First occurrence of *Golovinomyces bolayi* on *Lactuca tuberosa* in Iran

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**Abstract**

White powdery masses were observed on leaves of *Lactuca tuberosa* in southwestern Iran. Infected leaves were examined using a stereomicroscope and microscopic slides of fungal structures prepared in lactic acid 25%. Based on morphological characteristics of conidia, appressoria and foot cell of conidiophores, the fungus was identified as *Golovinomyces bolayi*. The fungus is an important pathogen causing powdery mildew disease and occurs on various *Lactuca* species. *G. bolayi* separated from *G. orontii* species complex and distinguished from other closely related species by having catenate conidia as well as nipple-shaped appressoria and sinuous-curved foot cell of conidiophores.

**Keywords** – Asteraceae – Erysiphales – Obligate parasite – Powdery mildew – Taxonomy

**Introduction**

Powdery mildew fungi are obligate parasites belonging to the Erysiphaceae (Ascomycota: Erysiphales), can infect more than 10,000 host plant species (Braun & Cook 2012). These fungi produce white powdery masses on the upper or lower surface of leaves, can be extended and cover a total surface of leaves (Braun 1987). Both anamorph (imperfect) and teleomorph (perfect) stages may be observed on infected plants. Some symptoms such as chlorosis or yellowing of leaves and decrease of growth can be followed by mycelial growth of powdery mildews (Braun 1987).

The genus *Lactuca* L. belonging to the plant family Asteraceae. According to Mozaffarian (2015), 12 species of the genus *Lactuca* are growing in different localities of Iran. Fungal pathogens belonging to the genera *Golovinomyces* (U. Braun) Heluta, *Podosphaera* Kunze, and *Leveillula* G. Arnaud. causing powdery mildew on various species of the genus *Lactuca*. (Braun 1987). The genus *Golovinomyces* was initially introduced by Heluta (1988) based on *Erysiphe* sect. *Golovinomyces* U. Braun. and characterized by ectophytic parasitism, polyascal ascomata and euoidium anamorph. The literature review indicated that taxonomy of the genus *Golovinomyces* had significantly been changed during the time (Braun et al. 2019).

Salmon (1900) was the first who introduced *Erysiphe cichoracearum* DC. (now as *Golovinomyces cichoracearum*) with broad host range in his monograph. *E. cichoracearum* is the most common causal agent of powdery mildew on Asteraceae (Braun 1987). In a taxonomic treatment, Blumer (1933) tried to segregate *E. cichoracearum* into several species and then he provided description of the species along with host list (Blu+mer 1967).
According to inoculation tests, Hammarlund (1945) showed plurivorous species within *E. cichoracearum sensu lato*. Hammett (1977) divided *Erysiphe cichoracearum sensu lato* into two groups – *E. cichoracearum sensu stricto* parasitic on taxa of *Asteraceae* and *E. cichoracearum sensu lato* parasitic on taxa of other plant families.

According to Matsuda & Takamatsu (2003), there is a close affinity between *Golovinomyces* and the host tribes of the *Asteraceae*. They suggested co-speciation between *Golovinomyces* species and their hosts and determined five significant groups in *Golovinomyces*, each of them is associated with a single host tribe of the *Asteraceae*. *Golovinomyces* on members of *Asteraceae* segregated into several species viz. *G. ambrosiae* (Schwein.) U. Braun & Cook RTA. on hosts belonging to tribe *Astereae* Cass., *G. macrocarpus* (Speer) U. Braun on hosts belonging to tribe *Anthemideae* Cass. and *G. montagnei* U. Braun on hosts belonging to tribe *Cardueae* Cass.

Braun & Cook (2012) divided *G. cichoracearum sensu lato* into more species on the basis of reassessment of morphological characteristics and molecular data, which is represented by a single host tribe of the *Asteraceae*. *G. cichoracearum sensu stricto* is now limited to host species of the genera *Scorzonera* L. and *Tragopogon* L. (Takamatsu et al. 2013).

Takamatsu et al. (2013) determined 11 lineages in the genus *Golovinomyces*, seven lineages include the *Asteraceae* as a host family and characterized by having hosts belonging to a single tribe of the *Asteraceae*. According to their phylogenetic analysis, powdery mildew on *Lactuca* spp. were included in *Golovinomyces orontii* group 3. This group had 19 host species of 10 families, indicating that the genus *Golovinomyces* expanded its host ranges to closely related plants in an early stage of evolution.

Lebeda & Mieslerová (2011) recognized large variability in virulence and existence of different races of *Golovinomyces cichoracearum*, concluded that interactions between various *Lactuca* species and pathogen are race-specific, following gene-for-gene hypothesis. Furthermore, Mieslerová et al. (2020) reported that disease incidence of *Golovinomyces bolayi* on populations of *Lactuca serriola*, varied between 29 and 75.2% and confirmed that *G. bolayi* isolates represented different combinations of reaction patterns on the *Lactuca* spp.

Braun et al. (2019) showed that *G. orontii sensu lato* which previously classified as groups one to three, split into three distinct main clusters. The third assemblage of sequences (group 3) comprises powdery mildews on hosts of the *Asteraceae* tribe *Cichorieae*, including *Cichorium* and *Lactuca* spp. and hosts belonging to various other plant families. Therefore, they proposed new species, *Golovinomyces bolayi* S. Takam., Lebeda & M. Götz.

Information of powdery mildew fungi in Iran are scattered in various regional reports (Pirnia et al. 2005, 2006, 2007, Pirnia 2014, Khodaparast et al. 2016, Arzanlou & Torbati 2016, Khodaparast et al. 2019). In a major attempt, Khodaparast & Abbasi (2009) provided comprehensive check-list of *Erysiphales* from Iran, which covers 90 species on 528 host plant species. In some taxonomic works, information of *Golovinomyces* species are involved (Khodaparast et al. 2001, Davari et al. 2014, Sharifi et al. 2013, 2014). Providing an update on powdery mildew diseases of *Lactuca* spp., was the main aim of this study.

**Materials & Methods**

Infected leaves of *Lactuca tuberosa* by anamorphic powdery mildew were collected from Fars Province, Iran, then dried between paper sheets and placed in the envelope. Features such as the location of mycelium and conidiophores on the leaves are examined using Stereomicroscope (Zeiss, Germany). Microscopic slides of fungal structures including conidiophores, foot-cells, conidia and appressoria prepared in lactic acid 25% without any staining and examined by means of standard light microscopy Olympus CH30, Japan (Pirnia 2014). Drawings were made using a drawing-tube attached to microscope.

Morphological characteristics of mycelium (colour, density), conidia (single or in chains, shape, size), conidiophores (shape, size), foot cell (shape, length, arrangement of the following cells on the foot cell) and appressoria (shape, diameter) were used to identify species (Braun et al. 2019).
The length and width of 30 conidia, foot cell and appressoria (×1000 magnification) were measured. Representative material is deposited as typically dried samples in the internationally recognized fungus reference collection of the Iranian Ministry of Agriculture “IRAN” at the Iranian Research Institute of Plant Protection under accession number IRAN 16776 F.

Results and discussion

On the basis of the combination of the production of conidia in chains, shape and dimension of conidia, structure and dimension of foot cell and nipple-shaped appressoria on the mycelia and host-plant association, the fungus was identified as *Golovinomyces bolayi* S. Takam., Lebeda & M. Götz. Only the anamorph stage of the fungus was found on the leaves.

Disease incidence of powdery mildew on *Lactuca tuberosa* is sporadic in Iran and only found in this study. Disease severity was estimated as a percentage of total leaf area of a single plant which was covered with powdery symptoms by giving maximum rating 1-5 scale as shown in Table 1 (Mulbrhan et al. 2016).

| Percentage of Disease | Rating scale |
|-----------------------|--------------|
| 1-10%                 | 1            |
| 11-25%                | 2            |
| 26-50%                | 3            |
| 51-75%                | 4            |
| >75%                  | 5            |

The formula cited in Mulbrhan et al. (2016), is used for calculating of disease severity.

\[ \text{Plant disease severity (PDS)} = \frac{\text{Number of individual ratings}}{\text{Number of plants assessed}} \times \frac{100}{\text{Maximum scale}} \]

In material examined, disease severity was estimated 40% (Percentage of disease: 11-25%, Rating scale: 2). *Golovinomyces bolayi* is common species on various *Lactuca* spp., foremost *Lactuca sativa* as edible vegetable. The powdery mildew can influence overall yield and yield quality of Lettuce which needs effective strategies for management of disease.

*Golovinomyces bolayi* S. Takam., Lebeda & M. Götz.

Mycelium amphigenous, forming white patches on the upper and lower surface of the leaves. Hyphal appressoria nipple-shaped, 3–5 μm diam. Conidiophores erect, solitary, straight. Foot cells cylindrical, sinuous-curved, 45–70 × 9–12 μm, followed by 2–3 (–4) cells that are shorter than the foot cell, 10–22 μm long, as wide as the foot-cells or somewhat wider. The basal septum of conidiophore adjacent to the mycelium. Conidia are forming in chains, without fibrosin bodies, ellipsoid-ovoid, doliform-subcylindrical, 25–34 × 14–18 μm. (Fig. 1).

Material examined – Iran, Fars Province, Noorabad, on living leaves of *Lactuca tuberosa* Jacq. (*Asteraceae*), 15 Jul. 2018, M. Pirnia (IRAN 16776 F).

Note – *Golovinomyces bolayi* has wide host range on hosts of tribe *Cichorieae, Asteraceae* (*Cichorium* spp. and *Lactuca* spp.) and some other plant families viz. *Bignoniaceae, Brassicaceae, Campanulaceae, Crassulaceae, Cucurbitaceae, Fabaceae, Lamiaceae, Linderniaceae, Plantaginaceae, Rosaceae* and *Solanaceae* (Braun et al. 2019). *Lactuca tuberosa* belongs to tribe *Cichorieae*. Hitherto no powdery mildew fungus was reported on *Lactuca tuberosa* in Iran and only one fungus, *Ramularia carletonii* (Ellis & Kellerm.) U. Braun was reported in this country (Pirnia & Braun 2018).
Fig. 1 – Golovinomyces bolayi. a Symptoms on leaves. b Conidiophores, catenate conidia, foot cells and basal septum. c Conidia. d Nipple-shaped appressoria. Scale bars = 25 µm.

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