

Contribution to the identification of *Cercospora* species in Iranکمک به شناسایی گونه‌های *Cercospora* در ایران*

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Abstract

Eight species of the genus *Cercospora* from different localities in Northern provinces of Iran were obtained and examined during spring-autumn 2010. *Cercospora acnidae* (from *Amaranthus chlorostachys* var. *chlorostachys*), *C. beticola* (from *Beta vulgaris*), *C. iridis* (from *Iris* sp.), *C. lactucae-sativae* (from *Lactuca sativa*), *C. mercurialis* (from *Mercurialis annua*), *C. sorghi* (from *Sorghum halepense*) and *C. zonata* (from *Vicia faba*) were identified. Furthermore, *C. apii* is identified from *Abutilon theophrasti*, *Euphorbia heterophylla*, *Solanum lycopersicum*, *Pelargonium zonale*, *Vigna sinensis* and *Zantheschia aethiopica*. Among these, *C. acnidae*, *C. apii*, *C. iridis*, and *C. mercurialis* are new records and *Sorghum halepense* is new host for *C. sorghi* in Iran.

Keywords: New record, fungi, host plant, leaf spot

چکیده

هشت گونه از جنس *Cercospora* از نقاط مختلف استان‌های شمالی ایران جمع‌آوری شدند و طی بهار تا پاییز سال ۱۳۸۹ مورد مطالعه قرار گرفتند. *Cercospora acnidae*, *C. lactucae-sativae*, *C. iridis*, *C. beticola*, *C. mercurialis*, *C. sorghi* و *C. zonata* به ترتیب از میزبان‌های *Amaranthus chlorostachys* var. *chlorostachys*، *Lactuca sativa*، *Iris* sp.، *Beta vulgaris*، *Mercurialis annua* و *Sorghum halepense* شناسایی شدند. علاوه بر این، *Cercospora apii* از میزبان‌های *Abutilon theophrasti*، *Euphorbia heterophylla*، *Solanum lycopersicum*، *Pelargonium zonale*، *Vigna sinensis* و *Zantheschia aethiopica* شناسایی شد. از بین این گونه‌ها، *C. acnidae*، *C. apii*، *C. iridis* و *C. mercurialis* برای اولین بار از ایران گزارش می‌شوند و *Sorghum halepense* میزبان جدیدی برای گونه *C. sorghi* در ایران می‌باشد.

واژه‌های کلیدی: رکورد جدید، قارچ، گیاه میزبان، لکه برگی

Introduction

Species of the hyphomycetous genus *Cercospora* Fresen. are associated with leaf spot diseases on a wide range of host plants (Ellis 1976, Crous & Braun 2003). Many *Cercospora* species produce a phytotoxic metabolite called cercosporin which was initially thought to be produced only by true *Cercospora* species (Fajola 1978), and those species not producing this toxin belong to other morphologically similar genera. Chupp (1954) in a monograph of the genus *Cercospora* listed over 1800 species names. In a major taxonomic treatment of the genus, Deighton (1967–79) segregated and reclassified many *Cercospora* species into several genera, including *Cercosporiella*, *Cercosporidium*, *Paracercospora*, *Pseudocercospora*, *Pseudocercosporiella* and *Pseudocercosporidium*. *Cercospora* species are characterized by having acicular, hyaline and septate conidia with a conspicuous hilum produced on pigmented, unbranched, septate and smooth conidiophores (Braun 1995, 1998). Crous & Braun (2003) published an annotated check-list of *Cercospora* and *Passalora* Fr. with 5720 taxa and taxonomic re-allocation of numerous species. Recently Braun & Crous (2007) described several new species, new combinations and new names after re-examination of type collections of *Cercospora* species and other related genera. Iranian records of *Cercospora* species have only been poorly studied. Ershad (1990, 2000, 2002) reported four *Cercospora* species from Iran. Scharif & Ershad (1966) presented a list of fungi on various host plants where also a few *Cercospora* species are named. Ershad (2009) listed 59 including 14 uncertain species of the genus *Cercospora* in his book 'Fungi of Iran'. Taxonomy of the genus *Cercospora* and other similar fungi have changed in recent decades, new species, combinations, names and nomenclatural clarifications have been introduced, therefore, taxonomic revision of this genus in Iran seems to be necessary.

Materials and Methods

Specimens with leaf spot symptoms from different localities in the north of Iran, including Guilan,

Mazandaran and Golestan provinces were collected during spring-autumn 2010. Some specimens were also provided by others. Potato Dextrose Agar (PDA) and Malt-extract Agar (MA) were used in order to grow the fungi. Conidia and stromata were streaked onto the media directly using a sharp sterilized needle under a binocular. No growth was occurred on these media after a month. Microscopic slides were prepared from stromata, conidiophores and conidia in 25% lactic acid. Characters such as, presence or absence of stromata and their development, pigmentation of conidia, conidiophores, conidiogenous loci (scars) and conidiogenous cells were used to identify the species. Drawings were made using a drawing tube attached to an Olympus BH-2 microscope.

Results and Discussion

Eight foliicolous *Cercospora* species, four of which being new records to Iran, were identified. From the species *Cercospora beticola*, *C. lactucae-sativae*, *C. sorghi* and *C. zonata* are reported from Iran (see Ershad 2009), therefore, these species are not illustrated here and only their host and locality are mentioned. All collected specimens are deposited in the fungus reference collection of the Ministry of Jihad-e-Agriculture (IRAN) at the Iranian Research Institute of Plant Protection, Tehran.

1. *Cercospora acnidae* Ellis & Everh., Proc. Acad. Nat. Sci. Philadelphia 43: 89 (1891)

Leaf spots circular to subcircular, numerous, brown to blackish-brown with grey centre and red margin, 2–7 mm in diameter, sometimes coalescing into large area; caespituli amphigenous, mostly epiphyllous, punctiform; stromata small to fairly prominent, brown, 20–35 µm wide; conidiophores in small fascicles, 5–10 stalks, arising from stromata, brown, paler and attenuated towards the tip, multiseptate, erect, geniculate in the upper part, not branched, smooth, thin, 60–110 × 3–5 µm; conidiogenous cells integrated, terminal and intercalary, 20–45 µm in length; conidial scars

conspicuous, thickened and darkened, terminal and lateral, 1.5–2 μm wide; conidia formed singly, hyaline, acicular, straight to slightly curved, smooth, thin, multiseptate, 5–11 septa, base truncate, tip acute to subacute, 45–100 \times 2–3.5 μm ; hilum thickened and darkened, 1–2.5 μm wide (Fig. 1).

Specimen examined: On *Amaranthus chlorostachys* Willd. var. *chlorostachys*, Golestan province, Shastkola forest, 8 Nov. 2010, coll.: M. Pirnia & R. Zare (IRAN 15015 F).

Chupp (1954) introduced *C. brachiata* on *Amaranthus* spp. in his monograph that is morphologically similar to *C. apii*, therefore, Crous & Braun (2003) merged the species into *C. apii* and provided another species *Cercospora acnidae* on *Amaranthus* that was originally isolated from *Acnida cannabina* and *A. commons* (*Amaranthaceae*). Morphology of the specimen examined fit well with *Cercospora acnidae* provided by Chupp (1954).

2. *Cercospora apii* Fresen., Beitr. Mycol. 3: 91 (1863)

Leaf spots subcircular to irregular, at first yellowish-brown, later dark-brown, margin indefinite or

surrounded by brown or dark reddish-brown border, scattered on leaf surface, mostly 5–12 mm in diameter; stromata lacking to well-developed, composed of swollen hyphal cells, subglobose to irregular, brown to olivaceous-brown, 10–50 μm wide; caespituli amphigenous, mostly hypophyllous, punctiform; conidiophores solitary or in small to moderately large fascicles, loose to dense, mostly arising from stromata, erect, straight, subcylindrical or basal part cylindrical and upper fertile part slightly to strongly geniculate-sinuuous, medium dark-brown to pale-brown, paler near the apex, unbranched, smooth, thin-walled, septate, 20–250 \times 4–7 μm ; conidiogenous cells integrated, terminal or intercalary, occasionally unilocal, determinate, but usually multilocal, sympodial, 10–70 μm in length; conidiogenous loci conspicuous, thickened and darkened, terminal and lateral, sometimes inconspicuous; conidia solitary, short conidia often cylindrical or obclavate-cylindrical, developed long conidia acicular, hyaline to olivaceous, straight to curved, smooth, thin-walled, multiseptate, 3–20 transverse septa, base truncate, tip acute to subacute, 40–250 \times 2–5 μm ; hilum thickened and darkened, 2–4 μm wide (Fig. 2).

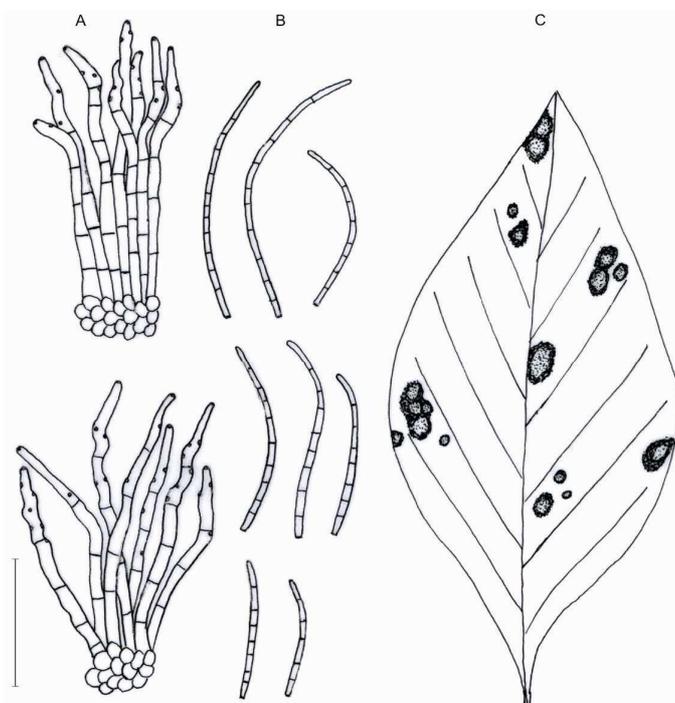


Fig. 1. *Cercospora acnidae* on *Amaranthus chlorostachys* var. *Chlorostachys*: A. Conidiophores, B. Conidia, C. Symptoms on leaf (Bar = 50 μm).

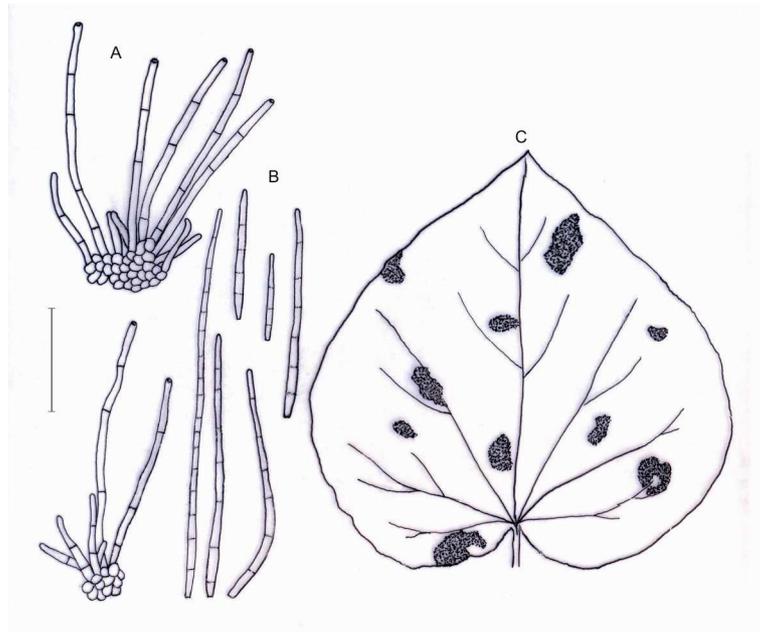


Fig. 2. *Cercospora apii* on *Abutilon theophrasti*: A. Conidiophores, B. Conidia, C. Symptoms on leaf (Bar = 50 μ m).

Among *Cercospora* species, *C. apii* is the oldest available name comprising of a large complex of morphologically indistinguishable taxa. Chupp (1954) stated that species of *Cercospora* are generally host-specific and described a large number of species based on host species. Crous & Braun (2003) believed that some *Cercospora* isolates on various host plants are morphologically indistinguishable and introduced *C. apii* s. lat. and linked 83 host genera to *C. apii*. Groenewald et al. (2006) revealed that morphology, host specificity and geographical location are not suitable characters in order to resolve the *C. apii* species complex. In this study, *C. apii* studied from six new host plants in Iran.

Specimens examined: On *Abutilon theophrasti* Medic., Golestan province, Shastkola forest, 8 Nov. 2010, coll.: M. Pirnia & R. Zare (IRAN 15016 F); on *Euphorbia heterophylla* L., Golestan province, Gorgan, 10 Nov. 2010, coll.: M. Pirnia & R. Zare (IRAN 15018 F); on *Solanum lycopersicum* Mill., Guilan province, Astaneh Ashrafieh, 24 June 2010, coll.: M. Pirnia (IRAN 15019 F); on *Pelargonium zonale* Medic., Guilan province, Sume'e-Sara, 25 June 2010, coll.: M. Pirnia (15020 F); on *Vigna sinensis* (L.) Endl., Guilan province, Astaneh Ashrafieh, 24 June 2010, coll.: M. Pirnia (IRAN 14808 F); Guilan province, Sume'e-Sara, 25 June 2010,

coll.: M. Pirnia (15021 F); Mazandaran province, Babol, 12 Sept. 2010, coll.: M.A. Aghajani (IRAN 15022 F); on *Zantheschia aethiopica* (L.) Spreng., Guilan province, Some'e-Sara, 15 July 2007, coll.: A. Khodaparast (IRAN 14807 F).

3. *Cercospora iridis* Chupp, A monograph of the fungus genus *Cercospora*: 260 (1954)

Leaf spots irregular, elongated, centre white, margin surrounded by brown or dark reddish-brown border, 10–30 mm in diameter; stromata present, subglobose to irregular, brown, 35–45 μ m wide; caespituli hypophyllous, punctiform; conidiophores in small fascicles, arising from stromata, erect, cylindrical, upper part geniculate to sinuous, brown to pale-brown, paler and sometimes narrower towards the tip, unbranched, smooth, thin-walled, 0–1 septate, 10–50 \times 3–5 μ m; conidiogenous cells integrated, terminal, 7–15 μ m in length; conidiogenous loci inconspicuous to slightly conspicuous; conidia solitary, obclavate-cylindrical, olivaceous brown, straight to slightly curved, smooth, thin-walled, 1–5 transverse septa, base truncate, tip rounded to obtuse, 15–40 \times 7–10 μ m; hilum darkened, 2–3 μ m wide (Fig. 3).

Specimen examined: On *Iris* sp. Guilan province, Bandar Anzali, 25 June 2010, coll.: M. Pirnia (IRAN 15025 F).

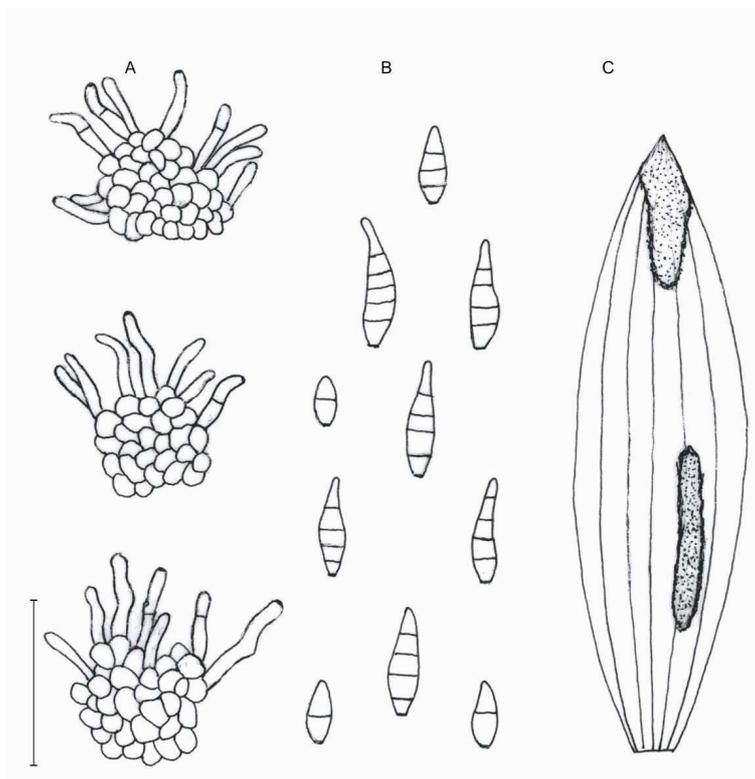


Fig. 3. *Cercospora iridis* on *Iris* sp.: A. Conidiophores, B. Conidia, C. Symptoms on leaf (Bar = 50 μ m).

Cercospora iridis has characteristic short conidiophores and obclavate-cylindrical conidia (Crous & Braun 2003). Chupp (1954) described this species with acicular and hyaline conidia, but specimen examined in this study has obclavate-cylindrical and olivaceous brown conidia similar to that reported by Crous & Braun (2003).

4. *Cercospora mercurialis* Pass., Mycoth. Univ., No. 783 (1877)

Leaf spots circular to irregular, pale-brown with a brown line border, yellowish grey centre, 2–8 mm in diameter; stromata present, small, substomatal, brown, 20–35 μ m wide; caespituli amphigenous, mostly hypophyllous, punctiform; conidiophores in small fascicles, 3–8 stalks, arising from stromata, through stomata, pale-brown to brown, erect, geniculate to sinuous, not branched, rarely dichotomously branched in the upper part, smooth, thin, irregular in width, narrower

towards the tip, aseptate or sparingly septate, (12–)15–67 \times 2.5–5 μ m; conidiogenous cells integrated, terminal, sometimes conidiophores reduced to conidiogenous cells, (12–)15–63 μ m in length; conidial scars conspicuous, thickened and darkened, terminal and lateral, 1–2 μ m wide; conidia formed singly, hyaline, cylindrical or rarely acicular, straight to slightly curved, smooth, thin, indistinctly multiseptate, base subtruncate to obconically truncate, tip mostly obtuse, 47–100 \times 2.5–5 μ m; hilum thickened and darkened, 1–2 μ m wide (Fig. 4).

Specimen examined: On *Mercurialis annua* L., Golestan province, Gorgan, 11 May 2010, coll.: M. Pirnia (IRAN 14809 F).

Morphology of the specimen examined agrees with the description provided by Chupp (1954). The species is distinguished from *C. apii* by moderately short and dichotomously branched conidiophores. This is the first report of this species from Iran.

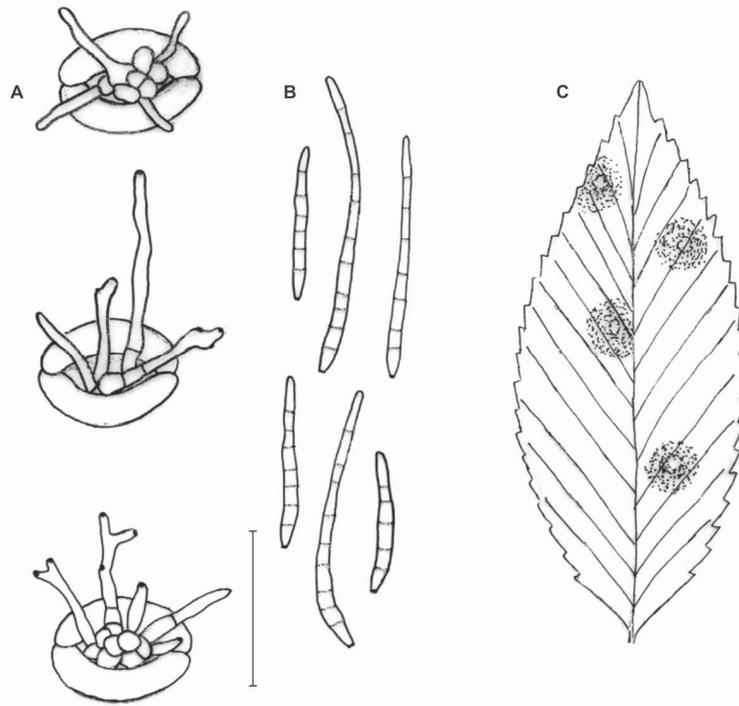


Fig. 4. *Cercospora mercurialis* on *Mercurialis annua*: A. Conidiophores, B. Conidia, C. Symptoms on leaf (Bar = 50 μ m).

5. *Cercospora beticola* Sacc., Nuovo Giorn. Bot. Ital. 8: 189 (1876)

Specimens examined: On *Beta vulgaris* L., Golestan province, Gorgan, 10 May 2010, coll.: M. Pirnia (IRAN 15023 F); Mazandaran province, Behshahr, 9 May 2010, coll.: M. Pirnia (IRAN 15024 F); Mazandaran province, Babol, 12 Sep. 2010, coll.: M.A. Aghajani (IRAN 15017 F).

Chupp (1954) characterized *C. beticola* mainly by moderately short conidiophores with almost hyaline tips and with one to several mild geniculations near the tip. Because of similar morphological characteristics, Crous & Braun (2003) redispersed *C. beticola* as a synonym of *C. apii* s. lato. However, Groenewald *et al.* (2006) showed that, *C. apii* and *C. beticola* s. stricto form two well-defined clades and, therefore, are probably two distinct species. Their data was obtained from a wide range of host plants. Our results from Iranian specimens classified under *C. beticola* shows that they have moderately short conidiophores, obclavate-cylindrical conidia identical to the description provided by Chupp (1954) for *C. beticola*. *Cercospora beticola* has

previously been reported from various localities in Iran during 1946–1973 (Ershad 2009).

6. *Cercospora lactucae-sativae* Sawada, Rep. Gov. Agric. Res. Inst. Taiwan 35: 111 (1928)

Specimen examined: On *Lactuca sativa* L., Golestan province, Gorgan, 11 Nov. 2010, coll.: M. Pirnia & R. Zare (IRAN 15026 F).

Cercospora lactucae-sativae was originally published as a synonym of *Cercospora longissima* by Chupp (1954). Crous & Braun (2003) introduced *C. lactucae-sativae* on *Lactuca* species and corrected *C. longissima* as a synonym of *C. lactucae-sativae*. Collections initially examined in Iran were published as *C. longissima*. Recently, Ershad (2009) has changed the name *C. longissima* to *C. lactucae-sativae* in his book 'Fungi of Iran'.

7. *Cercospora sorghi* Ellis & Everh., J. Mycol. 3: 15 (1887)

Specimen examined: On *Sorghum halepense* (L.) Pers., Guilan province, Astaneh Ashrafieh, 24 June 2010, coll.: M. Pirnia (IRAN 15027 F)

Morphology of the specimen examined agrees with the description of *C. sorghi* given by Chupp (1954). The species has already been reported from *Sorghum bicolor* (L.) Moench in Iran (Ershad 2009) and this is its first report from *Sorghum halepense*.

8. *Cercospora zonata* G. Winter, *Hedwigia* 23: 191 (1884)

Specimens examined: On *Vicia faba* L., Mazandaran province, Sari, 9 May 2010, coll.: M. Pirnia (IRAN 15028 F); Golestan province, Gorgan, 12 May 2010, coll.: M. Pirnia (IRAN 15029 F)

Morphology of the specimens examined agrees with the description provided by Chupp (1954) and Ellis (1976). Collections were already examined in Iran published as *C. fabae* by Scharif & Ershad (1966) and as *C. zonata* by Hedjaroude (1976) without any description or illustration. Ershad (2009) has also listed *C. zonata* on *Vicia faba*.

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