petaloid on CMA and MEA | No |

169

170 The most commonly isolated species was *P. cinnamomi*, found in 17 orchards, whereas *P.*
 171 *cryptogea* and *P. palmivora* were obtained from two other orchards. *Armillaria gallica* was detected
 172 in only one tree. *Phytophthora palmivora* was isolated only from soil, and *P. cryptogea* only from
 173 roots, while *P. cinnamomi* was isolated from both necrotic root and stem tissues, and from soil
 174 samples.

175

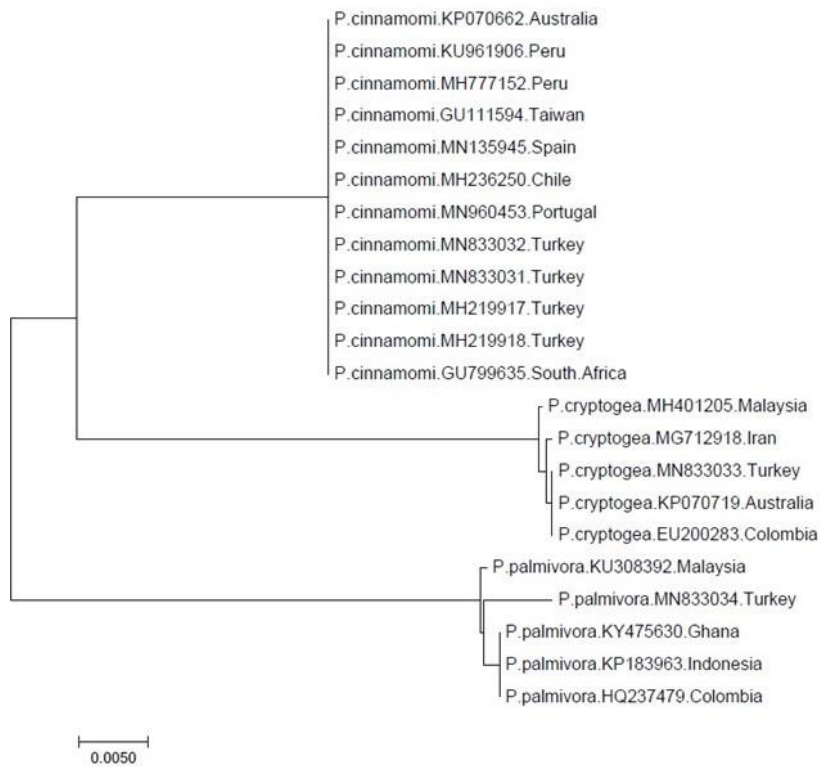


176

177

178

Figure 4. Sporangia (a) and chlamydospores (b) of *Phytophthora cinnamomi*; sporangia of *Phytophthora cryptogea* (c) and *Phytophthora palmivora* (d).



179

180

181

182

Figure 5. Dendrogram showing genetic relatedness of *P. cinnamomi* (4 isolates), *P. cryptogea* (1 isolate) and *P. palmivora* (1 isolate) isolated from roots of avocado planted in the south of Turkey, compared against sequences of eight isolates of *P. cinnamomi*, four isolates of *P. cryptogea* and four isolates of *P. palmivora* from GenBank (Table 1).

183 *Pathogenicity test*

184 Lesions between 1.4 – 5.4 cm in length appeared on stems inoculated with *Phytophthora* spp.
 185 (Table 3). In contrast, collenchyma callus of healthy appearance was produced over wound points
 186 over the same time period on control plants (Fig. 6). The inoculated *Phytophthora* spp. were re-
 187 isolated from symptomatic tissues. Lesion lengths were found to be significantly different ($P<0.05$)
 188 (Table 3). Data showed that *P. cryptogea* and three isolates of *P. cinnamomi* were in the same
 189 group, whereas *P. palmivora* and one isolate of *P. cinnamomi* were in a separate group.

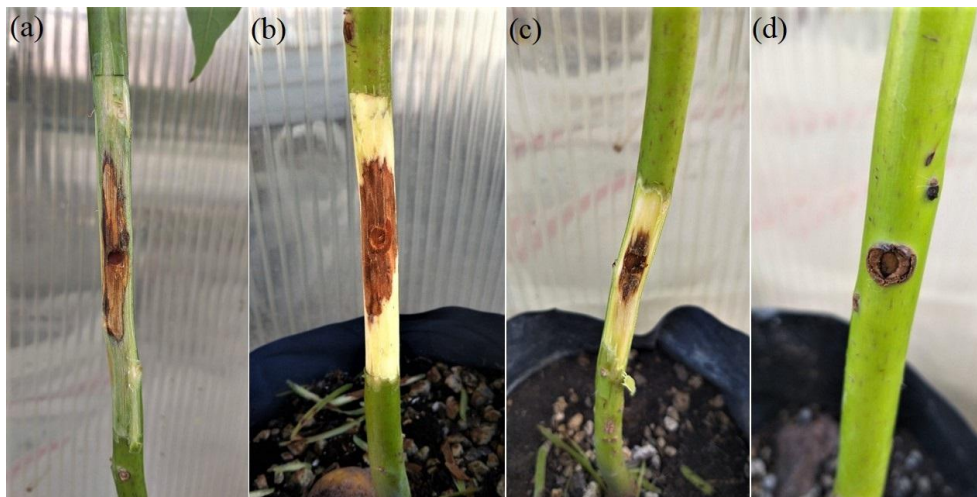
190 Table 3. Stem canker length averages of seedlings inoculated with six isolates of *Phytophthora* spp. obtained from avocado orchards in Turkey

| Isolate | GenBank accession | Min-max necrosis length (cm) | Necrosis length averages (cm) |
|---------------------|-------------------|------------------------------|-------------------------------|
| <i>P. cryptogea</i> | MN833033 | 3.5 – 5.4 | 4.32 A* |
| <i>P. cinnamomi</i> | MN833031 | 2.2 – 4.5 | 3.74 AB |
| <i>P. cinnamomi</i> | MH219918 | 2.5 – 4.1 | 3.42 ABC |
| <i>P. cinnamomi</i> | MH219917 | 2.2 – 4.5 | 3.32 ABC |
| <i>P. cinnamomi</i> | MN833032 | 1.7 – 3.4 | 2.62 BC |
| <i>P. palmivora</i> | MN833034 | 1.4 – 2.5 | 2.14 C |

CV (%)=0.24; $R^2=0.52$

*Means followed by the same letter are not statistically different according to Tukey test ($P\leq 0.05$)

191
192
193
194



195
196 Figure 6. Stem inoculation test: lesions caused on avocado plants by *Phytophthora cryptogea* (a), *P. cinnamomi* (b), *P. palmivora* (c) and no lesion on
197 control (d).
198

199 **4 Discussion**

200 This paper presents the first report of *P. cinnamomi*, *P. cryptogea* and *P. palmivora* causing
 201 avocado root rot in Turkey. It is a widespread disease in southern Turkey, such that at least one of
 202 the *Phytophthora* spp. was isolated from all but four of the avocado orchards surveyed. It is not
 203 surprising that *P. cinnamomi* was the most frequently isolated species. It is known that *P.*
 204 *cinnamomi* is the most destructive and widely distributed pathogen of avocados (Zentmyer, 1980;
 205 Coffey, 1992; Erwin & Ribeiro, 1996; Ploetz et al., 2002; Rodriguez-Padron et al., 2018). The fact
 206 that *P. cinnamomi* is the main species responsible for one the most severe plant disease epidemics

207 known, dieback of jarrah (*Eucalyptus marginata*) dominated forests in Western Australia clear
208 demonstrate the exceptionally destructive nature of this pathogen (Podger, 1972; Shea et al., 1983;
209 Erwin & Ribeiro, 1996).

210 Although *Phytophthora cinnamomi* was first found in Papua New Guinea (ref?), it is considered
211 highly adaptable and certainly causes major problems in warm temperate climates (Crandall et al.,
212 1945; Zentmyer, 1980; CABI, 1991; Balci et al., 2007). Extensive work in American oak forests
213 occurring above the 40°N latitude failed to find *P. cinnamomi* in soils (Balci et al., 2007). Cold
214 winter temperatures are unfavorable climatic condition its survival (Zentmyer, 1980; Brasier &
215 Scott, 1994; Balci & Halmschlager, 2003). Similarly, *P. cinnamomi* was obtained from various
216 Turkish coastal regions with temperate climates. For instance, it was isolated from rhizosphere soil
217 in oak forests (Balci & Halmschlager, 2003), and from soil around the roots of symptomatic
218 chestnut trees (Akillı et al., 2012, 2019) in temperate regions of Turkey. In addition, *P. cinnamomi*
219 caused root rot of walnut (Kurbetli, 2013) and *Protea* spp. (Tok & Avcı, 2015) in subtropical parts
220 of Turkey.

221 This work showed that, in addition to *P. cinnamomi*, other *Phytophthora* species such as *P.*
222 *cryptogea* and *P. palmivora*, are also present in avocado orchards in Turkey although these species
223 appear to be less widespread. *Phytophthora cryptogea*, one of the first *Phytophthora* species
224 identified, has a very wide range of hosts (Erwin and Ribeiro, 1996), is reported for the first time
225 associated with avocado in this work, although it was previously reported in orchards of several
226 fruits, including apple, kiwifruit and sweet cherry in Turkey (Kurbetli & Değirmenci, 2011;
227 Kurbetli & Ozan, 2013; Kurbetli, 2014). It may be widespread in fruit orchards in Turkey, but it
228 was found in only one avocado orchard in this study.

229 The *Phytophthora* isolates obtained from avocados here produced extending lesions on stems of
230 avocado seedlings, but it was surprising that the stem cankers caused by *P. cinnamomi* did not
231 progress as expected. Lesion lengths on plants inoculated with *P. cinnamomi* were not significantly
232 different from those caused by *P. cryptogea*. Some *Phytophthora* species, such as *P. quercina*, were
233 not effective in causing stem lesions of oak in pathogenicity tests (Balci and Halmschlager, 2003;
234 Bianco et al. 2003). Similarly, *Phytophthora megasperma* isolated from kiwifruit, almond and sour
235 cherry failed to cause cankers in stem inoculations on the same hosts (Kurbetli & Ozan, 2013;
236 Kurbetli et al., 2016, 2017). These species were highly pathogenic, however, when inoculated onto
237 root systems of oak, almond and sour cherry plants (Jung et al., 1999; Kurbetli et al., 2016, 2017).
238 Had root inoculations been carried out in the present work, it is possible that *P. cinnamomi* would
239 have proved to be much more aggressive than *P. cryptogea* or *P. palmivora* on avocado.

240 *Phytophthora palmivora* was the least pathogenic species in this work although Rodriguez-Padron
241 et al. (2018) reported that it showed high pathogenicity in stem inoculations on avocado.

242 It was interesting that the fan-shaped mycelium observed on the lower stem of a symptomatic tree
243 in an orchard infested with *P. cinnamomi* was of *Armillaria gallica* identified by both ITS and EF-
244 1 α sequences. Compared to rDNA sequences, the EF-1 α gene appears to better resolve closely
245 related *Armillaria* species, such as *A. gallica* and other related species, and this gene presents a
246 valuable diagnostic tool for the genus (Maphosa et al., 2006; Hasegawa et al., 2010; Klopfenstein et
247 al., 2017; Heinzelmann et al., 2019). *Armillaria gallica* behaves as a saprotroph or rarely as an
248 opportunistic pathogen (Tsykun et al., 2012; Heinzelmann et al., 2019). It is known that certain
249 symptoms, such as necrotic collar lesions at the stem base of ash (*Fraxinus* spp.) caused by
250 *Hymenoscyphus fraxineus* may be secondarily invaded by *Armillaria* species, which can further
251 reduce host health and accelerate decline (Husson et al., 2012; Chandelier et al., 2016; Marçais et
252 al., 2016; Enderle et al., 2017).

253 This study has increased knowledge of *Phytophthora* species associated with avocado crops, with
254 the first report of *P. cryptogea* from avocado, and the isolation of the previously described avocado
255 pathogens *P. cinnamomi* and *P. palmivora*. However, further investigation is required to clarify the
256 involvement of *Armillaria* and *Phytophthora* species other than *P. cinnamomi* in avocado decline in
257 Turkey.

258

259 **Acknowledgements**

260

261 This research was supported by Bati Akdeniz Agricultural Research Institute (BATEM), and we appreciate the help of
262 the staff of the Ministry of Agriculture and Forestry. We are also thankful to Dr Emrah Yıldırım for preparation of the
263 map.

264

265

265 **References**

- 266 Akilli, S., Serçe, Ç. U., Katircioğlu, Y. Z., & Maden, S. (2012). Involvement of *Phytophthora* spp. in chestnut decline
267 in the Black Sea region of Turkey. *Forest Pathology* 42, 377–386.
- 268 Akilli, S., Katircioğlu, Y. Z., Serçe, Ç. U., Çakar, D., Rigling, D., & Maden, S. (2019). *Phytophthora* species associated
269 with dieback of sweet chestnut in Western Turkey. *Forest Pathology* 49, e12533.
- 270 Balci, Y., & Halmschlager, E. (2003). *Phytophthora* species in oak ecosystems in Turkey and their association with
271 declining oak trees. *Plant Pathology* 52, 694–702.
- 272 Balci, Y., Balci, S., MacDonald, W. L., Juzwik, J., Long, R. P., & Gottschalk, K. W. (2007). *Phytophthora* spp.
273 associated with forest soils in eastern and north-central U.S. oak ecosystems. *Plant Disease* 91, 705–710.
- 274 Bianco, M. C., Di Brisco, D., Luisi, N., & Lerario, P. (2003). Pathogenicity of *Phytophthora* species on *Quercus*
275 seedlings. In: McComb JA, Hardy GE, Tommerup I, eds. *Phytophthora* in forests and natural ecosystems.

276 *Proceedings of the Second International Meeting of IUFRO Working Party 7.02.09*, 2001, Albany, Western
277 Australia. Perth, Australia: Murdoch University.

278 Brasier, C. M., & Scott, J.K. (1994). European oak declines and global warming: a theoretical assessment with special
279 reference to the activity of *Phytophthora cinnamomi*. *OEPP/EPPO Bulletin* 24, 221–32.

280 CABI, (1991). *Phytophthora cinnamomi*. *Distribution of the Maps of Plant Diseases*, No:302, CAB International,
281 Wallingford, UK, 2 pp.

282 Chandelier, A., Gerarts, F., San Martin, G., Herman, M., & Delahaye, L. (2016). Temporal evolution of collar lesions
283 associated with ash dieback and the occurrence of *Armillaria* in Belgian forests. *Forest Pathology* 46, 289–297.

284 Coffey, M. D. (1992). *Phytophthora* root rot of avocado, p. 423–444. In: Kumar, J., H.S. Chabe, U.S. Singh, and A.N.
285 Mukhopadhyay (eds.). *Plant Diseases of International Importance, Vol. III: Diseases of Fruit Crops*. Prentice Hall,
286 Englewood Cliffs, USA.

287 Crandall, B. S., Gravatt, G. F., & Ryan, M. M. (1945). Root disease of *Castanea* species and some coniferous and
288 broadleaf nursery stocks, caused by *Phytophthora cinnamomi*. *Phytopathology* 35, 162–180.

289 Enderle, R., Sander, F., & Metzler, B. (2017). Temporal development of collar necroses and butt rot in association with
290 ash dieback. *iForest* 10, 529–536.

291 Erwin, D. C. & Ribeiro, O. K. (1996). *Phytophthora Diseases Worldwide*, 2nd edn. St. Paul, MN, USA, APS Press.

292 Hasegawa, E., Ota, Y., Hattori, T., & Kikuchi, T. (2010). Sequence-based identification of Japanese *Armillaria* species
293 using the elongation factor-1 alpha gene. *Mycologia* 102, 898–910.

294 Heinzemann, R., Dutech, C., Tsykun, T., Labbé, F., Soularue, J. P., & Prospero, S. (2019). Latest advances and future
295 perspectives in *Armillaria* research. *Canadian Journal of Plant Pathology* 41, 1–23.

296 Henao, E. R. Arana, A. C., Valencia, A. L. E., & Florez, J. E. M. (2017). Evaluation of tolerance to *Phytophthora*
297 *cinnamomi* Rands in avocado (*Persea americana* Miller.) germplasm. *Acta Agronomyca* 66, 128–134.

298 FAOSTAT, (2019). <http://www.fao.org/faostat/en/#data/QC> (18.10.2019).

299 Husson, C., Cael, O., Grandjean, J. P., Nageleisen, L. M., & Marçais, B. (2012). Occurrence of *Hymenoscyphus*
300 *pseudoalbidus* on infected ash logs. *Plant Pathology* 61, 889–895.

301 Jung, T., Cooke, D. E. L., Blaschke, H., Duncan, J. M., & Oßwald, W. (1999). *Phytophthora quercina* sp. nov., causing
302 root rot of European oaks. *Mycological Research* 103, 785–798.

303 Klopfenstein, N. B., Stewart, J. E., Ota, Y., Hanna, J. W., Richardson, B. A., Ross-Davis, A.L., ... Kim, M. S. (2017).
304 Insights into the phylogeny of Northern Hemisphere *Armillaria*: Neighbor-net and Bayesian analyses of
305 translation elongation factor 1- α gene sequences. *Mycologia* 109, 75–91.

306 Kumar, S., Stecher, G., & Tamura, K. (2016). MEGA7: Molecular Evolutionary Genetics Analysis version 7.0 for
307 bigger datasets. *Molecular Biology and Evolution* 33, 1870–1874.

308 Kurbetli, İ., & Değirmenci, K. (2011). *Phytophthora* spp. determined in almond and apple orchards in Turkey.
309 *Proceedings of the Fourth Plant Protection Congress of Turkey*, 87. 28–30 June 2011, Kahramanmaraş, Turkey.

310 Kurbetli, İ. (2013). *Phytophthora cinnamomi* associated with root and crown rot of walnut in Turkey. *Journal of*
311 *Phytopathology* 161, 287–289.

312 Kurbetli, İ. & Ozan, S. (2013). Occurrence of *Phytophthora* root and stem rot of kiwifruit in Turkey. *Journal of*
313 *Phytopathology* 161, 887–889.

314 Kurbetli, İ. (2014). Involvement of *Phytophthora cryptogea* in sweet cherry decline in Turkey. *Phytoparasitica* 42,
315 627–630.

- 316 Kurbetli, İ., Yılmaz, A., Değirmenci, K., & Demirci, F. (2016): Almond decline caused by *Phytophthora megasperma*
317 in southeastern Anatolian region of Turkey. *The Journal of Turkish Phytopathology* 45, 13–20.
- 318 Kurbetli, İ., Aydoğdu, M., & Sülü, G. (2017). *Phytophthora chlamydospora* and *P. megasperma* associated with root
319 and crown rot of sour cherry in Turkey. *Journal of Plant Diseases and Protection* 124, 403–406.
- 320 Maphosa, L., Wingfield, B. D., Coetzee, M. P. A., Mwenje, E., & Wingfield, M. J. (2006). Phylogenetic relationships
321 among *Armillaria* species inferred from partial elongation factor 1-alpha DNA sequence data. *Australasian Plant*
322 *Pathology* 35, 513–520.
- 323 Marçais, B., Husson, C., Godart, L., & Caël, O. (2016). Influence of site and stand factors on *Hymenoscyphus*
324 *fraxineus*-induced basal lesions. *Plant Pathology* 65, 1452–1461.
- 325 Menge, J. A., Douhan, G. W., McKee, B., Pond, E., Bender, G. S., & Faber, B. (2012). Three new avocado rootstock
326 cultivars tolerant to *Phytophthora* Root Rot: ‘Zentmyer’, ‘Uzi’, and ‘Steddom’. *American Society for*
327 *Horticultural Science* 47, 1191–1194.
- 328 Milne, D. L., & Chamberlain, J. (1971). Experimental control of avocado *Phytophthora* root rot in South Africa.
329 *California Avocado Society Yearbook* 55, 144–147.
- 330 Pegg, K. G., Whiley, A. W., Langton, P. W., & Saranah, J. B. (1987). Comparison of phosetyl-al, phosphorous acid and
331 metalaxyl for the long-term control of *Phytophthora* root rot of avocado. *Australian Journal of Experimental*
332 *Agriculture* 27, 471–474.
- 333 Perez-Jimenez, R. M., Zea-Bonilla, T., & Lopez-Herrera, C.J. (2005). *Phytophthora* root rots in Andalusia: A review.
334 *South African Avocado Growers’ Association Yearbook* 28, 10–13.
- 335 Perez-Jimenez, R. M. (2008). Significant avocado diseases caused by fungi and oomycetes. *The European Journal of*
336 *Plant Science and Biotechnology* 2, 1–24.
- 337 Ploetz, R., Schnell, R. J., & Haynes, J. (2002). Variable response of open-pollinated seedling progeny of avocado to
338 *Phytophthora* root rot. *Phytoparasitica* 30, 262–268.
- 339 Podger, F. D. (1972). *Phytophthora cinnamomi*, a cause of lethal disease in indigenous plant communities in Western
340 Australia. *Phytopathology* 62, 972–981.
- 341 Robin, C., Smith, I., & Hansen, E. M. (2012). *Phytophthora cinnamomi*. *Forest Phytophthoras* 2.
- 342 Rodriguez-Padron, C., Siverio, F., Perez-Sierra, A., & Rodriguez, A. (2018). Isolation and pathogenicity of
343 *Phytophthora* species and *Phytophthium vexans* recovered from avocado orchards in the Canary Islands,
344 including *Phytophthora niederhauserii* as a new pathogen of avocado. *Phytopathologia Mediterranea* 57,
345 89–106.
- 346 Shea, S. R., Shearer, B. L., Tippet, J. T., & Deegan, P.M. (1983). Distribution, reproduction, and movement of
347 *Phytophthora cinnamomi* on sites highly conducive to jarrah dieback in south Western Australia. *Plant Disease*
348 67, 970–973.
- 349 Tok, F. M., & Avcı, F. (2015). First report of *Phytophthora* root rot caused by *Phytophthora cinnamomi* on
350 commercially cultivated proteas in Turkey. *Plant Disease* 99, 1181.
- 351 Tsykun, T., Rigling, D., Nikolaychuk, V., & Prospero, S. (2012). Diversity and ecology of *Armillaria* species in virgin
352 forests in the Ukrainian Carpathians. *Mycological Progress* 11, 403–414.
- 353 Zentmyer, G. A. (1980). *Phytophthora cinnamomi* and the diseases it causes. Monograph no: 10. The American
354 Phytopathological Society. St. Paul, Minnesota. USA.
- 355
- 356