Scientific Electronic Archives
Issue ID: Sci. Elec. Arch. Vol. 15 (8)
August 2022
DOI: http://dx.doi.org/10.36560/15820221589
Article link: https://sea.ufr.edu.br/SEA/article/view/1589

ISSN 2316-9281

First report of the phytopathogenic fungus *Phakopsora euvitis* Ono in leaves of *Plumeria rubra* L. (Apocynaceae)

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Abstract. *Phakopsora euvitis* is a phytopathological fungal species described in plants of *Vitis* sp. This study aimed to report the first case of fungal infestation of *P. euvitis* in the botanical genus *Plumeria*, *Plumeria rubra* in Rio Verde, Goiás, Brasil. Leaves showing *P. euvitis* urediniospore infestation were collected at Instituto Federal Goiano, Rio Verde, Goiás, Brasil, in the gardens of the central library. The plant and fungal species were identified by a specific dichotomous key for both genera. Slides were prepared with glycerin and micrographed under an optical microscope. As for the dimensions of *P. euvitis* urediniospores in this study, they presented length and width of 15.12 – 24.47 *x* 13.11 – 15.69 µm, sessile with slightly elongated peduncles, some abruptly elongated, ovoid, ovoid-ellipsoid and oblong-ellipsoid in only one hemisphere. Further studies should be carried out evaluating by histology the areas infected by *Phakopsora euvitis* in *Plumeria rubra* to understand the option of this fungus outside the genus Vitis and its evolution in this botanical species.
Keywords: *Plumeria* genus, *Phakopsora* genus, Urediniospores, Plant diseases.

Introduction

Several fungi are considered phytopathogens mainly in fruit and ornamental species (Evidente et al., 2019; Traversari et al., 2021). Among this exuberant fungal fauna, the genus *Phakopsora* stands out, infesting mainly vine orchards in the tropics, especially in Central and South America. *Phakopsora euvitis* Ono (2000) is popularly known as “vine rust or *ferrugem da videira*” because it prefers this group of vegetables, although it is not limited to this agricultural crop (Naruzawa et al., 2006).

P. euvitis is reported from Asia, Japan, Colombia, Venezuela, United States, Mexico, Australia and Brasil (Leu, 1988; Weinert et al., 2003; Tessmann et al., 2004). The complexity of the *Phakopsora* group according to Ono (2000), led *Phakopsora ampelopsidis* Diet. & Syd. the separation into three species according to their morphology and host specificity: *P. ampelosidis* parasite *Ampelosis* sp., *P. vitis sensu* P. Sydow presents development on *Parthenocissus* sp. and a new species, *P. euvitis* by Ono, infesting until then,

the genus *Vitis* spp. (Naruzawa et al., 2006; Menezes Filho et al., 2020).

Reports in Brasil of *P. euvitis* date back to 2001 on vines (*Vitis* spp.) in the municipality of Jandaia do Sul in the State of Paraná, by Tessmann et al. (2004), since then, this phytopathogenic fungus has been described in other Brazilian regions, including the states of Goiás, São Paulo, Rio Grande do Sul and Mato Grosso (Tessmann et al., 2004; Souza, 2004, Gomes et al., 2016; Menezes Filho et al., 2020).

The fungal *P. euvitis* causes the appearance of yellow pustules with urediniospores on the lower surface of the leaf blade and on the adaxial surface in the areas corresponding to the pustules, tissue necrosis is observed. Severe infection processes cause early defoliation, leading to physiological imbalance, promoting the loss of plant vigor. *P. euvitis* prefers to infect already developed leaves, although reports have observed it to a lesser extent in new shoots and rachis (Leu, 1988).

The plant diversity in the world presents a wealth of genera and species within numerous botanical families. Among these, we can mention the Apocynaceae family with 355 genera and around 3,700 species, within this rich diversity of genera we cite *Plumeria*, which is included in this family (Morales, 2005; Vettumperumal et al., 2018; Melo et al., 2021).

The genus *Plumeria* consists of large number of species distributed all around the world out of which 11 accepted species are growing in tropical and subtropical regions of the world (Bihani, 2021). Among this genus, we can mention several species used as ornamentals in parks and gardens, such as *Plumeria obtuse*, *P. alba* and *P. rubra*, the latter adapted to the different tropical and subtropical climates of Brasil. *P. rubra* is native to Mexico although it is also found in areas of the State of Hawaii in the United States.

P. rubra (Figure 1) has a semi-arboreal appearance, beautiful red flowers with a striking and sweet aroma, the latter being a characteristic that qualifies it as an excellent plant species for ornamental purposes. In addition to ornamental use, *P. rubra* has phytotherapeutic properties as initially described by a spanish priest, Francisco de Mendoza in 1522, however, it was Charles Plumier, a 17th century french botanist who named the species "*Plumeria*" (Idrees et al. al., 2019).

According to Bihani (2021), different parts of *P. rubra* are used to treat different conditions such as asthma (Patil et al., 2008), fever and skin irritation (Christophe, 2002) and also have numerous biological actions such as antifungal, antibacterial and antiviral (Jeven et al., 1979; Sticher, 1977; Vanden Berghe et al., 1978).

Vegetables have a "factory" of phytochemical compounds distributed in different classes such as flavonoids, tannins, saponins, quinones, coumarins, among others, which belong to the special metabolism used as means of defense, in pollination and against insects, herbivorous animals, viruses and even fungi (Suyal et al., 2021; Fernandez-Conradi et al., 2022).

Although plants are specialized in fighting various infections in their aerial and terrestrial organs, these phytopathogens negatively influence plant physiology, providing a rapid increase throughout the plant mainly in vital areas such as leaves that are involved in transpiration and gas exchange (Diagne et al., 2020; Domingos et al., 2021).

Fungi often have a preference for a certain plant group, however, these can infest and colonize plant tissues from other botanical groups.

The aim of this study was to describe the first record of *Phakopsora euvitis* Ono infesting leaves of *Plumeria rubra* L. in the State of Goiás, Brasil.

Figure 1. Individual of *Plumeria rubra* in flowering period, Rio Verde, Goiás State, Brasil. Source: Author, 2022.

Materials and Methods

Study location

The study was carried out at the Instituto Federal Goiano, Rio Verde, Goiás State, Brasil. Individuals of *P. rubra* are located in the garden of the central library of the geographically located educational institution (17°48'08.6''S and 50°54'18.4''W).

Collection and identification of plant species

The species of *P. rubra* was identified using a dichotomous key for the genus *Plumeria*. An exsiccate was herborized and deposited in the Herbarium of the Plant Systematics Laboratory, Department of Biological Sciences, Instituto Federal Goiano, Rio Verde, Goiás State, Brasil, with the following voucher (HRV 17778).

Fungal collection and identification

Leaf samples of *P. rubra* showing colonies of urediniospores were collected from live plants. The fungal sample was compared by visual and microscopic analysis with a standard sample deposited by Menezes Filho et al. (2020) with the following record (*P. euvitis* LQT ACPMF 01) in 2020 in the municipality of Rio Verde, Goiás State, Brasil.

A fungal sample for comparison was deposited in the author's mycological bank located in the duly documented Technological Chemistry laboratory.

Morphological characterization

The morphological characterization of the spores was performed as described by Menezes Filho et al. (2020) from counting 60 spores with the help of ImageJ software. To verify the viability of the spores, a solution of Toluidine Blue 1% (m/v) was used.

Microscopic observation was performed using an optical microscope at 4, 10 and 40x magnification. The dimensions of *P. euvitis* urediniospores were evaluated in terms of length and width expressed in (µm).

Results and discussion

Figure 2 shows *P. rubra* leaves infested with *P. euvitis* in the adaxial leaf portion. In the four individuals of *P. rubra*, infestation was observed only in the adaxial part and absence of necrosis in the abaxial part. It is suggested that this absence of necrosis is linked to the larger diameter of the leaf blade when compared to the leaf blade of *Vitis* spp.

Figure 2. Abaxial blade of *Plumeria rubra* infested with urediniospores colonies of *Phakopsora euvitis*, Rio Verde, Goiás State, Brazil.
Source: Author, 2022.

Figure 3 shows a micrograph of a urediniospore showing its equinulate-type ornamentation (Figure 4). Studies with P. euvitis in *Vitis* spp. show similarities in terms of ornamentation by Menezes Filho et al. (2020) in optical microscopy and Rasera et al. (2019) by scanning electron microscopy.

As for the dimensions of *P. euvitis* urediniospores in this study, they presented length and width of 15.12 – 24.47 *x* 13.11 – 15.69 µm, sessile with slightly elongated peduncles, some abruptly elongated, ovoid, ovoid-ellipsoid and oblong-ellipsoid in only one hemisphere. Studies by Gomes et al. (2016) and Menezes Filho et al. (2020) described findings of *P. euvitis* similar to this study, corroborating the microscopic morphology and identification key of the genus *Phakopsora*.

According to Xavier et al. (2012) and Gomes et al. (2016) urediniospores are produced in a disorganized way within the uredinia, where it is in this sense that we observe a great variety of forms in their morphology.

Although the aforementioned studies were carried out in a specific plant group, peculiarities were noted regarding its shape, as by Gomes et al. (2016) where they describe for *Vitis vinifera var. Isabel* average length and width of 14.6 – 23.3 *x* 11.5 – 15.4 µm with urediniospores yellowish and sessile with elongated peduncles in a single hemisphere. Xavier et al. (2012) verified dimensions

between 17.0 – 23.0 *x* 14.0 – 17.0 µm. Differences were also noted and described by Halfeld-Vieira et al. (2009) where they found uredia with cylindrical paraphyses, subepidermal and irrupting hypophiles, and urediniospores with dimensions between 24.0 – 26.0 *x* 15.0 – 18.0 µm and globoid shape. And Tessmann et al. (2004) with dimensions close to that of this study with 17.0 – 28.0 *x* 12.0 – 18.0 µm and morphology of ovoid, ovoid-ellipsoid and oblong-ellipsoid urediniospores.

Figure 4 shows some fungal spore ornamentation, as illustrated by Cummins and Hiratsuka (2003).

Figure 3. Micrograph of a urediniospore of *Phakopsora euvitis* in *Plumeria rubra*. Source: Author, 2022.

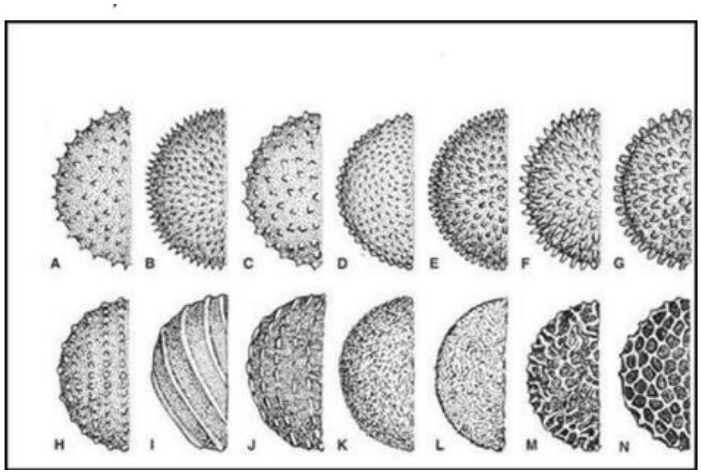

Figure 4. Fungal spore ornamentation. A-C: equinulate; D-G: warty; H: warty striatum; I: striped; J: dashed; K: rough; L: labyrinthine; M: pseudoreticulate; N: reticulate.
(Source: Cummins and Hiratsuka, 2003).

Conclusion

Phakopsora euvitis was initially described infesting plants of *Vitis* sp, however, as observed in this study, this fungus was described as the first report of infestation in the botanical genus of *Plumeria*, *Plumeria rubra* ornamental species in the world.

New studies should be carried out evaluating the biological control and the damage caused structurally in this *Plumeria* species.

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