BioInvasions Records (2020) Volume 9, Issue 1: 1–7

Rapid Communication

The first record of *Puccinia abrupta* var. *partheniicola*, on *Parthenium hysterophorus* an invasive alien plant species in Pakistan

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Citation: Iqbal IM, Ali K, Evans HC, Rehman A, Seier MK, Shabbir A, Weyl P (2020) The first record of *Puccinia abrupta* var. *partheniicola*, on *Parthenium hysterophorus* an invasive alien plant species in Pakistan. *BioInvasions Records* 9(1): 1–7, https://doi.org/10.3391/bir.2020.9.1.01

Received: 29 October 2019
Accepted: 13 December 2019
Published: 04 February 2020

Handling editor: Quentin Groom
Thematic editor: Stelios Katsanevakis

Abstract

Parthenium hysterophorus, commonly known as parthenium weed, is an invasive alien species in Pakistan. There are no specialized natural enemies native to Pakistan that attack the plant species inflicting serious damage and, thereby, keeping it under control. The rust species *Puccinia abrupta* var. *partheniicola,* commonly known as the winter rust, is closely associated with its host parthenium weed in its native range in the Americas. This study reports on the first record of the winter rust in Pakistan, with a likely pathway of introduction via India or Nepal. *Puccina abrupta* var. *partheniicola* was found to be widely present in several districts of the Punjab and Khyber Pakhtunkhwa Provinces. The sites where the most severe infestations of parthenium weed were observed were in the Lahore District of the Punjab Province. The climatic conditions prevalent in this region during the winter season 2018/2019 were favourable for the winter rust and caused an epiphytotic on parthenium weed. The presence of *P. abrupta* var. *partheniicola* marks the second natural enemy of *P. hysterophorus* recorded from Pakistan and is expected to aid the management of this invasive weed in the country.

Key words: winter rust, parthenium weed, natural enemy, fungal pathogen, invasive alien species

Introduction

Parthenium hysterophorus L. (Asteraceae), commonly known as parthenium weed, is a highly invasive plant outside its native range in the Americas causing severe damage to agriculture and ecosystems in more than 50 countries around the world (Shabbir et al. 2019). In Pakistan, *P. hysterophorus* was first reported in the Gujrat district of the Punjab Province in the 1980s and since then the weed has rapidly spread throughout the Province of Punjab, Islamabad Capital Territory (ICT) and parts of the Khyber Pukhtunkhwa (KP) Province (Shabbir et al. 2012) and has been reported from Sindh and Baluchistan Provinces. With its range still expanding,

parthenium weed is considered to be a dominant weed species of natural ecosystems (Iqbal 2015) and agriculture in Pakistan (Adkins and Navie 2006; Bajwa et al. 2019). Furthermore, this weed is known to be highly allergic to humans and toxic to livestock (Adkins and Shabbir 2014). Management options for *P. hysterophorus* in Pakistan are limited, with most emphasis on manual removal and herbicide application. However, both strategies have their limitations. Manual removal of the weed without wearing protective clothing is very risky due to contact dermatitis while large-scale control using herbicides is economically and environmentally not sustainable.

To date the strategy of biological control has received limited attention contributing little to the management of the weed in Pakistan. The leaf-feeding beetle *Zygogramma bicolorata* Pallister (Coleoptera: Chrysomelidae), a natural enemy of *P. hysterophorus* in its native range, accidentally arrived in the Punjab district, Pakistan in 2007 via India, where it had been introduced as a classical biological control agent in 1984 (Winston et al. 2014); however, to date the beetle has not been actively managed as a control agent and its impact in the region is currently unknown.

The rust pathogen *Puccinia abrupta* var. *partheniicola* (H.S. Jacks.) Parmelee, commonly known as the winter rust of *P. hysterophorus* with a native distribution in Mexico, Central America, Argentina, Bolivia and Brazil (Evans 1997), is another natural enemy of the weed used as a biological control agent. The pathogen, which has been shown to be highly host specific, was introduced as a biological control agent for *P. hysterophorus* in Australia in 1991 (Parker et al. 1994). While not deliberately introduced into any other country, the rust has also been reported from China, India, Ethiopia, Kenya, Mauritius Nepal and South Africa (Dhileepan and Strathie 2009; Shrestha 2012), with the most recent record from Tanzania (Winston et al. 2014).

Previously no rust infection has been reported on parthenium weed in Pakistan and here we present the first record and distribution data of the winter rust *P. abrupta* var. *partheniicola* from the Islamabad, Punjab and KP Provinces of Pakistan. The presence of this rust could potentially add to the management toolbox for *P. hysterophorus* in Pakistan.

Materials and methods

Typical symptoms of a rust infection were observed on *P. hysterophorus* at several locations near Lahore and Rawalpindi districts of the Punjab between February–May 2019 (Table S1). Samples of rust infected leaves and stems were collected from the Rawalpindi site (CABI grounds, 33°38′41.1″N; 73°04′57.4″E), pressed dry between blotting paper to eliminate any contamination and sent to CABI UK for identification. Upon arrival, rust spores were carefully dislodged from individual pustules on infected

Figure 1. Young pustules of the rust *Puccinia abrupta* var. *partheniicola* on *Parthenium hysterophorus* (A); rust spreading on top branches and leaves of *P. hysterophorus* (B); infection spreading on upper surface of leaf (C); chlorosis and curling observed on lower surface of leaf (D); close up of rust pustules showing ruptured leaf epidermis (E) Habit of infected *P. hysterophorus* (F). Photographs by Iram M. Iqbal.

leaves onto a microscope slide using a fine needle and stained with lacto-fuchsin. Morphological characteristics were assessed and measured on 50 randomly selected spores and photographically documented.

Field specimens were collected periodically between February and May 2019 in order to observe the stages of the rust life cycle present on *P. hysterophorus*, as well as to assess the level of disease severity. Surveys within the districts of Punjab and KP where *P. hysterophorus* is prolific, were conducted between February–May 2019 in order to determine the current distribution of the rust. Dried specimens of *P. hysterophorus* leaves infected with the rust were deposited at Herb IMI Royal Botanic Gardens Kew, UK under the accession number IMI 507031, as well as at the Pakistan Museum of Natural History (PMNH) Islamabad under the accession number 15680 (Fungi Herbarium PMNH).

Results

Rust infection on *P. hysterophorus* in the field was first noted as light brown pustules developing on the adaxial surface of affected leaves (Figure 1A).

Figure 2. Microscopic image of urediniospores of *P. abrupta* var. *partheniicola* stained with lacto-fuchsin. Single arrows point to subequatorial germ pores; double arrow points to apical germ pore. Photomicrograph by Harry C. Evans.

On severely infected plants, pustules also developed on the stems, flower involucres and flower stalks (Figure 1B). Chlorosis and curling were observed on infected leaves as a first symptom before sporulation (Figure 1C, D). The average diameter of individual pustules was 2 mm (Figure 1E). Individual pustules often merged over time with rust infection being more severe on older leaves of affected plants (Figure 1F). Rust infected plants of *P. hysterophorus* measured 30–100 cm in height with abundant flowers (Figure 1F). Rust infection of *P. hysterophorus* was not limited to any specific habitat, and infected plants were observed along roadsides, in wastelands, as well as in agricultural fields. Rust infection was not observed in the field after May 2019 when day-time temperatures exceeded 30 °C and night-time temperatures were in excess of 22 °C.

The identity of the winter rust *P. abrupta* var. *partheniicola* on *P. hysterophorus* was confirmed based on morphological parameters, specifically of the uredinia and urediniospores, which matched the type description of *P. abrupta* var. *partheniicola* given by Parmelee (1967). Examination of urediniospores showed the characteristic cuneate shape and two subequatorial germ pores plus one apical germ pore (Figure 2). Urediniospore size was measured as 22–26 × 20–24 µm, which is within the range given by Parmelee (1967). Telia and teliospores were absent from all samples collected during the February to May 2019 season.

Surveys for the rust covered 65 sites in 19 districts of Punjab and KP (Table S1). The winter rust was recorded at 21 sites in 8 districts (Figure 3, Table S1). Severe attack was observed in the northern part of Punjab including

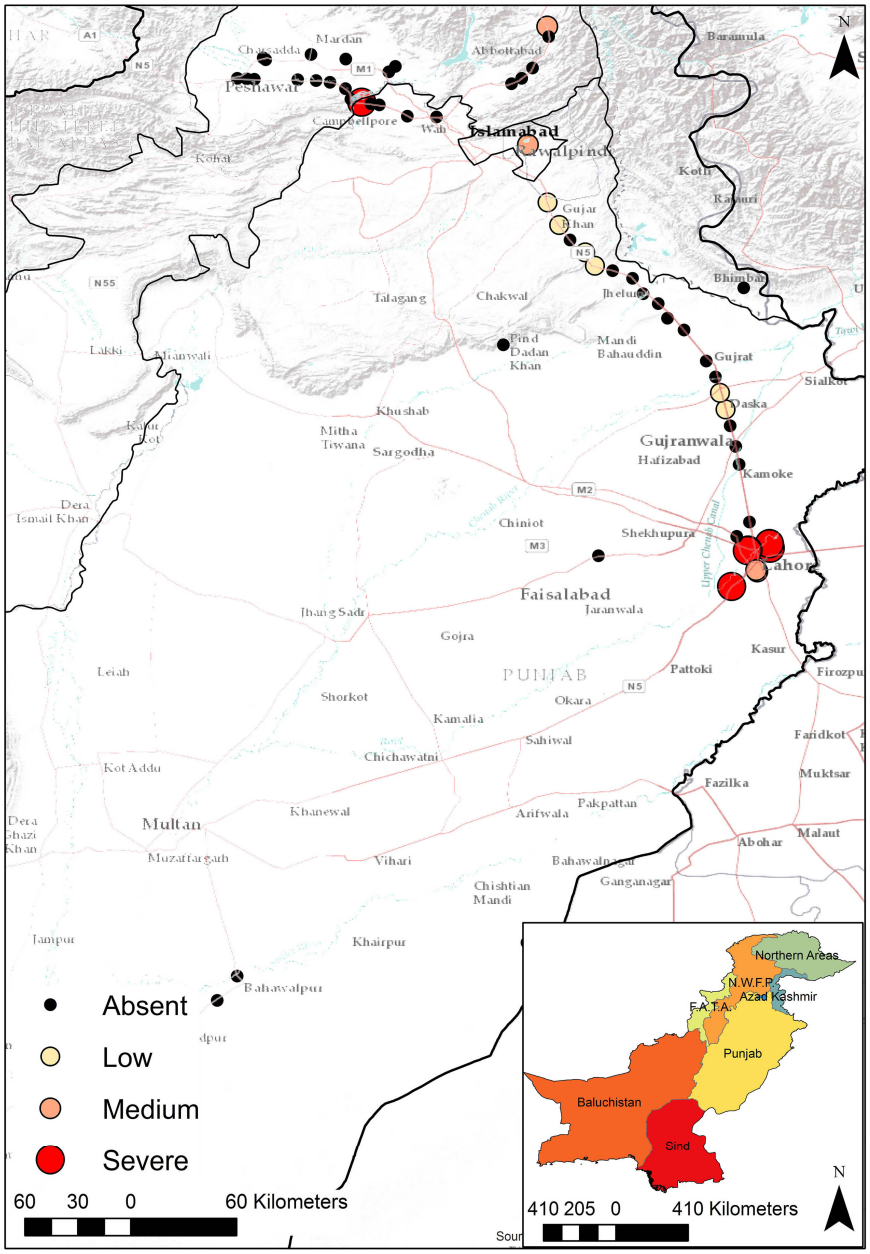

Figure 3. Survey sites showing current distribution of the winter rust *Puccinia abrupta* var. *partheniicola* in the Punjab and KP Provinces and Islamabad Capital Territory of Pakistan.

Lahore, Attock and Narowal districts, while in KP Province medium rust infection was recorded only in Mansehra district, and in other districts rust infection was considered low or absent (Figure 3, Table S1).

Discussion

The winter rust *P. abrupta* var. *partheniicola* has been confirmed to be present and relatively widespread in Pakistan. According to Evans (1987), the winter rust is a macrocyclic and autoecious species, which completes its life cycle on one host species, *Parthenium hysterophorus*, and does not require an alternate host.

Besides the beetle *Z. bicolorata*, the winter rust, *P. abrupta* var. *partheniicola* marks the second natural enemy against parthenium weed to be recorded

in Pakistan. In contrast to Australia, the rust has not been deliberately released in any other country, but has already been reported from India (Dhileepan and Strathie 2009), China (Day et al. 2018) and Nepal (Shrestha 2012). It is thus likely that the pathogen naturally dispersed to Pakistan, either from India or Nepal. The cool and moist conditions during the winter season 2018/2019 must have been conducive for *P. abrupta* var. *partheniicola* to cause an epiphytotic on its host in the Punjab and KP Provinces. The winter rust continued to sporulate on *P. hysterophorus* until the end of May, after which it disappeared in the field. Fauzi et al. (1999) suggested that the optimum temperature for infection and disease development of the winter rust is 15 ± 1 °C with an initial humid period, providing free water on leaf surfaces during the infection stage, of at least six hours. Urediniospore germination sharply decreases at temperatures above 20 °C (Fauzi et al. 1999). This would explain why when day and night time temperatures in the field exceeded 30 °C and 22 °C, respectively, combined with low humidity in May 2019, the rate of urediniospore infection and disease development of the winter rust on *P. hysterophorus* declined, ultimately leading to the absence of the pathogen in the field.

The winter rust has been considered a promising biological control agent against *P. hysterophorus* (Parker et al. 1994). In Australia, where the agent was deliberate introduced, *P. abrupta* var. *partheniicola* is reported to have established widely in south-eastern Queensland, but is restricted in its distribution by the abiotic requirements (temperature and dew period) for urediniospore infection and survival in other parts of the country which limits its overall impact on the weed (Dhileepan et al. 2018). Fauzi (2009) provides evidence that while the rust may not kill its host it can significantly reduce biomass and seed production, especially when applied at the rosette stage of *P. hysterophorus*. Further, the suppressive effect of rust was enhanced in the presence of competitive pasture plants and the winter rust is likely to maintain its effectiveness under elevated CO_2 concentrations (Shabbir et al. 2014). Therefore, we believe that the presence of *P. abrupta* var. *partheniicola* in Pakistan will aid the management of parthenium weed.

Acknowledgements

The authors acknowledge the contribution of the anonymous reviewers.

Funding Declaration

We wish to acknowledge the support of our donors who make Action on Invasives possible, in particular UK Aid (DFID) and the Directorate-General for International Cooperation (DGIS, Netherlands). CABI is an international intergovernmental organization, and we gratefully acknowledge the core financial support from our member countries (and lead agencies) including the United Kingdom (Department for International Development), China (Chinese Ministry of Agriculture), Australia (Australian Centre for International Agricultural Research), Canada (Agriculture and Agri-Food Canada), Netherlands (Directorate-General for International Cooperation), and Switzerland (Swiss Agency for Development and Cooperation). See http://www.cabi.org/about-cabi/who-we-work-with/key-donors/ for full details.

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Supplementary material

The following supplementary material is available for this article:

Table S1. Total Survey sites showing presence or absence of winter rust in different districts of Punjab, KP and Islamabad Provinces.

This material is available as part of online article from:
http://www.reabic.net/journals/bir/2020/Supplements/BIR_2020_Iqbal_etal_Table_S1.xlsx