Chapter 5 — The potential global geographical distribution of Citrus Black Spot caused by *Guignardia citricarpa* (Kiely), with emphasis on the likelihood of disease establishment in the European Union

5.1 Abstract

This study represents a climatic modelling approach towards assessing the risk that Citrus Black Spot (CBS) will spread to regions of the world where the disease does not occur. Globally, CBS is widespread, but is absent within countries of the European Union (EU). Thus, barriers to the trade of citrus fruit have been put in place to restrict the possible introduction of the pathogen *Guignardia citricarpa* Kiely. The objective of this study is to evaluate the climatic suitability of the European climate for the establishment of the pathogen and persistence of the disease. For this purpose, the CLIMEX model was used which allows for the prediction of the potential geographical distribution of a species using its observed geographical distribution. In this study, the climatic requirements of CBS were inferred from the distribution thereof in South Africa and Australia. The model output reflected the current known distribution of the pathogen around the world and indicated that climate provides a barrier to the establishment of the disease in Europe. The potential distribution of CBS was mainly limited by cold conditions. A global map was produced which indicates localities where the climate is suitable for the potential establishment of the pathogen.

5.2 Introduction

Citrus Black Spot (CBS), caused by *Guignardia citricarpa* Kiely [anamorph *Phyllosticta citricarpa* (McAlpine) van der Aa], is a foliar and fruit disease of citrus. The disease affects the rind of the fruit, causing superficial lesions (Kotzé, 1981; Snowdon, 1990). Most commercial citrus cultivars are susceptible to CBS to some degree, with lemons and Valencia oranges being highly susceptible (Kiely, 1948b; Kotzé, 2000). The disease may also cause significant losses on grapefruit and limes (Brodrick, 1969), and has been reported to occur on citron, pomelos and mandarins (Brodrick, 1969; Kiely, 1948b, 1970). Rough lemon is tolerant (Wager, 1952) and Persian limes (Timmer, 2005, Personal Communication) and sour orange are not susceptible to the pathogen (Kotzé, 1981).

The earliest official description of CBS was by Benson, (1895), from diseased fruit originating from citrus growing areas within New South Wales (NSW), Australia. He published drawings of diseased fruit, but did not study the causal organism. Subsequently, CBS was recorded from orange orchards near Sydney, NSW (Cobb, 1897). The first record of CBS in South Africa (SA) was in 1929 from areas around Pietermaritzburg (Doidge, 1929). The disease has also been recorded in other countries, including Argentina (Garran, 1996), Bhutan (European Union, 1998), Brazil (European Union, 2000a), China (European Union, 1998), Ghana (Timmer, 2005, Personal Communication), Indonesia, India (Brodrick, 1969), Kenya, Mozambique (European Union, 1998), Nigeria (Baayen et al., 2002), Philippines, Swaziland, Taiwan, Uruguay (Kotzé, 2000), West Indies (Calavan, 1960), Zambia and Zimbabwe (European Union, 1998).

Reports on the occurrence of CBS in New Zealand and Japan are conflicting and it is uncertain if the disease occurs in these countries (CABI/EPPO, 1998; Sutton & Waterson, 1966). Although a pathogen of citrus, *P. citricarpa* does occur in Japan, it causes fruit decay and not rind spotting (Kuramoto, 1981). Additionally, an isolate of *Guignardia* (PPRI 1567) from Japan, previously thought to be the pathogen *G. citricarpa*, was identified as the endophyte *G. mangiferae* (A.J. Roy) by Baayen et al. (2002). Citrus Black Spot symptoms have never been seen on citrus fruit in New Zealand (Everett & Hale, 2002; Dawson, 2003, Personal Communication; Tyson, 2003, Personal Communication).

The disease has not been recorded in citrus-producing Mediterranean and European countries (European Union, 1998; Kotzé, 2000) including Greece, Israel, Italy, Spain, Portugal, France and Turkey (Baayen et al., 2002; Bonants et al., 2003). It does not occur in Chile or the citrus growing areas of the USA (Baayen et al., 2002; Cook, 1975; European Union, 2000b; Kotzé, 1981).

Various citrus growing areas, within countries where the disease has been recorded, have remained free of CBS. In China, the disease has only been recorded in the provinces of Sichuan, Yunnan, Guangdong, Fujian and Zhejiang (European Union, 1998). In Brazil, it was first recorded in 1980 in the state of Rio de Janeiro and also in Rio Grande do Sul (in 1986) and São Paulo (in 1992) (European Union, 2000a). In SA, the Hartswater (USDA/APHIS, 2002),

Vaalharts and Warrenton citrus production regions in the Northern Cape (Mabiletsa, 2003) and all the citrus production regions within the south-western Western Cape are free of CBS (European Union, 1998; Mabiletsa, 2003). Despite movement of CBS-infected citrus fruit and potentially infected nursery trees [prior to the implementation of the restriction on the movement of citrus propagation material in 1983 (Agricultural Pests Act, 1983)] into this region (Kotzé, 2002, Personal Communication) it has remained free of CBS (European Union, 1998; Mabiletsa, 2003). Similarly, CBS infected fruit have been introduced to the inland citrus producing areas of NSW in Australia and the disease has also not been able to establish in this region (Barkley, 2003, Personal Communication; Whiteside, 1965). Other parts of Australia reported to be free of CBS are the Sunraysia and mid-Murray areas of Victoria and NSW, and the entire states of Western Australia and South Australia (including the Riverland region) (Barkley, 2003, Personal Communication; European Union, 1998). This suggests that climate plays an important role in the potential distribution of the disease.

World-wide, South Africa is the third largest exporter of fresh citrus fruit. Approximately 50% of South African citrus was exported during the 2001/2002 season (FAO, 2002). However, the export of fruit from regions infected with CBS to the European Union (EU) is restricted through quarantine regulations (Bonants et al., 2003; European Union, 2000c). Similar phytosanitary restrictions affect the export of citrus from Argentina (European Union, 2001a), Australia (European Union, 1998) and Brazil (European Union, 2000a) into the EU.

A Pest Risk Analysis (PRA) for the export of fresh citrus fruit from CBS-infected production regions in SA to the EU was conducted (Hattingh et al., 2000). This PRA included a review of the disease and results of studies conducted to determine the survival of conidia on fruit in the packinghouse. In addition, there was a need for a predictive climate matching study (European Union, 2001b). Therefore, the main objective of this study was to evaluate the suitability of European climates for the establishment of CBS. The premise of this approach was that CBS only occurs in areas with a climate suitable for disease development. Consequently, there would be no risk of inadvertently introducing the disease into regions where climatic conditions are unfavourable for disease development.

For this purpose, the Compare Locations function in CLIMEX (Hearne Scientific, Melbourne, Australia) was used. This function compares the relative potential for growth and persistence of a species in different localities and allows prediction of the potential geographic distribution of that species based on its climatic requirements (Sutherst & Maywald, 1985; Sutherst et al., 2003). The model is based on the assumption that organisms are efficient integrators of climate and other environmental variables. The seasonal phenology, geographical distribution and relative abundance of the population reflect the integration processes. It is difficult to capture these complex processes in experiments or in process-based simulation models. CLIMEX is especially useful in cases where there is a lack of process-related data. The aim of this model is therefore to describe the core responses of a species to climate by providing a single number to

indicate the climatic favourability of a location for a specific species (Sutherst & Maywald, 1985; Vera et al., 2002).

There have been examples where climatic modelling approaches have been used to determine the risk of potential establishment of plant pathogenic species. These include: leaf blight [Cylindrocladium quinqueseptatum (Boedijn and Reitsma)] of Eucalyptus in mainland Asia and globally (Booth et al., 2000a); Eucalyptus rust [Puccinia psidii (Winter)] in the Neotropics and Australia (Booth et al., 2000b); and Sphaeropsis sapinea [(Fr.) Dyko and Sutton] and Cryphonectria cubensis [(Bruner) Hodges] (pathogens of Pinus and Eucalyptus spp.) in South Africa (Van Staden et al., 2004). The risk of establishment of plant pathogens has also been successfully defined a priori using climatic modelling with CLIMEX, for example oak decline [Phytophthora cinnamomi (Rands)] in southern Europe (Brasier, 1996) and soybean rust [Phakopsora pachyrhizi (Sydow)] throughout the world (Pivonia & Yang, 2004).

CLIMEX aims to provide answers to practical problems about the control of invasive species and provides a modelling technique that allows the incorporation of new information as it becomes available. CLIMEX model outputs can assist in estimating the risk of introducing novel species. This will allow preventative or adaptive disease management approaches (Booth et al., 2000a) and can support decision-making with regards to quarantine issues, particularly when existing information is sparse (Worner, 1988).

Since citrus is a high value fruit crop and CBS is economically important and currently a technical barrier to trade for certain countries, the potential global distribution of this disease is of interest to all citrus producing countries of the world. Climate plays a key role in the epidemiology of this disease, especially since relatively high temperatures and the availability of moisture promote pathogen establishment, and high temperatures increase disease severity (Kotzé, 1971, 1996). The objective of this study is to explore the influence of climate on the potential for disease establishment in areas where CBS has previously not been recorded.

5.3 Methodology

5.3.1 Epidemiology of Citrus Black Spot

Two types of spores may cause infection of citrus, namely windborne ascospores (contained in perithecia) and waterborne conidia (contained in pycnidia) (Kiely, 1948b; Kotzé, 1963, 1996). Both kinds of fruiting bodies may be found on a single leaf, but only pycnidia occur on infected fruit (McOnie, 1965). In wet weather, conidia ooze from pycnidia. These conidia then require running water for dissemination and specific conditions for germination (Kiely, 1948b; Korf et al., 2001).

Ascospores are seen to be the most important source of inoculum (Kiely, 1948b; Kotzé, 1963, 1996). Successful maturation of perithecia is determined by prevailing weather conditions, as optimal maturation requires alternate wetting and sun drying of leaves, and fluctations in temperature (Kiely, 1948a, 1948b, 1949; Kotzé, 1981; McOnie, 1967). Discharge of ascospores

relies on wetting of the perithecia. Germination of ascospores and infection then requires the presence of free surface water and suitable microclimatic conditions (Kiely, 1948b).

The critical period for infection of fruit is from fruit set until up to five months later when the fruit becomes resistant to further infection (Kotzé, 1981). Mature fruit is not infected (Wager, 1949). Therefore, the onset of rain, ascospore release and infection period are strongly correlated (Kotzé, 1963; McOnie, 1964; Whiteside, 1967). Where these three factors do not coincide, no epidemic will develop (Kotzé, 2002, Personal Communication).

5.3.2 Outline of the CLIMEX model

The CLIMEX model integrates the weekly responses of a population to climate into a series of annual indices. A hydrological model is used to calculate weekly soil moisture from rainfall and estimated evaporation. Responses to temperature and moisture are combined into a weekly population Growth Index (Glw) for that species. The annual Temperature Index (TI) and annual Moisture Index (MI) summarize the response of the species to temperature and moisture respectively. Responses to detrimental conditions are reflected by a series of "stress indices" that estimate the harmful effects of either extreme or prolonged exposure to hot, cold, dry or wet weather. These stress indices may be applied on their own or in combination with each other, so the stress accumulated may include hot and dry, or, cold and wet stress. These growth and stress indices are combined into an Ecoclimatic Index (EI). The EI indicates the suitability of a certain geographical location for the multiplication, infection and persistence of the species on a scale from 0 to 100. Generally, an El value greater than 30 can be considered very favourable for population growth and persistence, while an El value equal to zero suggests that the species will be unable to persist under average climatic conditions (Sutherst et al., 2003). However, El values are based on the species occurrence and abundance and EI values indicating a highly favourable climate will differ per species based on the requirements of the species. So for instance, Vera et al. (2002) use El values of higher than 25 as highly favourable for the establishment of mediterranean fruit fly in Argentina and Australia, and D'Amamo et al (2002) use an EI of 20 and higher to circumscribe a highly favourable climate for the establishment of german wasps in Argentina.

The values of the CLIMEX model parameters, which reflect the climatic requirements of the species, are inferred from information on the currently known distribution of the species. The procedure is referred to as inverse, or inferential, modelling. In CLIMEX, this involves developing hypotheses as to which factors limit the distribution, and then manually adjusting parameter values until the simulated geographical distribution coincides as closely as possible with the observed distribution (Sutherst & Maywald, 1985). A specific parameter set should describe the climatic responses of the species. Once the parameter values have been estimated, the parameter set may be used to predict the potential occurrence of the species in other locations (McFadyen & Skarratt, 1996). Sutherst and Maywald (1985) describe the system in full and give details on the theoretical background and application of the program.

5.3.3 Data

5.3.3.1 The geographical distribution of Citrus Black Spot in South Africa and Australia

In 2002, six field pathologists, with extensive knowledge of CBS, mapped the geographical distribution of the CBS in SA (as described in 4.3.1, page 69) and this resulted in a map of the actual presence of the disease in SA at the time of assessment (Figure 5.). Information on the presence of CBS in Australia as obtained from the Australian Plant Pest Database (APPD), was provided by Patricia Barkley (formerly NSW Agriculture, Camden, Australia) and Andrew Miles (Queensland Department of Primary Industries, Indooroopilly). A map of the known occurrence of CBS in Australia was drawn up from these data (Figure 5.2).

5.3.3.2 Climate data

The meteorological database within CLIMEX Version 2. consists of values for monthly long-term average maximum and minimum temperature, rainfall and relative humidity for 3092 locations. Of these localities, 129 are in SA, 676 are in Australia and 285 are in Europe. The model was run for all localities.

5.3.4 Predicting the distribution of Citrus Black Spot

The CLIMEX parameter values were estimated using the known current distribution of the disease in SA (Figure 5.) and in Australia (Figure 5.2), also taking into consideration the officially recorded absences of CBS in the Western Cape of South Africa and the inland parts of NSW in Australia. The values were iteratively adjusted until there was a close visual match between the current known distribution and the predicted potential distribution. Particular attention was paid to areas around Nelspruit in SA, where the disease has been known to occur in serious epidemic proportions. The final parameter values were applied to predict the potential distribution of CBS on a global scale.

5.3.5 Validation

The EI values for the occurrence of CBS, as predicted by the model, were split into three categories: EI \leq 4, climate unfavourable for the persistence of the species; EI \geq 5 and EI \leq 10, climate marginally suitable for disease development; EI \geq 11, climate favourable for disease development (locations with EI values above 20 had climates highly favourable for CBS).

To test the validity of these categories, data on the presence or absence of CBS, as obtained from literature or through personal communication were collected for 537 meteorological localities. These localities were from the CLIMEX program, but were independent of those used to build the model. These localities were predominantly from Australia, as the most accurate data was available for Australia, but some localities were from South Africa, Brazil, Zimbabwe, Indonesia and Argentina. The localities were divided into sites where a citrus host was probably present (70 localities) and those where the presence of a citrus host was not known (467 localities). At each locality, El values greater than 4 were counted as predicted presences, while El values equal or smaller than 4 were counted as predicted absences. The predicted

presences and absences were then compared to the observed presences and absences at those same localities using the Kappa statistic (κ). κ is used to measure the agreement between two sets of categorical data while correcting for chance agreements between the categories. This statistic based on a confusion matrix which relies on both presence and absence records (Robertson & Palmer, 2002) (Table 5.1). The absolute counts for the localities where citrus was probably present is presented in Table 5.2 and the absolute counts for the localities where the presence of a citrus host was not known are presented in Table 5.3.

Table 5.1 — A confusion matrix used to calculate the Kappa statistic (κ). The paramaters a, b, c, and d represent absolute counts.

		Observed	
		presence	absence
Predicted	presence	а	b
Fredicted	absence	С	d

Table 5.2 — The confusion matrix used to calculate the Kappa statistic (κ) for the localities where citrus is probably present.

		Observed	
		presence	absence
Predicted	presence	13	0
Fredicted	absence	0	57

Table 5.3 — The confusion matrix used to calculate the Kappa statistic (κ) for the localities where the presence of a citrus host was not known.

		Observed	
		presence	absence
Predicted	presence	0	0
Fredicted	absence	0	476

The value for κ lies between zero and one. A value of greater than 0.75, indicates an excellent agreement beyond chance, a value smaller than 0.4 indicates poor agreement, and a value close to zero would indicate agreement that is no better than random (Landis & Koch, 1977; Monserud, 1990).



Figure 5.1 — The distribution of Citrus Black Spot in South Africa

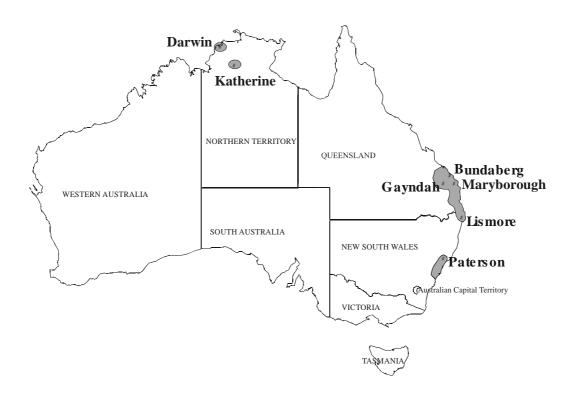


Figure 5.2 — The distribution of Citrus Black Spot in Australia

5.4 Results

5.4.1 Potential distribution of Citrus Black Spot

The parameter values that best describe the observed distribution are provided in Table 5.4. The predicted areas of occurrence of the disease in SA (Figure 5.3) and Australia (Figure 5.4) closely fit the observed distributions of the pathogen in these countries. The radii of the circles on the maps are proportional to the calculated EI values and give an indication of the climatic suitability for persistence of CBS.

Within SA, the model predicted EI values of above 30 at all the localities where the disease occurs abundantly, such as Barberton (EI=36), Komatipoort (EI=33) and Nelspruit (EI=37) (Figure 5.3), indicating that the climate at these localities is highly suitable for the establishment of the disease. Similarly, the model predicted EI values of higher than 20 for localities in Australia known to have a high occurrence of CBS, namely Lismore (EI=24) and Paterson (EI=21) in NSW, and Maryborough, (EI=25), Gayndah (EI=25) and Bundaberg (EI=24) in Queensland (Figure 5.4).

Interestingly, the EI values of several localities within SA where the disease has not been reported to occur are highly suitable for the occurrence of CBS. These localities include East London (EI=30) and Umtata (EI=21) in the Eastern Cape, and Ladysmith (EI=28) and Dundee (EI=20) in Kwazulu Natal. Similarly, in Australia, the model indicated a potential for establishment in localities where CBS has not yet been recorded, in coastal NSW (Casino, EI=31; Grafton, EI=30; Taree, EI=23; Kempsey, EI=23), and in Queensland (EI values above 25—Charters Towers, Gladstone, Mount Morgan and Rockhampton).

The indices describing cold stress were crucial in explaining the distribution of the disease in SA, Australia and around the globe. The main factor limiting the distribution of the disease was found to be cold stress in the form of continuous winter days with temperatures too low to allow the survival of the pathogen. All southern inland areas of Australia (including the citrus growing areas of Sunraysia, Riverland, Riverina) were excluded due to cold stress, but also as a result of a lack of sufficient moisture for growth, and the EI values in these areas never exceeded a value of one. Additionally, the EI values for localities within the inland parts of NSW (Bourke and Narromine) were smaller or equal to four mainly due to a lack of moisture. In the coastal and inland areas of the south-western Western Cape in SA, where all localities had EI values smaller or equal to four cold stress was the main limiting factor for occurrence of the disease. Although cold stress accumulated in the Northern Cape region of SA, the main factor limiting the distribution of the disease was insufficient moisture for growth.

Table 5.4 — CLIMEX parameter values giving the best fit to the distribution of Citrus Black Spot caused by *Guignardia citricarpa*.

Parameter Description	Value
Temperature parameters	
Lower threshold of temperature for population growth°C (DV0)	17
Lower optimal temperature for population growth°C (DV1)	24.5
Upper optimal temperature for population growth°C (DV2)	32
Upper threshold temperature for population growth°C (DV3)	40
Moisture parameters	
Lower threshold of soil moisture (SM0)	0.18
Lower limit of optimal range of soil moisture (SM1)	0.45
Upper limit of optimal range of soil moisture (SM2)	0.85
Upper threshold of soil moisture (SM3)	1.0
Stress indices	
Cold stress	
Cold stress temperature threshold°C (TTCS) (below which cold stress accumulates)	11
Cold stress temperature rate (THCS)	-0.0001
Cold stress degree-day threshold (DTCS)	6
Cold stress degree-day rate (DHCS)	-0.00025
Dry Stress (not used)	
Heat stress	
Heat stress temperature threshold°C (TTHS) (above which heat stress accumulates)	40
Heat stress temperature rate (THHS)	0.001
Heat stress degree-day threshold (DTHS)	25
Heat stress degree-day rate (DHHS)	0.001
Wet stress	
Wet stress threshold (SMWS)	1.45
Wet stress rate (HWS)	0.0001

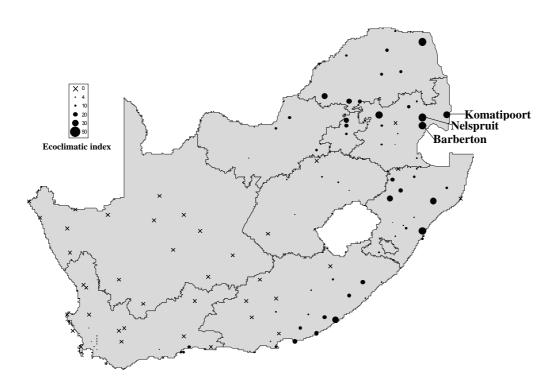


Figure 5.3 — The potential geographical distribution of Citrus Black Spot in South Africa as fitted by the CLIMEX model. The Ecoclimatic Index (EI) is proportional to the radii of the circles. Locations indicated with an x represent those with an EI of 0.

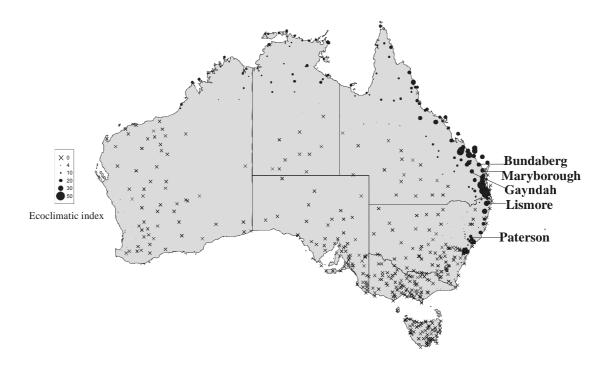


Figure 5.4 — The potential geographical distribution of Citrus Black Spot in Australia as fitted by the CLIMEX model. The Ecoclimatic Index (EI) is proportional to the radii of the circles. Locations indicated with an x represent those with an EI of 0.

University of Pretoria etd – Paul, I (2006)

Globally the distribution, especially in the northern hemisphere, was limited by cold stress, whereas heat stress limited the distribution in Northern Africa and parts of India, Pakistan, Iraq and Saudi Arabia.

Currently, the disease does not occur in Europe (Baayen et al., 2002). The CLIMEX model indicated that of the 285 localities in Europe 228 had an EI=0, indicating that the species will be unable to establish or persist at those localities. An EI=0 was found at all the localities in Austria, Belgium, Denmark, Finland, Germany, Ireland, Luxembourg, The Netherlands, Sweden and the United Kingdom (Figure 5.5). Furthermore, another twenty-one localities had an EI=1, thirteen had an EI=2, ten had an EI=3, seven had an EI=4, three had an EI=5, one locality an EI=6, and one had an EI=7. The highest EI obtained was EI=8 for the Las Palmas Island of Spain (Figure 5.6).

On a global scale the model mapped the potential distribution of the disease in all the countries where CBS has been reported, except for Taiwan and Bhutan (Figure 5.7). In all of these countries, the El values were favourable for the establishment of CBS with values higher than 20 (at least at one locality), except for China (maximum El=19) and Swaziland, (maximum El=13). The model also reflected the lack of climatic suitability of some areas where the disease is currently known to be absent. All of the localities in Chile, had an El=0, with the exception of one locality with an El=1. Likewise, all the localities in California had an El=0, with the exception of one locality with an El=1.

5.4.2 Validation

The EI values of 537 meteorological localities within CLIMEX were statistically compared to the known occurrence of CBS at that locality. This was done using the Kappa statistic, a value that measures the agreement of categorical data (Landis & Koch, 1977). The 537 localities included 70 localities where the citrus host was probably present and 437 localities where the presence of the citrus host is not known. When the kappa value was caluculated for these groups of localities it was equal to one for both groups, indicating that the CLIMEX model was in exact agreement with the current true occurrence of CBS at those localities.



Figure 5.5 — The potential geographical distribution of Citrus Black Spot in Europe as predicted by the CLIMEX model. The Ecoclimatic Index (EI) is proportional to the radii of the circles. Locations indicated with an x represent those where the EI amounts to 0.

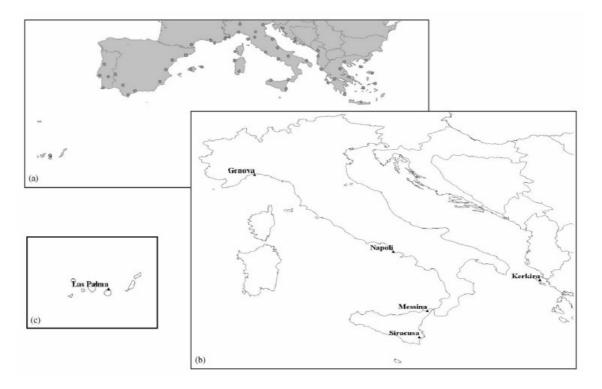


Figure 5.6 — Localities in Europe indicating: a) All localities with Ecoclimatic Index (EI) values lower than eight, b) Those localities with an EI value greater than four and c) The Las Palmas Island of Spain (EI=8).

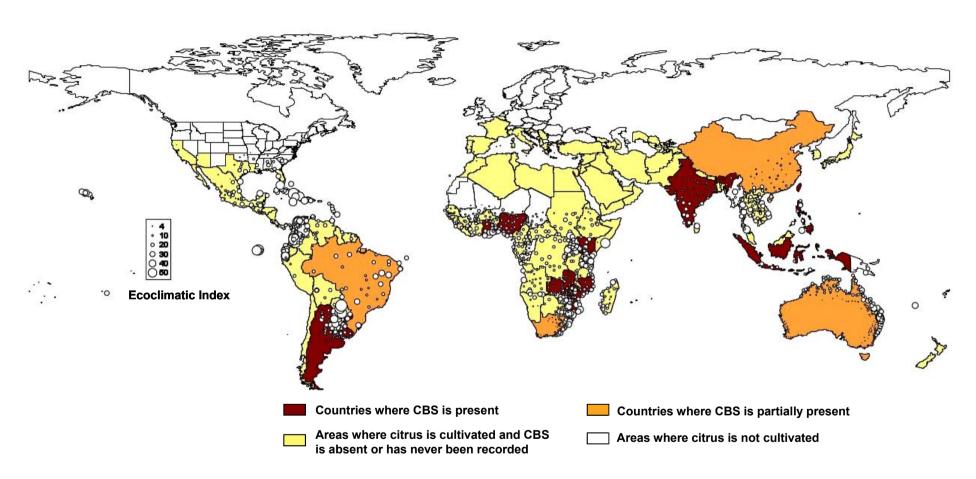


Figure 5.7 — The potential geographical distribution of Citrus Black Spot world-wide as fitted by the CLIMEX model. The Ecoclimatic Index (EI) is proportional to the radii of the circles.

5.5 Discussion

The model successfully describes the current known distribution of the pathogen around the world and indicates that climate appears to provide an effective barrier to CBS establishment. However, in SA (Figure 5.3) and Australia (Figure 5.4), some localities where CBS has never been recorded were predicted to have climates suitable for the occurrence of CBS. This may be because the host and/pathogen are absent or the presence of the disease has not been reported in the literature. In any event, the climates at these localities may be suitable for the establishment of CBS, and these results should serve as an early warning for the expansion of citrus production into the areas demarcated climatically suitable for the establishment of CBS.

On a global scale (Figure 5.7), the model mapped the potential distribution of the disease in all countries where CBS has been reported, with the exceptions of Taiwan and Bhutan. Only two climate data localities for Taiwan were available within the meteorological database of CLIMEX. Thus, the exclusion of Taiwan by the model may be because specific weather data points did not fall within citrus production regions of the country, where CBS may potentially occur. There were also no weather data localities for Bhutan within the meteorological database of CLIMEX (See Appendix A). Furthermore, these results support reports on the absence of CBS in Japan (Kotzé, 1996) and New Zealand (Sutton & Waterson, 1966), since there were no localities in either of these countries with favourable EI values.

All localities in the south-western Western Cape and inland parts of NSW where CBS is absent, despite the introduction of the pathogen, had EI values smaller than or equal to four. Since the outbreak of a plant disease requires the presence of a pathogen, a susceptible host and favourable climatic conditions (Booth et al., 2000a), the inability of a pathogenic species to establish in an area where both the pathogenic species and susceptible host are present may be attributed to unfavourable climatic conditions. Therefore, it is reasonable to conclude that an EI value equal to, or lower than four reflects climatic unsuitability for establishment of the CBS.

This study suggests that the climate of EU countries is unsuitable for establishment of the CBS disease-causing organism. However, five localities in Europe had an EI value higher than four (Figure 5.6). Three of these localities had an EI=5, namely Kerkira (Corfu) in Greece, and Napoli and Siracusa (Sicily) in Italy. One locality had an EI=6, namely Messina (Sicily, Italy), one locality, Genova had an EI=7 and the Las Palmas Island of Spain had an EI=8. Kerkira, Siracusa, Messina (Sicily) and the Las Palma Islands are island localities. Napoli and Genova are the only sites with an EI value greater than four that are on the European mainland and citrus is not cultivated at either of these localities (Anonymous, 1992), so the risk of introduction of CBS to commercial citrus growing operations appears to be very low. Furthermore, citrus trees require minimum temperatures above 7°C to survive (Davies & Albrigo, 1994; Spiegel-Roy & Goldschmidt, 1996; Srivastava & Singh, 2002). At both these localities, average temperatures below 7°C are experienced for two to three months of the year, indicating that citrus trees, the hosts of CBS, are unlikely to survive at these localities.

University of Pretoria etd – Paul, I (2006)

Whereas climatic unsuitability alone can provide a good indication that the risk of an organism establishing in a region is low, evaluating the phytosanitary risk posed by trade must combine the climatic suitability of a given region with a multitude of other risk mitigation considerations through a PRA. Standardized guidelines for PRAs have been developed by the International Plant Protection Convention (1996).

The importation of citrus into the EU is restricted to fruits and seeds and excludes any other vegetative citrus material such as leaves. Fruit are thus required to be free of peduncles and leaves (European Union, 2000c). It is known that only conidia and no ascospores (primary inoculum source) are associated with CBS-infected fruit, and together with the outcomes of this climatic modelling exercise, the risk of CBS introduction and establishment in the EU as a result of commercial trade in fresh citrus fruit, even from CBS infected areas, appears negligible.

The climate in some citrus producing countries near Europe such as Egypt and Turkey are also unsuitable for the establishment of CBS. Dry conditions restricts the survival of the species in Egypt while, in Turkey, cold temperatures are detrimental to the persistence of the species. Likewise, the El values in Israel and Morocco were always ≤ 4 except for one locality in Morocco, Larache (El=5) and two localities in Israel namely Gaza (El=5) and Haifa (El=6). Availability of moisture restrict the potential occurrence of the species in Israel and in Morocco climatic conditions are mainly too cold and too dry.

Several important citrus producing countries where the disease has not yet been reported have climates suitable for the establishment of CBS. This is especially true for the USA (Florida and Texas), which is second only to Brazil in citrus production, and Mexico, the world's largest producer of limes. Localities in these two countries frequently had EI values of above 20 with the highest values obtained in Tampa, Florida, USA (EI=28), at Brownsville, Texas, USA (EI=34) and at Progreso, Mexico (EI=36). Citrus is produced commercially at all of these localities. Other citrus producing countries that had climates suitable for disease establishment included Colombia, Cuba, Ecuador, Vietnam and Thailand.

It is important to realise that the values for the EI as provided by CLIMEX is not an absolute value and should be interpreted in a comparative or relative manner (Worner, 1988). CLIMEX parameter sets are not overly sensitive and small changes in the parameter set do not change the outcome appreciably. Because the total response of the species to climate is of interest, this information is valuable in the absence of more detailed studies and can be derived from relatively incomplete data.

CLIMEX analyses only consider the effects of climate on the species, therefore the output should be interpreted with caution. Decision-making processes should take account of the potential effects of competition from other species and human influences (such as effective control methods and irrigation) within the areas where CBS is predicted to occur (Worner, 1988).

5.6 References

- Agricultural Pests Act (1983) Act number 36 of 1983. Directorate of Plant Health and Quality. South African National Department of Agriculture, South Africa.
- Anonymous (1992) Citriculture in Italy today. In Proceedings of the International Society of Citriculture, pp. 1-12, Italy.
- Baayen, R. P., Bonants, P. J. M., Verkley, G., Carrol, G. C., van der Aa, H. A., de Weerdt, I. R., van Brouwershaven, Schutte, G. C., Maccheroni Jr., W., Glienke de Blanco, C. & Azevedo, J. L. (2002) Nonpathogenic isolates of the citrus black spot fungus, *Guignardia citricarpa*, identified as a cosmopolitan endophyte of woody plants, *Guignardia mangiferae* (*Phylosticta capitalensis*). Phytopathology, 92, 464-477.
- Barkley, P. (2003) Personal Communication. Formerly New South Wales Agriculture, Camden, Australia.
- Benson, A. H. (1895) Black spot of the orange. Agricultural Gazette of New South Wales, 6, 249.
- Bonants, P. J. M., Carroll, G. C., de Weerdt, M., van Brouwershaven, I. R. & Baayen, R. P. (2003) Development and validation of a fast PCR-based detection method for pathogenic isolates of the Citrus Black Spot fungus, *Guignardia citricarpa*. European Journal of Plant Pathology, 109, 503-513.
- Booth, T. H., Jovanovic, T., Old, K. M. & Dudzunski, M. J. (2000a) Climatic mapping to identify high-risk areas for *Cylindrocladium quinqueseptatum* leaf blight on eucalypts in mainland South East Asia and around the world. Environmental Pollution, 108, 365-372.
- Booth, T. H., Old, K. M. & Jovanovic, T. (2000b) A preliminary assessment of high risk areas for *Puccinia psidii* (Eucalyptus rust) in the Neotropics and Australia. Agriculture, Ecosystems and Environment, 82, 295-301.
- Brasier, C. M. (1996) *Phytophthora cinnamomi* and oak decline in southern Europe. Environmental constraints including climate change. Annales Des Sciences Forestières, 53, 347-358.
- Brodrick, H. T. (1969) Physiological studies with *Guignardia citricarpa* Kiely. DSc.Thesis, University of Pretoria.
- CABI/EPPO (1998) *Guignardia citricarpa*. Distribution maps of quarantine pests for Europe, Wallingford, U.K. CAB International.
- Calavan, E. C. (1960) Black spot of citrus. Californian Citrograph, 46, 4, 18, 22-24.
- Cobb, N. A. (1897) Letters on the diseases of plants: black spot of the orange. Agricultural Gazette of New South Wales, 8, 229-231.
- Cook, A. A. (1975) Diseases of Tropical and Subtropical Fruits and Nuts. Hafner Press, New York, U.S.A.
- D'Adamo, P., Sackman, P., Corley, J. C. & Rabinovich, M. (2002) The potential distribution of German wasps (Vespula germanica) in Argentina. New Zealand Journal of Zoology, 29, 79-85.
- Davies, F. S. & Albrigo, L. G. (1994) Citrus. CAB International, Wallingford, U.K.
- Dawson, T. (2003) Personal Communication. HortResearch, Kerikeri Research Centre, Kerikeri, New Zealand.
- Doidge, E. M. (1929) Some diseases of citrus prevalent in South Africa. South African Journal of Science, 26, 320-325.

- European Union (1998) Commission Decision of 8 January 1998 recognizing certain third countries and certain areas of third countries as being free of *Xanthomonas campestris* (all strains pathogenic to Citrus), *Cercospora angolensis* Carv. et Mendes and *Guignardia citricarpa* Kiely (all strains pathogenic to Citrus). Official Journal of the European Community, L 015, 21/01/1998, 41-42.
- European Union (2000a) Final report of a mission carried out in Brazil from 3–7 July 2000 in order to evaluate the pre-export inspections on citrus fruits originating in Brazil and imported to the European Union, Report: DG(SANCO)/1180/2000-MR (final). European Commission Health & Consumer Protection Directorate-General.
- European Union (2000b) Report of a mission carried out in the United States from 3–10 March 2000 in order to evaluate the phytosanitary situation and pre-export inspections on citrus fruit exported to the European Union, Report: DG(SANCO)/1114/2000-MR. European Commission Health & Consumer Protection Directorate-General.
- European Union (2000c) Special requirements for import of plants, plant products and other objects originating in third countries. Official Journal of the European Community, 169, 44-45.
- European Union (2001a) Final report of a mission carried out in Argentina from 10–14
 September 2001 in order to evaluate the inspection procedures for citrus fruit originating in
 Argentina and exported to the European Union, Report: DG(SANCO)/4303/2001-MR (final).
 European Commission Health & Consumer Protection Directorate-General.
- European Union (2001b) Report of the Commission Working Group on evaluation of the Pest Risk Assessment (PRA) prepared by South Africa on Citrus Black Spot (CBS), Brussels, 24/1/2001, Report: DG(SANCO)/1114/2000-MR. European Commission Health & Consumer Protection Directorate-General.
- Everett, K.R. & Hale, C (2002) New technologies for pathogen detection and bio-security. Agricultural Science, 10, 5.
- FAO (2002) Citrus fresh and processed: Annual statistics, http://www.fao.org/es/ESC/common/ecg/28189 en CitrusCMAENbull2002.pdf
- Garran, S. M. (1996) Citrus Black spot in the North East of Entre Rios: etiology epidemiology and control. In Proceedings of the International Society of Citriculture, pp. 466-471.
- Hattingh, V., le Roux, H. & Schutte, G. C. (2000) Citrus Black Spot: Pest Risk Assessment document for the review of current phytosanitary regulations pertaining to the export of fresh citrus fruit from the Republic of South Africa to the EU, Report. Submitted to the Agriculture Commission of the European Communities by the South African National Department of Agriculture, Directorate Plant Health and Quality.
- IPPC (1996) International Standards for Phytosanitary Measures, Part 1 Import Regulations, Guidelines for Pest Risk Analysis, Report. International Plant Protection Convention (IPPC), Food and Agricultural Organization of the United Nations, Rome.
- Kiely, T. B. (1948a) *Guignardia citricarpa* (n.sp.) and its relationship to the black spot disease of citrus in coastal orchards of New South Wales. Journal of Australian Industrial Agricultural Science, 14, 81-83.
- Kiely, T. B. (1948b) Preliminary studies on *Guignardia citricarpa* (n. sp.), the ascigerous stage of *Phoma citricarpa* McAlp., and its relation to blackspot of citrus. Proceedings of the Linnaean Society of New South Wales, 73, 249-292.
- Kiely, T. B. (1949) Black spot of citrus in New South Wales coastal orchards. Agricultural Gazette of New South Wales, 60, 17-20.

- Kiely, T. B. (1970) Black Spot of Citrus. The Fruit and World Market Grower, February, 57-60.
- Korf, H. J. G., Schutte, G. C. & Kotzé, J. M. (2001) The effect of packhouse procedures on the viability of *Phyllosticta citricarpa*, anamorph of the citrus black spot pathogen. African Plant Protection, 7, 103-109.
- Kotzé, J. M. (1963) Studies on the black spot disease of citrus caused by *Guignardia citricarpa* Kiely, with particular reference to its epiphytology and control at Letaba. D.Sc. Thesis, University of Pretoria, Pretoria, South Africa.
- Kotzé, J. M. (1971) Uitwerking van droogte op ontwikkeling van swartvlek van sitrus. Citrus and Sub-Tropical Fruit Journal, August, 9.
- Kotzé, J. M. (1981) Epidemiology and control of citrus black spot in South Africa. Plant Disease, 65, 945-950.
- Kotzé, J. M. (1996) History and epidemiology of Citrus Black Spot in South Africa. In Proceedings of the International Society of Citriculture, pp. 1296-1299.
- Kotzé, J. M. (2000) Black Spot. In Compendium of citrus diseases (eds L. W. Timmer, S. M. Garnsey & J. H. Graham). American Phytopathological Society Press, St. Paul, Minnesota, U.S.A.
- Kotzé, J. M. (2002) Personal Communication. Private consultant and Citrus Black Spot research co-ordinator for Citrus Research International, Pretoria, South Africa.
- Kuramoto, T. (1981) Citrus Diseases in Japan. Japan Plant Protection Association.
- Landis, J. & Koch, G. G. (1977) The measurement of observer agreement for categorical data. Biometrics, 33, 159-174.
- Mabiletsa, P. (2003) Republic of South Africa, Citrus Semi-Annual 2003, Report: SF3017. Global Agriculture Information Network.
- McFadyen, R. C. & Skarratt, B. (1996) Potential distribution of *Chromolaena odorata* (Siam Weed) in Australia, Africa and Oceania. Agriculture, Ecosystems and Environment, 59, 89-96.
- McOnie, K. C. (1964) Source of inoculum of *Guignardia citricarpa*, the Citrus Black Spot pathogen. Phytopathology, 54, 64-67.
- McOnie, K. C. (1965) Source of infection for black spot of citrus. South African Citrus Journal, 378, 5,6,9.
- McOnie, K. C. (1967) Germination and infection of citrus by ascospores of *Guignardia citricarpa* in relation to control of black spot. Phytopathology, 57, 743-746.
- Pivonia, S. & Yang, X. B. (2004) Assessment of the potential year-round establishment of soybean rust throughout the world. Plant Disease, 88, 523-529.
- Robertson, M. P. & Palmer, A. R. (2002) Predicting the extent of succulent thicket under current and future climate scenarios. African Journal of Range & Forage Science, 19, 21-28.
- Snowdon, A. L. (1990) A colour atlas of post-harvest diseases and disorders of fruit and vegetables. Volume I: General Introduction and Fruits. Wolfe Scientific Ltd., Barcelona, Spain.
- Spiegel-Roy, P. & Goldschmidt, E. E. (1996) Biology of Citrus. Cambridge University Press, Cambridge, U.K.
- Srivastava, A. K. & Singh, S. (2002) Citrus: Climate and soil. International Book Distributing Company, India.
- Sutherst, R. W. & Maywald, G. F. (1985) A computerised system for matching climates in ecology. Agriculture, Ecosystems and Environment, 13, 281-299.

- Sutherst, R. W., Maywald, G. F., Bottomley, W. & Bourne, A. (2003) CLIMEX v2, User's Guide. Hearne Scientific Software, Melbourne, Australia.
- Sutton, B. C. & Waterson, J. M. (1966) *Guignardia citricarpa*. Commonwealth Mycological Institute, descriptions of pathogenic fungi and bacteria No 85. The Eastern Press, London and Reading, U.K.
- Timmer, L.W. (2005) Personal Communication. Proffesor, University of Florida, U.S.A.
- Tyson, J. (2003) Personal Communication. HortResearch, Auckland, New Zealand.
- USDA/APHIS (2002) Importation of fruits and vegetables proposed rules 7 CFR Parts 300 and 319 [Docket No. 02–026–1], Vol. 67, pp. 61547-61564. United States of America Federal Register.
- Van Staden, V., Erasmus, B. F. N., Roux, J., Wingfield, M. J. & van Jaarsveld, A. S. (2004) Modelling the spatial distribution of two important South African plantation forestry pathogens. Forest Ecology and Management, 187, 61-73.
- Vera, M. T., Rodriguez, R., Segura, D. F., Cladera, J. L. & Sutherst, R. W. (2002) Potential geographical distribution of the Mediterranean fruit fly, *Ceratitis capitata* (Diptera: Tephritidae), with emphasis on Argentina and Australia. Environmental Entomology, 31, 1009-1022.
- Wager, V. A. (1949) The occurrence of the black spot fungus in citrus areas of South Africa. Farming in South Africa, 24, 367-374.
- Wager, V. A. (1952) The black spot disease of citrus in South Africa. Union of South Africa Department of Agriculture, Science Bulletin No. 303, 1-52.
- Whiteside, J. O. (1965) Black spot disease in Rhodesia. Rhodesian Agricultural Journal, 63, 87-91.
- Whiteside, J. O. (1967) Sources of inoculum of the black spot fungus, *Guignardia citricarpa*, in infected Rhodesian orchards. Rhodesia, Zambia and Malawi Journal of Agricultural Research, 5, 171-177.
- Worner, S. P. (1988) Ecoclimatic assessment of potential establishment of exotic pests. Journal of Economic Entomology, 81, 973-983.