



# *Pucciniastrum minimum* is the causal agent of rust on blueberries in New Zealand

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## Abstract

The causal agent of rust on blueberries in New Zealand was determined by molecular and morphological analyses as *Pucciniastrum minimum* rather than *Naohidemyces vaccinatorum* as first reported in 2004.

**Keywords** Blueberry · Pucciniales · Rust fungi · *Thekopsora minima* · *Vaccinium*

Three rust fungi were recognised by Sato et al. (1993) on blueberry plants (*Vaccinium* spp.); *Pucciniastrum minimum* (as *Thekopsora minima*) and two species which they placed in a new genus, *Naohidemyces*. Of the latter two species, *Naohidemyces fujisanensis* is restricted to Japan, but *N. vaccinatorum* (= *Melampsora vaccinatorum*) (Liang and Kakishima 2011) has a much wider distribution in North America, Europe and Japan. In 1980 and again in 1981, a rust identified as *P. vaccinii* was found on *Vaccinium vitis-idaea* plants growing in quarantine in Auckland, New Zealand. The plants, which had been imported from Germany, were destroyed.

In 2004, a rust was found infecting blueberry plants in an orchard in the Waikato region, North Island of New Zealand and was deposited in the New Zealand Fungarium with the accession number PDD 78424. Only urediniospores were observed and this specimen was identified as *Naohidemyces vaccinatorum* (Anonymous 2006).

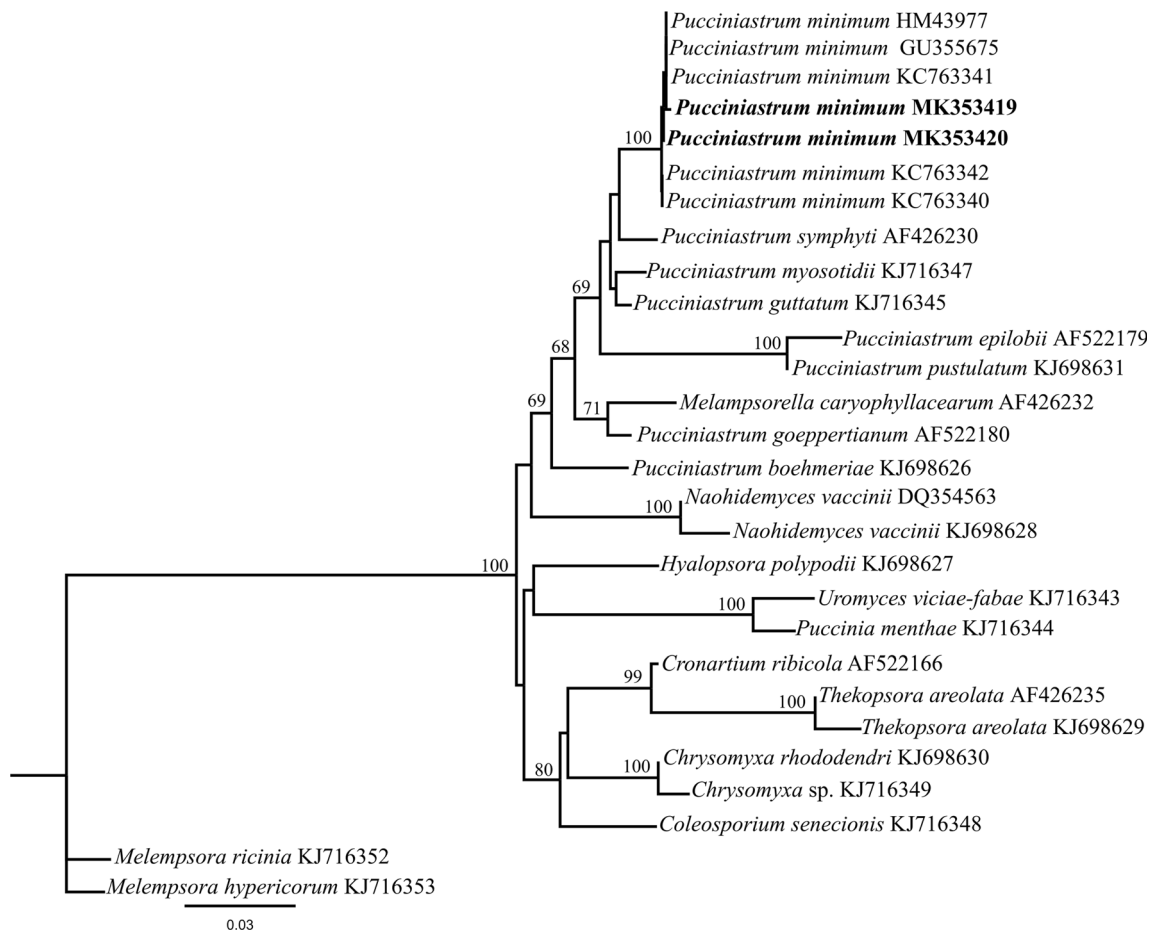
A recent publication (McTaggart et al. 2013) demonstrated that *Pucciniastrum minimum* (as *Thekopsora minima*) caused a rust on *Vaccinium corymbosum* in Queensland and New South Wales

in Australia, which prompted a re-examination of the New Zealand collections of rust fungi on blueberries. DNA from two recent collections (PDD 104137 and PDD 102321) was extracted and the 28S region (nuLSU) was amplified according to the protocol outlined in Padamsee and McKenzie (2014). The LSU sequence of PDD 104137 and PDD 102321 (GenBank MK353419 and MK353420, respectively) had 99% identity (with 100% query coverage) to *Thekopsora minima* (HM439777) in a BLAST search.

The newly generated sequences were aligned with five other sequences of *P. minimum* (as *T. minima*) deposited in GenBank and 21 sequences from the Padamsee and McKenzie (2014) study, which demonstrated that *Thekopsora minima* is recovered in a clade that encompasses *Pucciniastrum* sensu stricto and as a result the appropriate name for this species should be *Pucciniastrum minimum*. The dataset was analysed in GARLI (Zwickl 2006) using the GTR +  $\Gamma$  + I model of evolution and 1000 maximum likelihood bootstrap support (MLBS) values were generated in PhyML 3.0 (Guindon and Gascuel 2003). In the resulting phylogeny (Fig. 1), the generated sequences of the rust on blueberry in New Zealand were recovered in a highly-supported clade (100% MLBS) with the other five sequences of *P. minimum* (as *T. minima*). The phylogeny also demonstrates that this fungal species is recovered in the moderately supported clade (69% MLBS) with other *Pucciniastrum* species, including the generic type *P. epilobii*.

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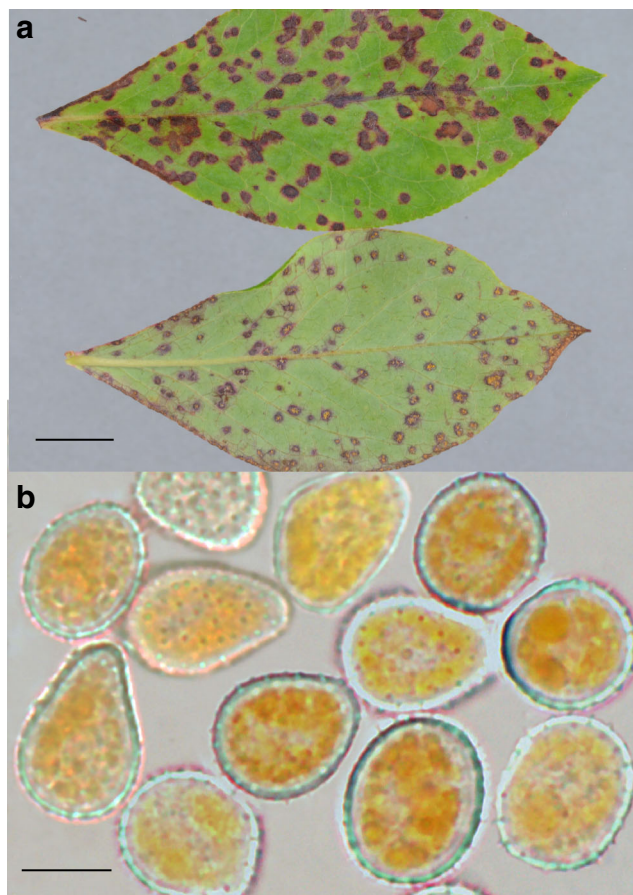


**Fig. 1** Phylogram obtained from maximum likelihood analysis of nuclear LSU rDNA. Bootstrap support values (> 65%) from a maximum likelihood search with 1000 replicates shown

The original 2004 specimen and several later collections were examined: on *Vaccinium* sp. 'Jersey', Auckland, Warkworth, Omaha, Jones Road, 5 Apr 2005, C.F. Hill (PDD 82858—II); on *Vaccinium* sp. 'Misty', Mt. Albert, 27 Nov 2014, P. Wilkie (PDD 105401—II); on *Vaccinium* sp., Waikato, Te Awamutu, Ngaroto Nurseries, 15 Jan 2004, S. Ganey (PDD 78424—II); on *Vaccinium* sp., SH 1B bypass of Hamilton, south of crossing east coast main trunk railway, 13 Jan 2012, E.H.C. McKenzie (PDD 102321—II), GenBank MK353419; on *V. corymbosum*, Mid Canterbury, West Melton, Weedons Ross Road, 22 Jan 2014, G. Logan (PDD 104137—II), GenBank MK353419. All specimens had the following morphology. Uredinia were hypophyllous, golden-yellow, scattered or 4–5 grouped on angular, vein-limited brown spots and a dark brown spot with a red border on the corresponding upper-leaf surface, uredinia circular, up to 0.25 mm diam., bullate,

opening by a central pore, and becoming pulverulent (Fig. 2a). Urediniospores were subglobose, ellipsoidal or obovoid, 15–21(–23) × (12–)13–15(–18) µm (mean 18.5 × 14.1 µm), with yellow contents, and a wall (0.5–)1(–1.5) µm thick, hyaline, and echinulate; germ pores were not seen (Fig. 2b).

*Pucciniastrum minimum* (as *T. minima*) has been recently recorded in South Africa (Mostert et al. 2010) and in Mexico (Rebollar-Alviter et al. 2011). Although it proved impossible to extract DNA from the rust on the blueberry plants imported to New Zealand from Germany in the 1980s, the known world distribution of *P. minimum* and *Naohidemyces vaccinatorum* would suggest that it was *N. vaccinatorum*. As those plants with their rust were destroyed, we may conclude that the only rust fungus present on blueberries in New Zealand currently is *P. minimum*.



**Fig. 2** *Pucciniastrum minimum* on *Vaccinium corymbosum*. **a** Abaxial and adaxial leaf symptoms; **(b)** Urediniospores. Scale a= 5 cm; b= 10  $\mu$ m

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