Characterisation of *Botryosphaeria* species from

mango in South Africa

Submitted by

René Jacobs

A thesis submitted in partial fulfilment of the requirements for the degree of

MAGISTER SCIENTIAE

In the Faculty of Natural and Agricultural Sciences, Department of Plant Pathology and

Microbiology, Forestry and Agricultural Biotechnology Institute, University of Pretoria,

Pretoria, South Africa

July 2002

Supervisors: Prof. L. Korsten

Mr. B. Slippers

Prof. M.J. Wingfield

DECLARATION

I, the undersigned, hereby declare that the thesis submitted herewith for the degree Magister Scientiae to the University of Pretoria, contain my own independent work and has not been submitted for any degree at any other University.

René Jacobs

July 2002

TABLE OF CONTENTS

ACKNOWLEDGEMENTS

This thesis is dedicated to my husband, Francois for your unconditional love and support. You help me fulfill all my dreams and aspirations. Thank you for everything!

I would like to express my most sincere thanks and gratitude to the following:

'When the going gets tough, the tough gets going', but my parents have kept me going all the time and I thank you from the bottom of my heart.

Bernard Slippers for your enthusiasm, guidance, encouragement and endless support. You have made me see the molehill in every situation when I could only see the mountain in front of me and I will be eternally grateful for that.

Mike Wingfield and Lise Korsten for helping me keep 'my eye on the ball' and for your willing support, guidance and the reviewing of this thesis.

Fanus Swart (DuRoi-UP alliance) for your time, effort and field knowledge. It is much appreciated.

For financial support, I would like to thank the National Research Foundation (NRF), South African Mango Growers Association (SAMGA) and the University of Pretoria and THRIP.

Most importantly, I thank my heavenly Farther for His eternal love and light upon my path.

PREFACE

Botryosphaeria spp. are well-known endophytes and opportunistic pathogens of many woody hosts, including mango (*Mangiferae indica* Linn.). In South Africa, *Botryosphaeria* spp. commonly cause cankers, twig die-back, blossom blight and fruit rot of mango, which result in significant economic losses annually. Limited control is available for these diseases, which is partly due to the lack of knowledge concerning the identity and epidemiology of the causal agent. There is, however, confusion regarding the taxonomy of the causal agent. This thesis aims to address the current lack of knowledge and problems with taxonomy, efficient identification and etiology of *Botryosphaeria* spp. on mango in South Africa.

In the first chapter, the literature concerning *Botryosphaeria* spp. that occur on mango is reviewed. Information is summarized and reviewed under three main themes, namely taxonomy, epidemiology and current control of *Botryosphaeria* diseases of mango. The taxonomy of *Botryosphaeria* spp. is currently in disarray due to difficulties in identifying species based on morphological characteristics. Various species have been implicated as causal agents of mango diseases previously. Due to the discrepancies over the taxonomic status of the pathogen, chritical epidemiological issues and aspects could not be clarified, thus, making the development of an effective disease management strategy difficult. This has a complicating effect on the control of the pathogen, which is currently achieved with limited success. The need for a revision of the taxonomy of the *Botryosphaeria* pathogens involved in mango diseases is emphasised, as correct identification of the pathogen is the first step in developing effective control strategies and quarantine regulations.

6

The taxonomy of *Botryosphaeria* is currently confusing, since various species, namely *Hendersonia creberrima*, *Dothiorella dominicana*, *Nattrassia mangiferae* and *Lasiodiplodia theobromae*, are names of fungi assumed to cause various diseases on mango. Most of these taxa are, however, not validly described and the identity of these species is suspect. These pathogens were previously identified by using only anamorph morphological characteristics. The use of morphology alone is, however, questionable due to the overlap between some species. In Chapter two, traditional morphological characterisation was combined with molecular sequence data to identify the *Botryosphaeria* spp. from mango in South Africa. This was compared and related to species affecting mango in other parts of the world.

Confirming the pathogenicity of the different *Botryosphaeria* spp. occurring on mango in South Africa will be important for implementing disease control and quarantine strategies. Little information is currently available regarding the pathogenicity and role of the four *Botryosphaeria* spp. identified from mango in South Africa. Two of these *Botryosphaeria* spp. are newly described and their pathogenicity has not been determined. The objective in the third chapter of this thesis was to utilise apple and potted tree inoculation trials, to determine pathogenicity of all *Botryosphaeria* spp. from mango in South Africa. The identity of the most and least pathogenic *Botryosphaeria* spp. on a resistant and susceptible commercial mango cultivar in South Africa was also investigated.

Identification of *Botryosphaeria* spp. is focused on combined morphological and sequence data. Morphological data is not always reliable and the use of sequence data in species identification of large numbers of isolates is impractical. For this reason, a PCR-RFLP technique was considered in Chapter four as an alternative approach to achieve rapid and reliable identifications for *Botryosphaeria* spp. from mango. This technique was then used to

7

identify a large number of isolates collected during a survey of *Botryosphaeria* spp. from South Africa and Australia.