

# **Taxonomy, phylogeny and population biology of the red band needle blight pathogen and related species**

BY

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## DECLARATION

I, the undersigned, hereby declare that the thesis submitted herewith for the degree *Philosophiae Doctor* to the University of Pretoria, contains my own independent work and has hitherto not been submitted for any degree at any other University.

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## PREFACE

Chapter one of this thesis presents the literature pertaining to biological invasions and the different stages of invasions in terms of plant pathogens. The review focuses on determining areas where studying the population genetics of a pathogen might complement the knowledge of the ecology of the pathogen in order to gain a better understanding of their evolutionary and invasive potential. *Dothistroma septosporum*, which causes Dothistroma needle blight (DNB), is one of the better known examples of an invasive plant pathogen and is the focus of this thesis. After presenting an overview of the pathogen, it is concluded that a thorough knowledge of the ecological aspects of the pathogen has been gained by many intensive studies but that there is a distinct, and perhaps dangerous, lack of knowledge pertaining to the global population genetics of this pathogen.

Throughout the years of this study, DNB has become a global concern as more epidemics were being reported, not only in the Southern Hemisphere where the disease has been problematic for over 50 years, but also in the Northern Hemisphere in both native and non-native forests. An “International Dothistroma Alliance Committee” was established in 2004 among researchers world-wide with the aim being to share and integrate knowledge about the pathogen and the disease it causes. The main focus of this group incorporates aspects such as diagnostics and monitoring, assessing pathogen risk (pest-risk), disease impact, management and pathogen variability as well as population diversity. The research in this thesis greatly assists in the latter two aims of the committee.

In the second chapter, multigene phylogenies were constructed from *Dothistroma septosporum* isolates obtained worldwide. These studies revealed that the disease is not caused by one pathogen with varying morphotypes, but that two, very closely related sibling species are responsible. A thorough description of the two species, named *Dothistroma septosporum* and *D. pini*, is provided. It is also shown in Chapter six that both species are capable of infecting the same needle and can co-exist within the same conidioma. There is thus a distinct potential for hybridisation to occur between these two species.

Coincident with the initiation of this research is an increase globally in the incidence of DNB. This increased the knowledge base about the pathogens around the world and also made it easier to obtain cultures and isolates for this study from different sources. As a consequence, an understanding of the distribution and occurrence of each species was possible and is

documented in Chapters three and six. The molecular techniques developed in this work have enabled the development of a robust technique to distinguish between the two pathogens causing DNB. A combination of the species-specific mating type markers designed by Groenewald *et al.* (2007), the diagnostic Primer\_A, and an effective species-specific RFLP test, provide a quick and effective means of identifying the DNB pathogens, directly or indirectly, from conidiomata on infected needles.

In order to study the population diversity of the DNB fungi, twelve microsatellite markers were developed in Chapter three. In Chapter four, the preliminary applications of these markers provide a first glimpse of the global diversity of *D. septosporum* which has caused the most devastation world-wide. From this research it is also obvious that the pattern of diversity reflects the movement of its host (pine) from its native Northern Hemisphere to various countries within the Southern Hemisphere.

*Dothistroma septosporum* is an important disease of both plantation grown pines and native forests. Climate change and the continual movement of infected plant material pose great threats to existing forests. The fact that a sibling species has been discovered shows the evolutionary potential of the species to adapt to changing environments. Successful quarantine and monitoring will aid in curbing the further spread of the disease into areas where it could, potentially, be more devastating.

The chapters in this thesis correspond to different research projects and are represented in the text in the format of a publication. Due to the nature of this style, however, there is some unavoidable repetition in the text, especially within the introduction of each chapter. Three of the chapters within this thesis have been published in internationally recognised ISI rated journals.