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Research article

A Study on the identification of powdery mildew fungi (Erysiphaceae) in Ardabil landscape, Iran

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Abstract: The Erysiphaceae are obligatory parasitic fungi that cause powdery mildew disease of green space plants. During this study, powdery mildew fungi were collected and identified from different localities of Ardabil landscape, Iran. *Erysiphe rayssiae* (on *Spartium junceum*), *Erysiphe robiniae* var. *robiniae* (on *Robinia pseudoacacia*) and *Euoidium* cf. *agerati* (on *Ageratum houstonianum*) are new records for mycobiota of Iran. *Jasminum* sp. is reported as Matrix nova for *Erysiphe syringae-japonicae*. Also this is the first record of *Golovinomyces montagnei* on *Cirsium arvense* and *Golovinomyces orontii* on *Antirrhinum majus* in Iran. The taxa including *Erysiphe astragali* on *Astragalus* sp., *Erysiphe crucifearum* on *Brassica elongata*, *Erysiphe polygoni* on *Rumex* sp., *Golovinomyces cichoracearum* on *Cichorium intybus* and *Golovinomyces sordidus* on *Plantago* sp. are newly found in Ardabil province. Furthermore, some specimens belonging to *Podosphaera fusca* s.l. were assessed. According to the new species concept, concerning these taxa, *Podosphaera fusca* s.s. was redescribed, and *Podosphaera erigerontis-canadensis* on *Taraxacum* sp. is reported for the first time, although previously reported from Iran under the *Podosphaera fusca*.

Keywords: taxonomy, Erysiphales, powdery mildew, new record, new host

Introduction

Worldwide, Erysiphales is by far the largest powdery mildew order (Talgo *et al*., 2011). Powdery mildews are destructive fungi on wild and cultivated plants aerial parts such as leaves, shoots and stems and cannot be cultured on artificial media. Both anamorphic and teleomorphic states may be observed on infected parts, the characteristics of all structures are important in taxonomic studies (To-anun *et al*., 2005). The development of the powdery mildew anamorphs taxonomy proceeded largely independently of the teleomorphs (Braun and Cook, 2012). The first systematic trial to identify powdery mildew conidial states at species level was made by Ferraris (1912) who grouped species of *Oidium* according to the size and shape of their conidia and created a key to its species (To-anun *et al.*, 2005). Records of powdery mildews on new host plants, often as anamorphic states, are not rare (Ale-Agha *et al.*, 2008). Valuable information on phylogeny and classification of these fungi have been published in recent years (Saenz and Taylor, 1999; Mori *et al*., 2000; Takamatsu *et al*., 1998, 1999, 2000 and 2008; Takamatsu, 2004; Matsuda and Takamatsu, 2003; Khodaparast *et al.*, 2003, 2005; Cunnington *et al*., 2010; Hirata *et al*. 2000). The classification of powdery mildew is based on the location and type of the mycelium

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and the characteristics of the asci, ascospores and cleistothecial appendages produced by their perfect states (Boesewinkel 1980). The fruiting bodies of the powdery mildews are more or less spherical to somewhat flattened, closed (non-ostiolate) ascomata, which have been classified as chasmothecia (Braun and Cook 2012). Pirnia *et al*. (2005, 2006, 2007) showed that morphology of penicillate cells in various species of some powdery mildews can be used as useful character for species delimitation. The knowledge of the species of powdery mildews in a particular area is important concerning the biology and taxonomy of these fungi and for phytopathological purposes (Khodaparast and Abbasi, 2009). The knowledge on the Erysiphales fungi on Ardabil green space plants isscant. Therefore, present research was aimed to investigate the powdery mildew species in Ardabil landscape.

Materials and Methods

Samples were collected from summer to autumn 2012 and transferred to the laboratory of Plant Pathology, University of Mohaghegh Ardabili. Host plants were identified by specialists. For microscopic analysis, different organs of the fungus were prepared in lactic acid 50% and were examined by light microscopy (ZEISS AXIOLAB). Morphological characteristics of sexual and asexual stages, kind of host and other information related to each species were investigated. Images were provided by digital camera (Sony, DSH-HX) attached onto an Olympus BH2 microscope. The images were put together and were edited using Photoshop (Adobe Photoshop CS). Exact identification and confirmation of taxa were done using Braun (1987, 1995), Braun and Takamatsu (2000), Cook and Braun (2009) and Braun and Cook (2012). All collected specimens are deposited in the fungal collection of the Department of Plant Protection, College of Agriculture, University of Mohaghegh Ardabili. Reference numbers are presented in parentheses after collector´s name.

Results and Discussion

The list of identified species from Ardabil landscape alphabetically is as follow: *Erysiphe astragali* on *Astragalus* sp., *Erysiphe crucifearum* on *Brassica elongata*, *Erysiphe polygoni* on *Rumex* sp., *Erysiphe rayssiae* on *Spartium junceum*, *Erysiphe robiniae* var. *robiniae* on *Robinia pseudoacacia*, *Erysiphe syringae-japonicae* on *Jasminum* sp., *Euoidium* cf. *agerati* on *Ageratum houstonianum*, *Golovinomyces cichoracearum* on *Cichorium intybus*, *Golovinomyces montagnei* on *Cirsium arvense*, *Golovinomyces orontii* on *Antirrhinum majus*, *Golovinomyces sordidus* on *Plantago* sp. and *Podosphaera erigerontis-canadensis* on *Taraxacum* sp. Among these, *Erysiphe rayssiae* and *Erysiphe robiniae* var. *robiniae* and *Euoidium* cf. *agerati* were new records for mycobiota of Iran. Other species were newly found in Ardabil province and reported previously from Iran (see Khodaparast and Abbasi, 2009), therefore, these species are not illustrated here and only their host and locality are mentioned. Furthermore, new host plants were identified for some previously reported taxa.

***Erysiphe rayssiae* (Mayor) U. Braun & S. Takam., Schlechtendalia, 4: 13, 2000**

Material examined: On leaves of *Spartium junceum* (Fabaceae), Iran, Ardabil, 9. Sep. 2012, K. Sharifi, (FCUMA1001).

Mycelium amphigenous, white, almost persistent, hyphae 4.8-7.2 µm wide; hyphal appressoria more or less lobed; conidiophores arising from upper surface of mother cell, erect, 45-80 µm long, foot-cells straight to slightly sinuous, about 27.5-50 × 4.8-7.2 µm, usually followed by 1-2 shorter cells, conidia formed singly; ellipsoid-ovoid to cylindrical-doliform, 25-37.5 × 12.5-17.5 µm (Fig. 1).

Leveilulla taurica has been reported previously on *S. junceum* in Iran (Amano 1980). *Erysiphe rayssiae* differs from two close species (*Erysiphe pisi* var *pisi* and *Erysiphe trifoliorum*) by dimensions of conidiophores and foot-cells (Braun and Cook, 2012). This is the first report of *Erysiphe rayssiae* from Iran.

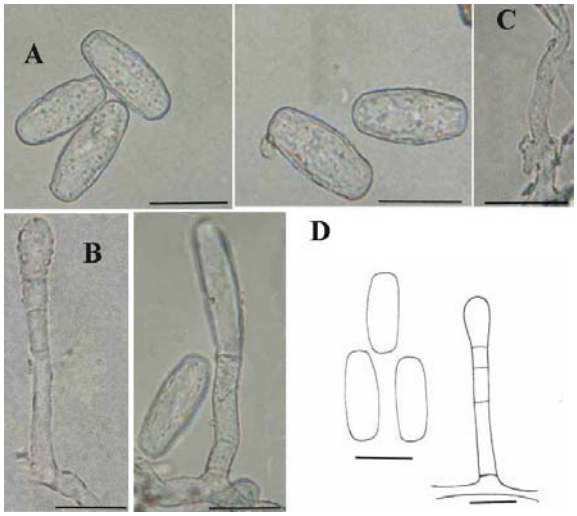

Figure 1 *Erysiphe rayssiae*, A: Conidia; B: Conidiophore; C: Appressorium; D: Drawing of conidia and conidiophore, scale bar = 20µm.

***Erysiphe syringae-japonicae* (U. Braun) U. Braun & S. Takam., Schlechtendalia, 4: 14, 2000**

Material examined: on leaves of *Jasminum* sp. (Oleaceae), Iran, Ardabil, 20. Sep. 2012, K. Sharifi, (FCUMA1002)

Mycelium amphigenous, effuse or in patches, evanescent to almost persistent, chasmothecia scattered to gregarious, mostly 75-115 µm diam, appendages equatorial, stiff, straight to somewhat curved, usually 0.75-1.25 times as long as the chasmothecial diam, apices 4-6 times tightly and regularly dichotomously branched strictly in two dimensions, occasionally deeply forked, tips recurved; asci 3-10, broadly ellipsoid-obovoid, 35-57.6 × 33.6-48 µm, sessile or short-stalked, 5-8 spored; ascospores ellipsoid-ovoid, 15-23 × 7.5-14 µm, colorless (Fig. 2).

According to Braun and Cook (2012), *Erysiphe syringae-japonicae* is distinguished from *Erysiphe syringae* by having appendages with more pigmented, mostly thick-walled and asci with more ascospores. Furthermore, *Erysiphe ligustri* differs from *E. syringae-japonicae* in forming three-dimensionally branched chasmothecial appendages, although these are genetically very close to each other (Seko *et al*. 2008). Previously, *Oidium jasmini* was reported on *Jasminum* sp. form Iran (Pirnia 2013). *Erysiphe syringae japonicae* on *Jasminum* sp. was recorded as new hosts for the world.

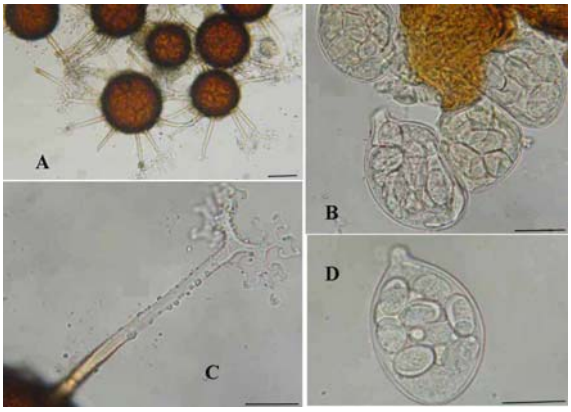

Figure 2 *Erysiphe syringae japonicae*, A: chasmothecia; B: Crushed chasmothecium with asci; C: Appendages; D: Ascus, scale bars A = 50 µm, B, C and D = 20 µm.

***Erysiphe robiniae* var. *robiniae* Grev., F1. edin.: 460, 1824**

Material examined: on leaves of *Robinia pseudoacacia* (Fabaceae), Iran, Ardabil, 17. Oct. 2012, K. Sharifi, (FCUMA1003).

Mycelium amphigenous, effuse or in patches, often covering the entire surface of the leaves, persistent or evanescent, hyphae 4-6µm wide; conidiophores arising from the upper surface of the mother cell, erect, 45-87.5 µm long, foot cell cylindrical, straight to slightly sinuous, 30-50.5 × 8.2-10 µm, followed by 1-2 shorter cells; conidia formed singly, ellipsoid (cylindrical) or doliform, about 27.5-45 × 15-18 µm; chasmothecia scattered to almost gregarious, depressed globose, 70-130 µm diam; peridium cells irregularly polygonal, 12-23 µm diam; appendages numerous, in the lower half, usually not turning upward, sinuous, but not irregularly shaped, 54.6-730 µm, 5-9 µm wide, 1-6 septate, apices mostly simple, dichotomously branched, loose, forked widely, tips straight; asci 4-9, ellipsoid-obovoid, saccate-clavate, 54.6-75 × 31.2-33.8 µm, sessile or short-stalked, 4-6 spored; ascospores

ellipsoid-ovoid, 18.2-25 × 10.4-15.5 µm, colorless (Fig. 3).

Braun (1987) listed name of various species as synonyms of *E. trifolii*, e.g. *Erysiphe robiniae* (now *E. trifoliorum*) (Braun *et al*., 2010). Although *Erysiphe robiniae* is morphologically very close to *E. trifoliorum*, but separated by having distinctly thickened chasmothecial appendages and irregularly shaped peridium cells (Braun and Cook 2012). This is the first report of *Erysiphe robiniae* var. *robiniae* from Iran.

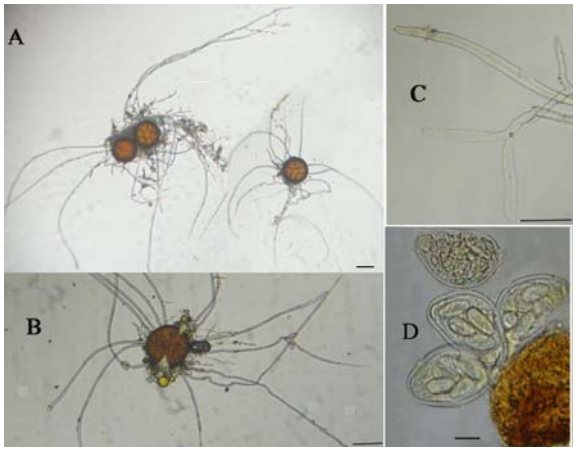

Figure 3 *Erysiphe robiniae* var. *robiniae*, A: chasmothecia; B: chasmothecia with ascus; C:Appendages; D: Asci, scale bars A and B = 50 µm, C and D = 20 µm.

***Euoidium* cf. *agerati* (J. M. Yen) U. Braun and R. T. A. Cook**

Material examined: on stems and leaves of *Ageratum houstonianum* (Asteraceae), Iran, Ardabil, 22. Aug. 2012, K. Sharifi, (FCUMA1004)

Mycelium on stem and leaves, amphigenous, effuse or in irregular patches, covering the entire leaf surface, evanescent to usually persistent; hyphae branched, septate, hyaline, thin walled, about 4.8-7.2 µm wide, hyphal appressoria nipple-shaped, with somewhat crenulated surface or slightly to moderately lobed; conidiophores erect, arising from superficial hyphae, on top of the mother cells, 65-155 × 9.6-12 µm, foot-cells straight, cylindrical, about 35-60 × 9.6-12 µm, followed by 1-2 shorter cells; conidia in chains, ellipsoid-ovoid to doliform-cylindrical, without fibrosin bodies, 26-37.5 × 12-23 µm, germ tubes arising from one end, short to moderately long, simple, tips unlobed, with club-shaped appressorium (Fig. 4).

The conidia in this anamorph in chains and without any fibrosin bodies, thus this species is a *Golovinomyces* anamorph (Bappamal *et al*., 1995). Anamorph names are used where teleomorphs have not been found. Often *Euoidium agerati* can represent anamorph of *Golovinomyces circumfusus*, although this often has curved foot-cells (Braun and Cook, 2012). This is the first report of *Euoidium* cf. *agerati* from Iran.

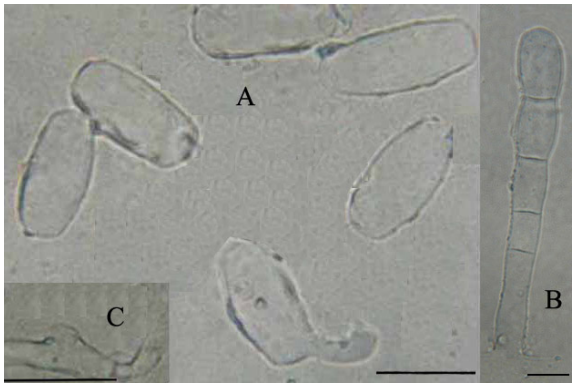

Figure 4 *Euoidium* cf. *agerati*, A: Conidia with germinated conidia; B: Conidiophore; C: Appressorium, scale bars = 20 µm

***Golovinomyces montagnei* U. Braun (2012)**

Material examined: on leaves of *Cirsium arvense* (Asteraceae), Iran, Ardabil, 16. Oct. 2012, K. Sharifi, (FCUMA1005)

Mycelium on stem and leaves, amphigenous, sometimes also caulicolous, effuse or in irregular patches, sometimes covering the entire leaf surface, evanescent to usually persistent; hyphae thin walled, about 4.8-7.2 µm wide, hyphal appressoria nipple-shaped, solitary, sometimes in opposite pairs, occasionally with somewhat crenulated surface, 3-6 µm diam; conidiophores erect, arising from upper surface to somewhat laterally from the

hyphal mother cell, 65-155 × 9.6-12 µm, foot-cells straight, cylindrical, about 35-80 × 9.6-12 µm, followed by 1-3 shorter cells, basal septum usually at the junction with the mother cell, sometimes raised up to 10µm; conidia in chains, ellipsoid-ovoid to doliform-limoniform, 25-40 × 15-22.8 µm, germ tubes arising fromone end, short to moderately long, mostly clavate, apex with somewhat swollen apperssorium (Fig. 5).

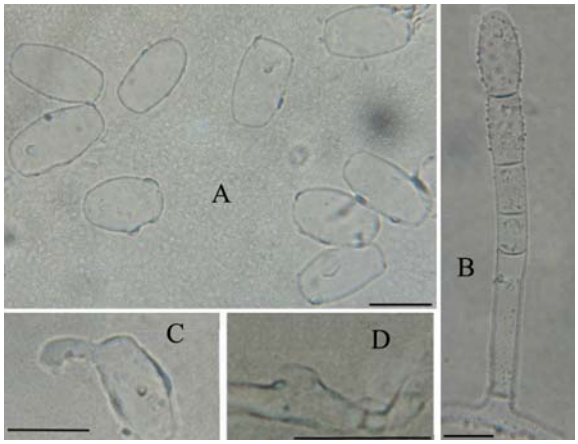

Figure 5 *Golovinomyces montagnei*, A: Conidia; B: Conidiophores C: germinated conidium; D: Appressoria, scale bars = 20 µm.

According to the phylogenetic relationships reconstruct among the *Golovinomyces* species, this species diverged with the phylogeny of host subfamily and tribes (Matsuda and Takamatsu, 2003). Based on the last revision about taxonomy of *Golovinomyces cichoracearum s.l.* (Braun and Cook, 2012), this species was divided into more species. *Golovinomyces montagnei* is introduced as the cause of powdery mildew on *Carduoideae* (Asteraceae). *Golovinomyces montagnei* is distinguished from *Golovinomyces cichoracearum* by its conidiophores with straight foot-cells, unlobed appressoria, short chasmothecial appendages and asci which are occasionally three-spored (Blumer, 1933, 1967). *Cirsium arvense* is identified as new host for *Golovinomyces montagnei* in Iran.

***Golovinomyces orontii* (Castagne) V. P. Heluta, Ukrainskiy Botanichnyi Zhurnal, 45 (5): 63, 1988**

Material examined: on leaves of *Antirrhinum majus* (*Scrophulariaceae*), Iran, Ardabil, 10. Aug. 2012, K. Sharifi, (FCUMA1006).

Mycelium amphigenous, effuse or in patches, evanescent or persistent, white, hyphae about 4.8-7.2 µm wide; conidiophores erect, arising laterally or from the upper surface of hyphal mother cells, and positioned almost centrally or towards one end of the cells, about 62.5-105 × 9.6-12 µm, foot-cells straight or often curved in the basal half, about 35-57.5 × 7.2-12 µm, followed by 1-4 shorter cells; conidia usually in short chains, ellipsoid- ovoid to doliform-subcylindrical, 25-35 × 12.5-17.5 µm, germ tubes arising from an end, occasionally from a side, usually fairly short, apically often with a somewhat swollen appressorium (Fig. 6).

Often *Golovinomyces orontii* has been confused with *Golovinomyces cichoracearum*. *G. orontii* differs from *G. cichoracearum* in host range, developing conidia in unswollen form and foot-cells of conidiophores that are usually curved at the base (Braun 1987, Matsuda and Takamatsu, 2003 and Braun and Cook, 2012).

Antirrhinum majus is identified as new host for *G. orontii* in Iran.

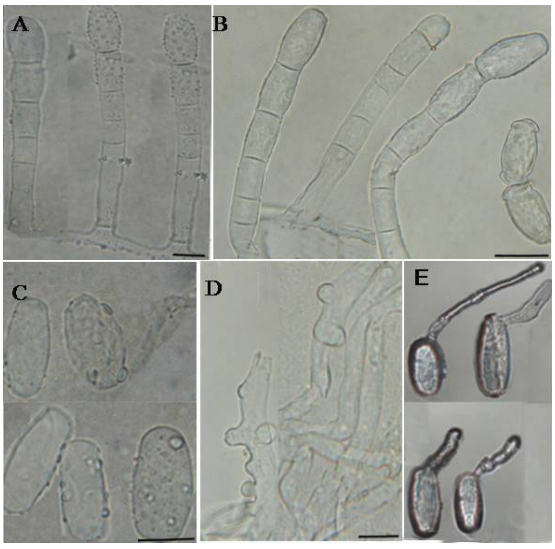

Figure 6 *Golovinomyces orontii*, A and B: Conidiophores; C: Conidia; D: Appressoria; E: Germinated conidia, scale bars = 20µm.

***Podosphaera erigerontis-canadensis* (Lev.) U. Braun & T. Z. Liu, in Liu, The *Erysiphaceae* of Inner Mongolia: 198, Chifeng 2010**

Material examined: on leaves of *Taraxacum* sp. (Asteraceae), Iran, Ardabil, 12. Aug. 2012, K. Sharifi, (FCUMA1007)

Mycelium on leaves, amphigenous, white, effused or in irregular patches, covering almost the entire leaf surface, white when young, old persistent mycelium turning brownish diffuse brown patches together with abundant chasmothecia, chasmothecia scattered to loosely aggregated, subglobose, 67.2-90 µm diam; peridium cells irregularly polygonal to irregular, 24-50 µm diam, appendages few, in the lower half of the ascomata, mycelioid, simple, 1-3.2 times as long as the chasmothecial diam; 4.8-10 µm wide, ascus broadly ellipsoid-ovoid, 52.8-79.2 × 45.6-60 µm, sessile or almost so, terminal oculus small, 11.25-15 µm diam; 6-8 spored (Fig. 7).

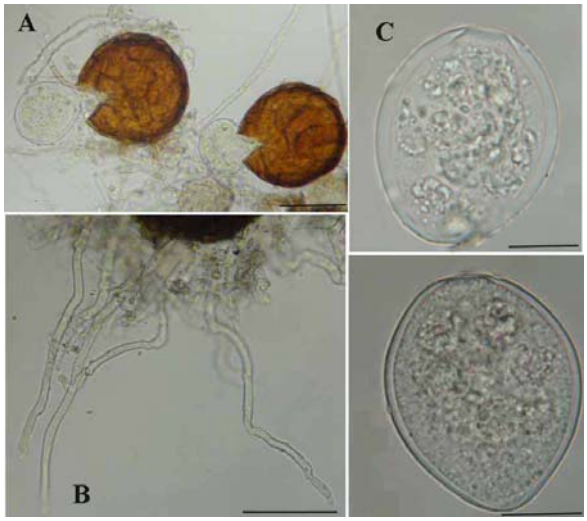

Figure 7 *Podosphaera erigerontis canadensis*, A: chasmothecia with ascus; B: Appendages; C: Ascus, scale bars A and B = 50 µm, C = 20 µm.

Based on reassessment on *Podosphaera fusca s.l.* and molecular sequence analyses by Ito and Takamatsu (2010), *Podosphaera erigerontis-canadensis* is separated from *P. fusca* by having small ascomata and asci with small terminal oculi (Braun and Cook 2012). *Podosphaera erigerontis-canadensis* is described for the first time from Iran, although this fungus has been reported frequently in the past as *Podosphaera fusca*.

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مطالعه قارچ‌های عامل سفیدک پودری (Erysiphaceae) فضای سبز اردبیل

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چکیده: قارچ‌های متعلق به تیره Erysiphaceae به‌عنوان انگل‌های اجباری، عامل بیماری سفیدک-پودری گیاهان از جمله گیاهان مورد استفاده در فضای‌سبز شهرها می‌باشند. طی این تحقیق، قارچ‌های سفیدک سطحی از مناطق مختلف فضای‌سبز شهر اردبیل جمع‌آوری و مورد شناسایی قرار گرفتند. گونه‌های *Erysiphe rayssiae* (روی *Spartium junceum*)، *Erysiphe robiniae* var. *robiniae* (روی *Robinia pseudoacacia*) و *Euoidium* cf. *agerati* (روی *Ageratum houstonianum*) برای فلور قارچی ایران جدید می‌باشند. همچنین *Jasminum* sp. به‌عنوان میزبان جدیدی (Matrix nova) برای *Erysiphe syringae-japonicae* شناخته شد. همچنین این اولین گزارش از *Golovinomyces montagnei* روی *Cirsium arvense* و *Golovinomyces orontii* روی *Antirrhinum majus* در ایران است. گونه‌های *Erysiphe astragali* روی *Astragalus* sp.، *Erysiphe crucifearum* روی *Brassica elongata*، *Erysiphe polygoni* روی *Rumex* sp.، *Golovinomyces cichoracearum* روی *Cichorium intybus* و *Golovinomyces sordidus* روی *Plantago* sp. برای استان اردبیل جدید می‌باشند. به‌علاوه، برخی گونه‌های متعلق‌به *Podosphaera fusca* s. l. مورد ارزیابی قرار گرفتند، با توجه به مفهوم جدید گونه در این مورد، *Podosphaera fusca* s. s. مجدداً توصیف شد و گونه *Podosphaera erigerontis-canadensis* روی *Taraxacum* sp. برای نخستین بار از ایران شرح داده شد، اگرچه این قارچ در گذشته با نام *Podosphaera fusca* گزارش شده بود.

واژگان کلیدی: تاکسونومی، Erysiphales، سفیدک‌های سطحی، رکورد جدید، میزبان جدید