

## Climate Change Influence on Disease Control Patterns in the Okanagan Tree Fruit Industry: A Monitoring Tool for Growers

Funding for this project has been provided by the Governments of Canada and British Columbia through Growing Forward 2, a federal-provincial-territorial initiative. The program is delivered by the Investment Agriculture Foundation of BC.

Opinions expressed in this document are those of the author and not necessarily those of the Governments of Canada and British Columbia or the Investment Agriculture Foundation of BC. The Governments of Canada and British Columbia, and the Investment Agriculture Foundation of BC, and their directors, agents, employees, or contractors will not be liable for any claims, damages, or losses of any kind whatsoever arising out of the use of, or reliance upon, this information.

DELIVERED BY

FUNDING PROVIDED BY









# Climate Change Influence on Disease Control Patterns in the Okanagan Tree Fruit Industry: A Monitoring Tool for Growers

### Canadian Agricultural Services CAS

W. J. McPhee Tree Fruit Consultant Bsc., MSc., PhD Plant Pathologist

January, 2017

#### **Acknowledgments**

Funding for this project was provided in part by the Governments of Canada and British Columbia through the Investment Agriculture Foundation of BC under Growing Forward 2, a federal-provincial-territorial initiative. The program is delivered by the BC Agriculture & Food Climate Action Initiative.













#### **Disclaimer**

Opinions expressed in this document are those of the author and not necessarily those of the Governments of Canada and British Columbia. The Governments of Canada and British Columbia, and their directors, agents, employees, or contractors will not be liable for any claims, damages, or losses of any kind whatsoever arising out of the use of, or reliance upon, this information.

Introduction	5
Project Rationale:	
Understanding and predicting disease patterns	5
Methods Used	6
Project Structure (Segments)	6
Input Sources (Segment 1)	7
Study Area (Segment 2)	8
Areas of Evaluation (Segment 3)	9
Pathogen Characteristics Evaluation (Segment 4)	10
Data Entry/Compilation (Segment 5)	11
Data Analysis (Segment 6)	11
Identification of Patterns and/or Changes (Segment 7)	12
Summary of Key Data	13
Evaluation of the Growing Days Accumulation Data	13
Isolate Distribution	15
Isolate distribution frequency by region	16
Isolation and Identification of Imports	18
Fungicide Efficacy Testing	19
Alternaria Isolates	20
Botrytis Isolates	21
Monilinia	23
Growth Rates	24
Growth Rate Variability at Three Temperatures	24
Specific Examples	27
Sporulation Patterns	34
Tree Cankers and Sporulation	38
Spore Germination	39
Morphology	42
Tree Cankers	43
Root Disorders	45
Soil Microbiology	48
Tree Assessments	49
Possible next steps	51
Conclusions	51

#### Introduction

The general areas of investigation for the project were fruit rots, cankers and soil pathogens within orchards and through the supply chain, including an assessment of some of the physical characteristics that relate to the aggressiveness of disease potential. The main objective of the project was to connect horticultural and pathological data to a geographical mapping system via computer programming to determine the uniformity, stability and potential shifts resulting from natural variability within pathogen populations initiated by climate change. The elements of the project are outlined below in Figure 1.

#### Project Rationale: Understanding and predicting disease patterns

From the late 1970's to the present, there have been significant changes in disease patterns within local tree fruit populations. For example, in the late 1970's there was great concern about the market losses in stone fruits, particularly cherries, because of the fruit rot disease Brown Rot. However, today Brown Rot is less common and only occurs as frequently as Botrytis rot and Alternaria rot. Similarly, from the late 1980's onward, Cytospora Canker was rampant throughout cherry orchards in the Okanagan Valley, but was rarely seen on apple. Today the disease is common on apple trees and appears to be much more aggressive than it was in the 1980's and 90's. There also appears to be a shift in species from mainly C. cincta to C. leucostoma.

These are two examples from many disease pattern-shifts that have occurred within the industry. These shifts may, in some cases, be related to management and control strategies. However, it is also plausible that changes in a climate pattern have had some influence on these shifts and, if this is the case, it raises the question of how the potential for a shift to occur can be monitored. The main purpose of this research is to develop and test a system designed to help visualize changes that occur over time and correlate these to weather patterns and weather pattern changes. When temperature patterns shift, degree-day accumulation patterns also shift, which is more indicative of changes in climate patterns. By coordinating climate changes with pathogen characteristics, it is possible to predict pathogen aggression and adaptation capabilities.

The software ArcGIS is a geographic information system capable of layering data on a map to show spatial relationships. As the database expands, and if the information in the database reflect the characteristics of pathogens, then interpretation of the data and elucidation of patterns can provide meaningful indications of pathogen shifts. For example, there are obvious morphological variation among Botrytis colonies collected from the field, variations in fungicide efficacy between field isolates, documented shifts in sporulation patterns, and growth rate differences show a variation in optimal growth temperatures for isolates. These and other pathogen characteristics are reported in more detail in the Summary of Key Data section. If and how these characteristics are influenced by climate change can be key to control strategies. Although dealing directly with pathogens and their characteristics constitutes the basic emphasis of this project, these pathogens are so intimately tied to general horticulture and to the structure of the industry that many "spin-offs" have been included. The main pathogen growth rates, chemical control patterns, sporulation, germination and visual morphology. These key issues will be discussed in detail in the Summary of Key Data section. Spin-off effects that are important to interpretations regarding climate change impact are also discussed.

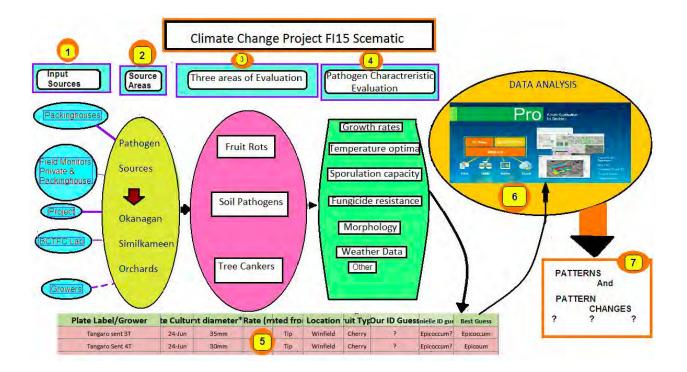
#### Methods Used

All methods used were standard procedures and are detailed in Appendix 4. A main concern in selecting methods was to strive for dependable, yet efficient techniques that could be applied in the field or in simple office-lab settings, and require only inexpensive equipment. This was an important part of the project concept because ease of monitoring is an important factor in expanding the database, which is necessary to see pattern changes over time. Independent monitors with limited time and facilities should be able to carry out many of these tests themselves.

#### Project Structure (Segments)

Figure 1 summarizes the structure of the project in seven segments. The approach for tracking climate impact is based on analysing pathogen characteristics that may be dependent in some way on temperature regimes. Each segment within the figure is designed to support the final goal: to predict shifts in disease patterns that require modification of control strategies.

Figure 1: Project structure



#### Input Sources (Segment 1)

The collection of information on the characteristics that define isolates within the chosen parameters is essential to the basic hypothesis that changes in climate pattern can shift isolate distribution patterns by making environmental conditions more or less favourable to individual variants. Under this umbrella are the two segments, "input sources" and "source areas." The input sources are important to the success of the project because the more data, the more likely the data will enable patterns to be determined. Data collection during the project provides a baseline of data, but long term data collection depends on segments of the industry to "buy in" and continue data collection.

In the early stages of the project the "select core" of data came from six orchards within the Okanagan and made up the bulk of the pathogen-characteristic studies. As the project became more well known the source of samples expanded. The core was maintained as a base for the more detailed information input such as soil microbiology and soil nutrient analysis. These latter two areas fit into the category loosely described as spin-offs and will be discussed in detail in the Summary of Key Data section.

Because field monitoring input is voluntary and depends on the interest generated by the project, participation could not be guaranteed. Initially most enthusiasm came from the private (non-co-op) field people, which might be expected since this group does not have access to any professional support groups. However, as the power of the information feed became evident, and through the efforts of Dr. Hirkala and her staff, participation by the co-op field staff has become routine. Aside from adding information to the database, the project has created an information flow that did not exist before the project began. It is important to note that independent growers (and their field representatives) have not been privy to the real time data, a disparity that needs to be corrected and may require setting up a separate matrix database.

Growers did not widely participate in data collection, but did respond when approached individually. They were interested in specific problems directly related to their own daily management. The resulting interactions allowed for an opportunity to address the necessity of being aware of the potential impact climate change may have on their daily routine. Over the course of the project numerous direct contacts were made with growers, extension personnel, packing plant owners and professionals to deliver the message and tie the project goal to needs (see Appendix 1 for the contact table).

The BC Tree Fruits Cooperative (BCTFC) lab, under the direction of Dr. Danielle Hirkala, was the main supplier of isolates as the lab already routinely monitors post harvest rot problems. Dr. Hirkala is also overseeing a replant program that brings in soil for analysis and runs routine assessments of canker disease problems in orchards. These services, made available by the Winfield lab, were a major supplier of data to the matrix and are key to the long term feasibility of the program.

#### Study Area (Segment 2)

The project drew pathogen samples mainly from the Okanagan/Similkameen valleys from a variety of in-the-field sources where disease inoculum can survive in discarded fruit, in ground litter, in soil, on tree surfaces, on alternate hosts, etc. To track change, it is important to address a broad spectrum of pathogen sources over a wide geographical area as well as evaluate variation in pathogenic aggression within that spectrum.

The Okanagan/Similkameen valleys (see figure 2 below) consist of numerous microclimates. It is well known that the level of correlation between weather and infection

varies significantly throughout the region, and that infection periods, which influence the potential for disease development, also vary from location to location. The project accessed daily recordings of temperatures, moisture, rainfall, soil temperatures, etc., for several weather stations throughout the Okanagan/Similkameen valleys in order to compare pathogen patterns with weather data.

Kamloops Vernon Kelowna Penticton

Figure 2: Map of Okanagan and Similkameen valleys

#### Areas of Evaluation (Segment 3)

As noted, the three main areas of evaluation for the project include fruit rots, soil pathogens and tree cankers. Location details for each main area were also noted. For

example, whether the isolated pathogen was associated with the fruit, the tree or the soil would be noted. Fruit rot pathogens were randomly picked from the orchard floor and from trees from the entire valley. Soil samples and wood rot cankers, because of the difficult nature of

handling and isolating these, were more selectively chosen and in every case each isolate Was given a set of coordinates to pinpoint the sample location.

#### Pathogen Characteristics Evaluation (Segment 4)

Segment 4 identifies the characteristics that will be monitored as the project continues beyond 2017. These characteristics will help to identify expressions of genetic variation within the populations they represent. Single spore isolates were purposely avoided because broader population characteristics were felt to be more representative of natural conditions.

It was not possible to examine all of these characteristics for each of the numerous isolates collected. Isolate characteristics fall into five categories:

- 1. **Growth Rate** Growth rate at room temperature (22°C) data has been collected on all isolates tested for fungicide efficacy screening, while determining low temperature (2°C) and high temperature (30°C) growth rates was carried out during the summer of 2017.
- 2. **Sporulation Patterns** The ability to produce large spore numbers is a survival plus for pathogens. Determining the sporulation precocity variation is therefore an important characteristic that might correlate with temperature patterns.
- 3. **Spore Germination** The germination capacity of the spores produced is important to understanding if fungicide tolerant isolates have the capacity to germinate normally and hence retain their capacity to infect.
- 4. **Visual variations** in colony form is a distinguishing factor that can be used to determine distribution patterns.
- 5. **DNA patterns** may also distinguish variations in distribution patterns among isolates. Although this was not a specific goal for the project it is a logical extension for assessment in the future.

These pathogen characteristics are important to survival potential, therefore investigating them is an effective way of predicting potential changes in population dynamics, and the need for changes in control strategies. In addition, climate data extracted from the 25 weather stations in the project area was important to this segment. Access was provided to the weather stations through the BCTFC lab in Winfield. Pattern changes expressed in weather station data (over time) is an important element of assessing possible linkages to changes in disease and distribution pressure.

Average temperatures, rainfall and soil temperatures and moisture can be used to represent the area surrounding the station to describe the "weather". Graphs of weather station data are available for all regions in Appendix 3.

#### Data Entry/Compilation (Segment 5)

Data was recorded and stored in Excel files (referred to as the matrix). A large amount of data on pathogen characteristics is stored within the main matrix. Any characteristic that can be tied to a particular isolate may be important. Information such as: where the isolate was found in the orchard, i.e., in the tree, on the ground, on fruit etc.; the sample date; and the GPS coordinates, are all potentially significant in separating pathogenicity potential and are separated in the matrix.

The original (Excel-based) matrix was expanded upon by Dr. Hirkala as an extension tool and contains all the information gathered through the commercial lab system in addition to project data. Although this is beyond the original scope of the project, it expands the long term sustainability of the program, making it more applicable to technology transfer and adopting it as a standard tool utilized by all packinghouse field personnel and independent extension specialists.

#### Data Analysis (Segment 6)

The ArcGIS system was installed on the BCTRF computer in 2016 as part of the database analysis segment of the project. It was to be managed by the BCTF diagnostic lab in Winfield, BC and made available to both BC Tree Fruit and independent field staff. Dr. Hirkala was trained on the program and set it up in the BCTF system. An app was created

for the BCTF extension group, which allows them to access their diagnostic information in real-time. However, this access was limited to the packinghouse group, contrary to the original intent of making the program accessible to both coop and independent staff.

ArcGIS software is capable of scanning the data to look for characteristic patterns that correlate to geographical position and to climate. This project included access to several weather stations strategically located throughout the Okanagan-Similkameen valleys that can be downloaded into the database. The computer program can handle huge volumes of data and there is an opportunity to continue to expand the database on an ongoing basis to increase the accuracy of detecting climate related patterns.

The mapping program ArcGIS for Desktop Basic can be utilized to draw together one or multiple characteristics from the Excel files and to place these on an area map, which allows the spatial data to be viewed, to create layered maps, and perform basic spatial analysis. The ArcGIS mapping system is set up like an access database. To generate maps to show spatial relationships queries need to be generated that can be pictured on a map. The program requires fixed data in order to make comparisons, which then allows a comparison between graph points to be made, such as average monthly temperatures, rainfall etc. The

data can be mined endlessly over time to look for changes in patterns. While this capability has been demonstrated, a detailed analysis of the data to evaluate for weather data relationships has not yet been completed. This requires extraction of average temperatures and rainfall, which is pending. With this information queries can include temperature maximums in specific time periods rather than yearly degree day accumulation data.

#### Identification of Patterns and/or Changes (Segment 7)

The final conclusions of the research initiated with this project are from the elucidation of patterns that are imbedded within the data. For example, the following questions may be answered through the data generation and analysis:

- Are there distinct pathogen-variant-distributions within the Okanagan area? For example, are certain variants more prevalent in the north than in the south?
- Are these variants responding to different control strategies?
- Will a shift in climate pattern result in a shift in variant distribution or change the variants themselves?

Such questions can only be asked and possibly answered with the current approach if the

database is large enough to be statistically meaningful. It is important to note that the existence of patterns and potential control-strategy changes for pathogens are, at this point, a hypothesis that needs to be investigated. Variants within the pathogen populations are real. However, how they may be impacted by climate change shifts still remains to be seen.

#### Summary of Key Data

#### Evaluation of the Growing Days Accumulation Data

Thirteen of the area weather stations were assessed for growing day accumulation (GDA) over the decades where data was available. This data (Appendix 3) shows a stepwise increase in the degree day accumulation value over the past decades for all regions which is consistent with the hypothesis that the regional climate is changing. Using the ArcGIS mapping program this information can be used to link and display data groups to geographical region based on heat units (GDA). Running queries through the ArcGIS program to find pathogen distributions patterns should, over time, when superimposed on weather data, demonstrate if the shift in pathogen distribution is weather influenced.

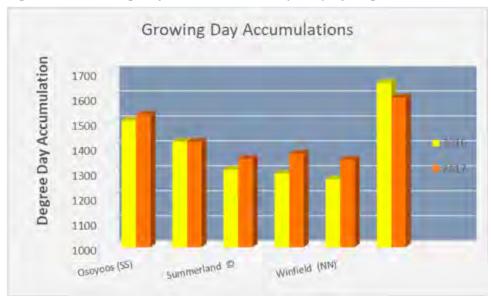
The data in Appendix 3 shows: decade averages from the start up time of the station to the present, a decade average, and an overall average. This demonstrates a distinct trend toward increased growing-days accumulation over time for each area and although this is not new information, regional values can be used to illustrate the distribution of pathogen characteristics in relation to the growing-day accumulation on a yearly basis.

Table 1 defines the locations of each region used in Excel data sorting. The graph below the table shows a comparison between 2016 and 2017 growing day accumulations by region. It also illustrates clearly that the Keremeos/Cawston region is significantly hotter for both years than the Okanagan Valley regions.

Table 1: General description of the regions considered in this study

Region Designation	Region Description	General Coordinates Latitude Boundary		Average Growing Day Accumulation by Region		
				2016	2017	
SS	The Osoyoos north area running from the US border to Rd 19	49	49.1108	1507.8	1531.7	
S	Running from Rd 19 north to Vaseau Lake	49.1108	49.2521	1421.3	1422.0	
С	OK Falls to just south of Peachland	49.2521	49.8051	1309.7	1353.5	
N	Westbank north to the Kelowna Airport	49.8051	49.9809	1294.4	1374.1	
NN	South end of Ellison Lake to Vernon north	49.9809	50.3378	1271.8	1350.45	
К	Keremeos Cawston region	49.223	49.1344	1659.3	1598.9	
CR	Creston Valley					
IMP	Isolates from outside the interior region, includes Fraser Valley to Mexico					

Figure 3: Growing Day Accumulations (GDA) by region



#### **Isolate Distribution**

The total number of fruit rot samples isolated during the project is 750 (not including the soil and root isolates, which are reported below). Figure 4 and 5 show the general frequency of fruit-rot-pathogen isolations, giving a general picture of the potential disease pressure in the Okanagan.

The potential for importing isolates from other areas that may have different environment requirements are shown on a separate graph. This data is further broken down in Figure 6 to examine if there are regional differences. The Okanagan and Similkameen valleys were divided into six regions for this purpose (regions with distinct differences in micro climates). The general coordinates are shown in Table 1.

Initially samples were randomly collected out of the field from the entire Okanagan Valley to build a sample base of pathogens, and they mainly represented fruit rot fungi. As the project progressed, more fruit rot samples were obtained from industry storages, field monitors, growers, imported fruit at retail outlets, etc., which broadened the scope of the data matrix and defined the distribution of pathogens and their individual growth characteristics. The canker and soil samples were more selective and were drawn mainly from diseased trees or soil associated with trees in a state of decline. The isolate mix shown in Figure 4 is from local orchards and from imported fruit samples and demonstrates the variation in the field in 2016.

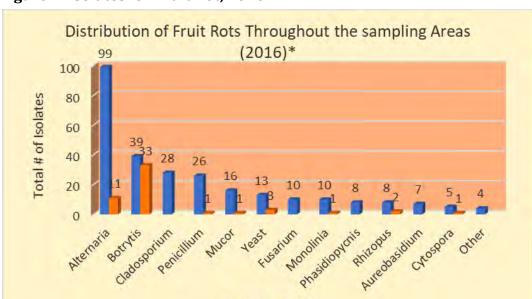
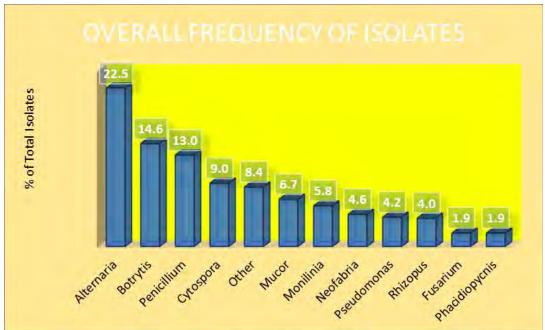


Figure 4: Isolates for Fruit Rot, 2016

Figure 5 shows the distribution of isolates over 2016 and 2017. This shows Alternaria is the number one isolate over the two years. Alternaria can be a major market problem for fruit shipped to distant markets yet is not managed as a key pathogen by the industry. It attacks flower parts at blossom, is favoured by wet weather, but is not identified as an issue until late season or post harvest unless specifically looked for. There are no registered products for this disease, which is a dangerous position to be in if the pathogen influence is increasing and little is being done to generate a strategy to address it.

Figure 5: Total frequency of isolates

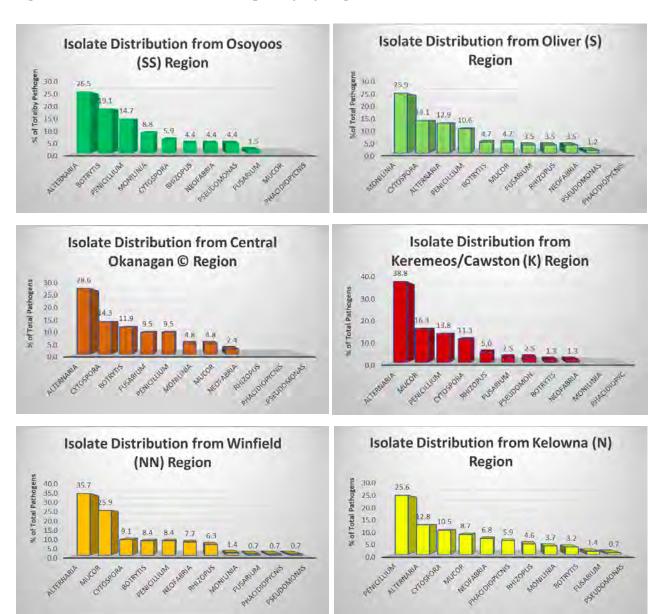


Samples containing spoiled fruit were also pulled from retail stores in order to compare isolates coming from outside the area. A summary of those isolates is shown in Figure 7. The most common isolate imported on cherries and berries was Botrytis rot, also a common rot problem in local BC orchards.

#### Isolate distribution frequency by region

The following series of graphs look at the isolate distribution frequency by region. These maps are based on information extracted from the matrix of all data as of end of 2017. In general, Alternaria is the most prominent fruit rot isolated over the project period and causes significant problem in markets.

Figure 6: Isolate distribution frequency by region



Assessing the pathogens by geographical region shows Alternaria is the most common isolate in four of the six regions. Uniformity of pathogen distribution runs through the Okanagan and Similkameen valleys. For regions S and N, Alternaria is at significant levels. Penicillium is the prominent isolate in Region N, and Monilinia in Region S. An explanation for the prominence of Penicillium in region N is that there could be a skew in the sample selection. A number of the samples were post harvest samples from the packinghouse line. A more precise analysis with the ArcGIS program should be capable of addressing biases as the data set increases.

Where Monilinia is the prominent isolate (region S) there are two possible explanations. First, samples from this area had a bias toward peach samples so Monilinia levels could be biased because of sample dynamics. High levels of Monilinia could also be related to micro climate to some degree. The area from where these samples were taken tends to be wetter than than average for the region. Fifty percent of all the Monilinia isolated were found in region S.

#### **Isolation and Identification of Imports**

Figure 7 shows the most common pathogens in imported berries, coming mainly from California and Mexico, is Botrytis. Alternaria was also isolated in significant numbers from imported fruit. Distribution of pathogens world wide is a part of international agricultural trade. It is prudent to assess these imported pathogens to assess the potential for survival and aggressiveness that could make them more adaptable to higher temperatures.

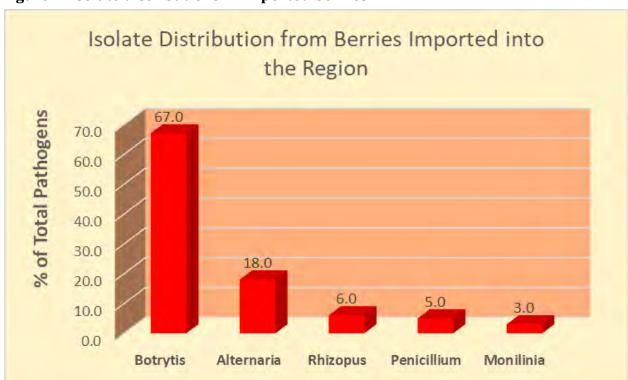


Figure 7: Isolate distributions in imported berries

#### **Fungicide Efficacy Testing**

It has been common over the years to see significant variation in response to fungicide treatment by different isolates. When assessing pathogen populations there is a gradient of impact by chemical controls within any population. The range from sensitive to resistant generally follows a decline slope with variants positioned along that slope.

Stability of a given characteristic may be difficult to define, but from a population sense, there is evidence that survival of any one characteristic in nature may be dependent on a second characteristic. An example is the suppression of an individual within the population with a resistant gene, because of its inability to reproduce as fast as those individuals with low resistance.

The shift from a sensitive to a resistant population represents a stable state for that population once reproduction efficiency is superseded by a more significant selection factor. The level of resistance is usually step wise, each step stable under defined environmental and abiotic conditions. Although there are individuals within the population that fit along the sensitive/resistant slope, one basic characteristic of the population can be defined by the optimum growth rate of the population. This may be quite important because, if the temperature pattern changes in such a way as to favour one of the suppressed individuals within the population, then the basic characteristics of the population could change. Based on agar plate tests, examples of the efficacy variation between fungicides, and an indication of the variation from isolate to isolate, are shown in the Figures 8 and 9.

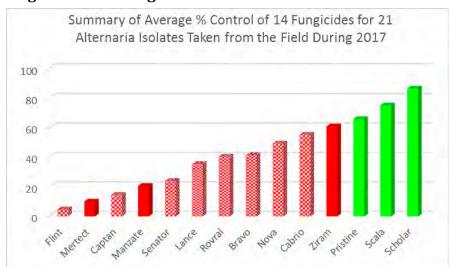


Figure 8: Average control of fungicides for Alternaria isolates

19

Figure 8 illustrates the average control by 14 fungicides for 23 field isolates of Alternaria. Although growers believe that Brown Rot-controlling fungicides also control other fruit rots, it is clear that only three of the Fungicides listed in the guide for control of Brown Rot also control Alternaria.

#### Alternaria Isolates

Figure 9 is an example of the variability in field control by a key fungicide for the major pathogen, Alternaria. It illustrates the potential weakness of using a single pesticide repeatedly within the season. What we are looking for within this project are inherent characteristics in the vulnerable group that can be influenced by climate shift to become more aggressive or more dominant within the total population. In that light, Figure 9 indicates 42.2% of the isolates tested were capable of surviving the control product.

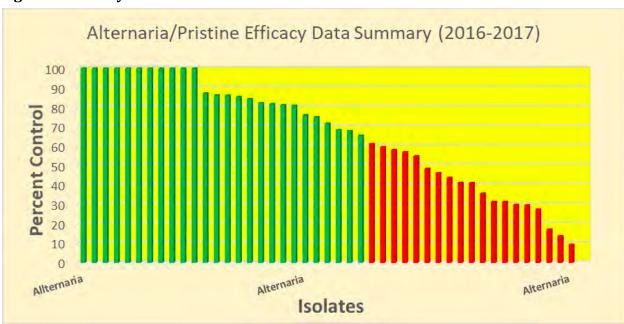


Figure 9: Efficacy of Pristine on Alternaria isolates

Figure 9 shows the individual control by Pristine for each of 45 isolates. Pristine is considered a very effective product by growers yet 35% of the field isolates tested were not controlled at a commercially acceptable level. The green and amber bars represent the average pathogen control for all fruit rots and for Alternaria respectively. Figure 10 illustrates the complete failure of the fungicide Flint against this pathogen. In fact, this chemical significantly enhances the growth of some isolates.

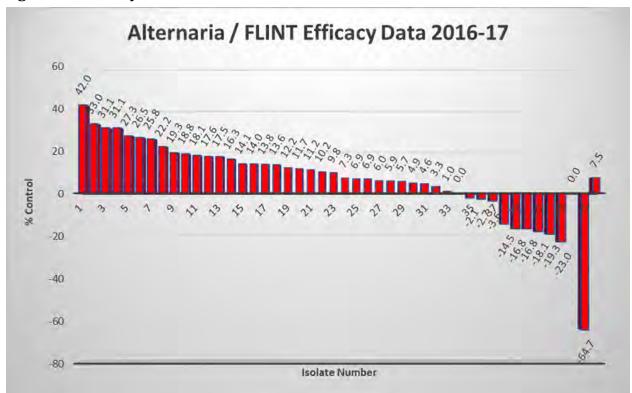


Figure 10: Efficacy of FLINT on Alternaria isolates

The variations in response between isolates as shown in the figures and discussed above, support the goal in the original proposal to define variations in the survival and pathogenicity of the pathogens in the field and track climate shifts.

#### **Botrytis Isolates**

Botrytis was another common fruit rot pathogen found throughout the test regions. It has been identified as a major cause of fruit damage, though the industry has had very little support for registration of control methods. The general recommendation is that this pathogen is controlled by Brown Rot sprays. The assumption is that growers control botrytis while applying regular brown rot sprays.

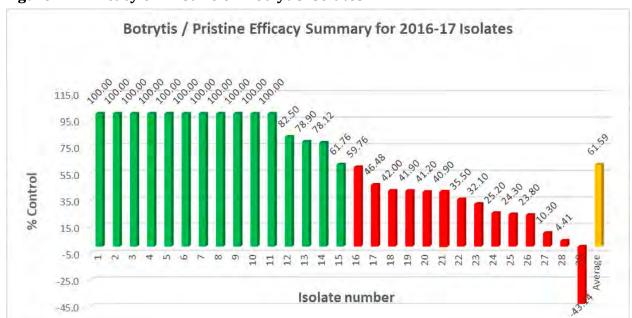


Figure 11: Efficacy of Pristine on Botrytis isolates

However, 50% of the botrytis randomly isolated are not controlled effectively with Pristine as expected. Captan is a popular and widely used fungicide for control of brown rot and is used under the assumption that it covers other fruit rots, such as Botrytis. Figure 12 demonstrates that the control of Botrytis with Captan is very poor.

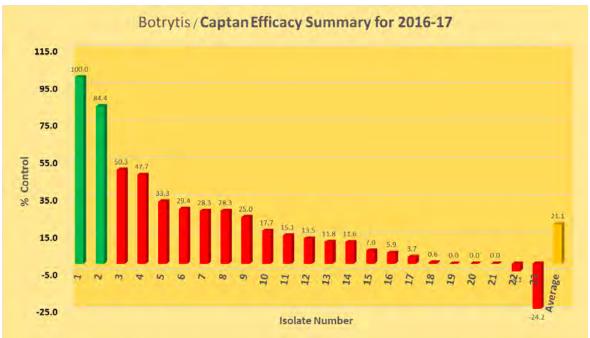


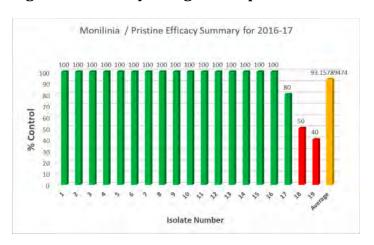
Figure 12: Efficacy of Captan on Botrytis isolates

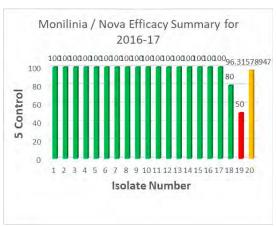
More significantly, 40% of the isolates with very poor control with Captan are the same isolates that are not controlled with Pristine. This creates the potential to favour Pristine resistant strains by using Captan.

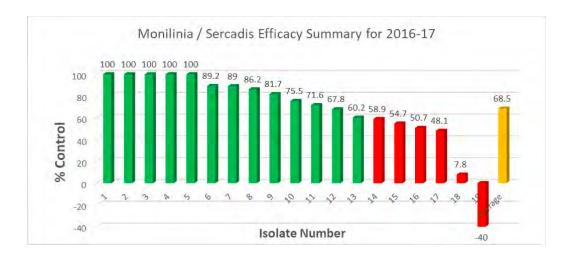
#### **Monilinia Isolates**

The resistance data for Monilinia illustrates the effectiveness of fungicides registered specifically for the pathogen as shown in Figure 13. All products registered for Monilinia (brown rot) performed similarly well. Figure 14 shows the one fungicide tested against Monilinia that screened poorly, Flint, a non-registered product. Its profile is more like those for Alternaria and Botrytis.

Figure 13: Efficacy of registered products to control Monilinia







23

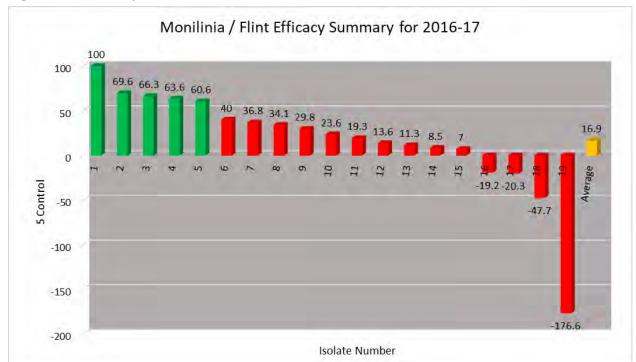


Figure 14: Efficacy of Flint for Monilinia

The figures above and those in Appendix 5 explain the potential for fungicides to fail to varying degrees in the field. With so much chemical tolerance in these random isolates, shifting the population makeup and changing the efficacy pattern may just require a shift in growth rate optimal for isolates with inherent resistance. It has been demonstrated that growth rate can be a major factor in an individual's dominance within a population under defined conditions. Further evidence of how climate change could accommodate shifts in competitiveness may be found in the growth rate trials below.

#### **Growth Rates**

#### **Growth Rate Variability at Three Temperatures**

In this project, isolations were made from groups of spores removed from sporulating wounds, from the flesh of rotting fruit, from bark, wood chips, soil, etc. Single spore isolates were purposely avoided as not representative of the normal variation of the pathogen population existing within any field sample. The intent was to duplicate the variability that exists naturally.

An initial sample of the growth variations across all pathogens for three distinct temperatures (2°C, 22°C and 32°C) are illustrated in the figures below.

The growth rate of a particular strain or variable can have a major impact on the survival of that particular strain. As pointed out earlier, pathogen populations in the wild consist of a mixed genetic make up of individuals. The population dynamic, composed of this mix of individuals, remains stable within the confines of the current environmental pressures.

To use one parameter as an example, a slow-growing component will remain a minor portion of the individuals within the population unless the environmental conditions shift in its favour. If the limiting factor is growth-rate, the dynamics will remain stable. If the relative growth-rates are related to temperature, then a shift in temperature to favour the slower growing individual can give that individual the competitive advantage it needs to dominate the population.

In this segment of the project the goal was to determine if individual isolates had distinct growth differences at low or high temperature. Table 2 shows the pathogens tested and the total number tested for each. There are three broad categories in Table 2 covering fruit rot fungi, tree canker fungi and root rot fungi.

Table 2: Fruit Rot, Wood Rot and Root Rot pathogens

Pathogen genus	No. of samples	Pathogen genus	No. of samples	
Fruit rot pathogens		Wood rot pathogens		
Alternaria	251	Neofabria	103	
Penicillium	170	Phacidiopycnis	32	
Cytospora	126	Coriolus	15	
Monilinia	86	Nectria	10	
Mucor	83	Aureobasidium	3	
Botrytis	80			
Rhizopus	36	Root rot pathogens		
Cladosporium	33	Fusarium	29	
Venturia	5	Cylindocarpon	4	
Yeast	5			

Pathogens in each temperature category display a range of growth rates and overall, the best growth rates are at moderate temperatures. Growth is generally reduced at the two temperature extremes. Figure 15 is for a series of Alternaria isolates and illustrates the typical pattern for that pathogen. It also shows that growth is infinitely variable. Figure 16 is a plot for all isolates covering several fruit rot pathogens and exhibits the same general shape curve, which indicates that there is a continuous range of growth rates within the general population. This might be expected since there are a number of different genuses being represented, however, the pattern is consistent for each independent pathogen in the fruit rot group.

Range of Growth Rates for all Alternaria Isolates at 22oC, 32oC and 0oC

20,00

15,00

5,00

6,00

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

1,000

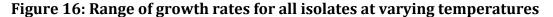
1,000

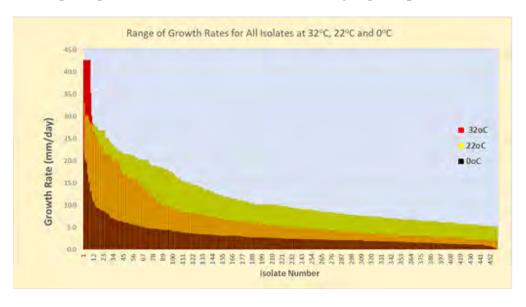
1,000

1,000

1

Figure 15: Range of growth rates for Alternaria isolates at varying temperatures





The left side of Figure 15 and 16 show that the growth rate of some isolates was stronger but, as demonstrated below, the relative temperature response for any one isolate is variable itself. Some isolates grow relatively poorly at moderate temperatures and exceptionally well at one or both extremes.

This type of variation, which in some cases can be described as aggressiveness, lends itself to climate change influence. Figures 17 to X demonstrate this and support the premise that a pathogen can be suppressed at one temperature by the more rapidly growing competitors, yet have an advantage over those same competitors at another temperature.

This same general pattern of infinite variability shows up for all pathogen characteristics including fungicide resistance, sporulation, germination, etc. The variation in characteristics, especially in growth rate at various temperatures, indicates a strong potential for shifts in dominance among isolates.

#### **Specific Examples**

Three regions, (SS, S, and N) are represented in the group of Alternaria isolates tested for growth rate comparisons. These graph patterns reveal little by their basic distribution shape. However, at 32°C there is one isolate that stands out with a growth rate almost double the growth rate of the next isolate, in Figure 17. This same isolate is the slowest growing isolate from this group at 22°C, in Figure 18, thus revealing a possible competitive advantage in the relatively hot south Okanagan and Similkameen where extended hot spells will be most severe, but a relatively non competitive disadvantage at moderate temperatures.

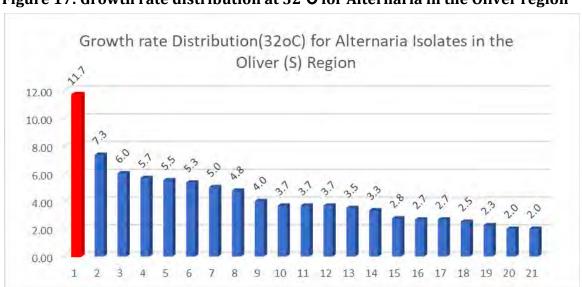


Figure 17: Growth rate distribution at 32°C for Alternaria in the Oliver region

27

Growth Rate Distribution (22oC) for Alternaria Isolates for the Oliver
(S) Region

16.0

16.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

10.0

1

Figure 18: Growth rate distribution at 22°C for Alternaria in the Oliver region

A second example of the potential influence of climate change shows up in screening the Monilinia isolates in the same region. One particular isolate (red bars) is the most aggressive at moderate temperature, shown in Figure 19, but not at higher or lower temperature extremes (shown in Figure 20 and 21). At lower temperatures it is a relatively poor competitor when compared to the other isolates, particularly isolate #1 (stippled red bar at far left). Higher temperatures will give this isolate a competitive advantage among this particular group of isolates but it is easily out-competed at lower temperatures.



Figure 19: Growth rate distribution at 22℃ for Monilinia

Figure 20: Growth rate distribution at 32°C for Monilinia in the Oliver region

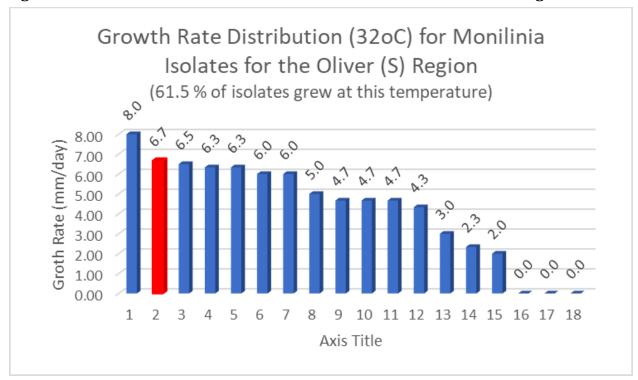
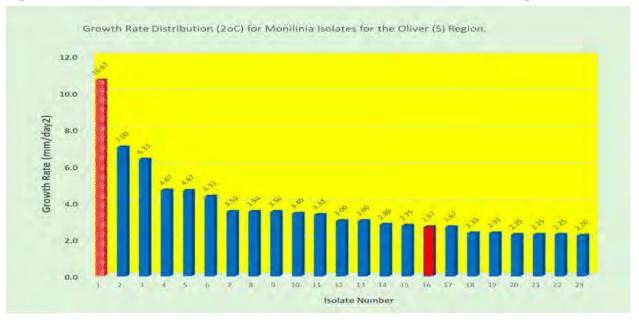


Figure 21: Growth rate distribution at 2°C for Monilinia in the Oliver region



A third example for potential pattern shift within a pathogen group stands out in the Keremeos area. In the south, the fruit rot Mucor has a minor presence. In the Keremeos area with a higher growing day accumulation value, Mucor is the second most common

fruit rot pathogen isolated. The relative growth rates for the Mucor isolates at the three temperatures, 32°C, 22°C and 2°C, are shown in Figure 22. Figure 22 shows that seven Mucor isolates clearly have a growth rate advantage at 32°C.

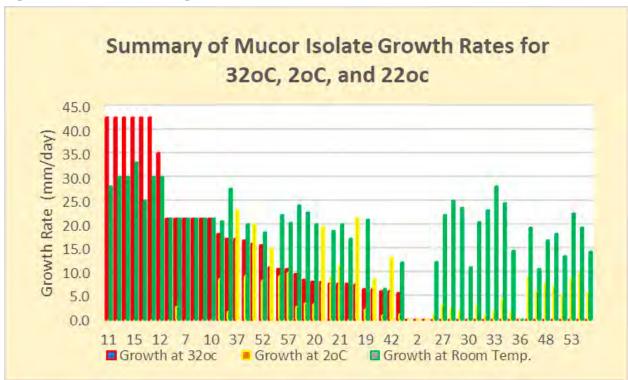


Figure 22: Mucor isolate growth rates for 32°C, 22°C and 2°C

When isolate growth rates at the three temperatures are separated from the main graph, (Figure 23-25 are plotted on the same scale for comparison), the growth characteristics of the group of isolates is more clear. At moderate temperatures (22°C) the majority of the Mucor isolates grew well. At the two extremes they varied. They generally grew poorly at 2°C, but few did exceptionally well at higher temperatures, although at the higher temperature only 62% survived. In general, higher temperatures would favour a few isolates giving them a competitive advantage.

Figure 23: Summary of Mucor isolate growth rates at 32°C

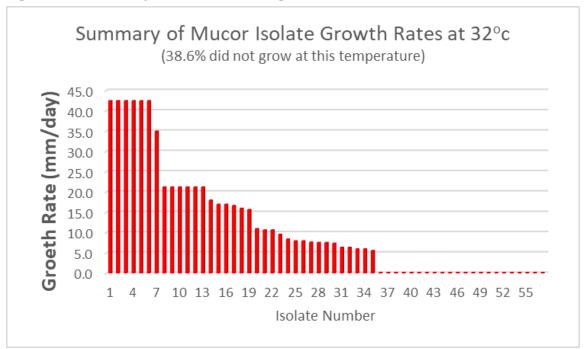
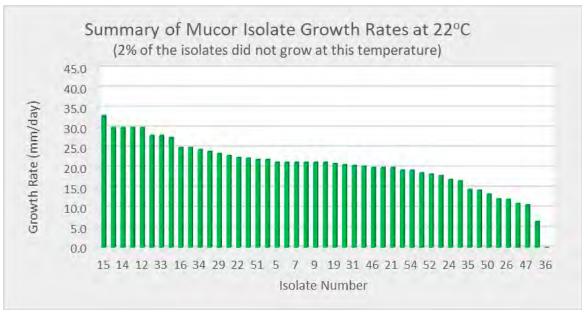
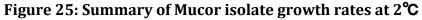
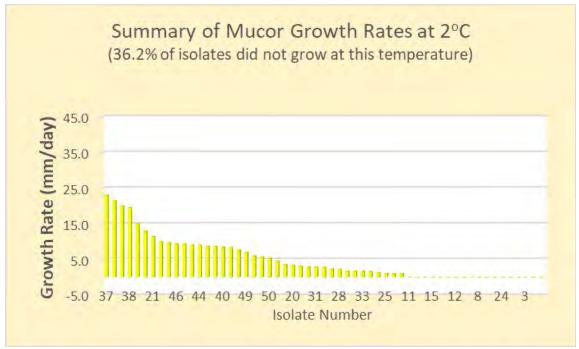


Figure 24: Summary of Mucor isolate growth rates at 22°C



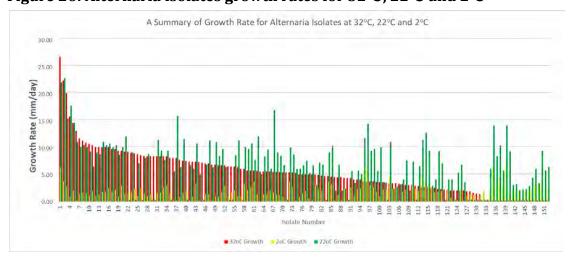




Mucor is usually associated with storage rot, but has been an orchard problem in the Similkameen Valley in the past on peaches and apricots. As data mounts in the database, the relationship between frequency and temperature maximums can be assessed.

Alternaria was the most frequently isolated fruit rot pathogen in the Keremeos area. A similar look at this pathogen (Figures 26-29) tells a different scenario with regard to growth at different temperature extremes.

Figure 26: Alternaria isolates growth rates for 32°C, 22°C and 2°C

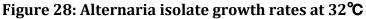


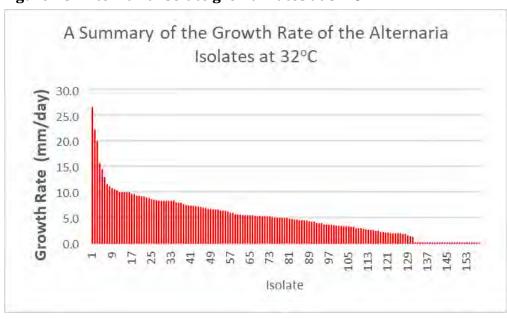
This combination plot (Figure 26), sorted highest to lowest for the 32°C group, accentuates the isolate growth rates at the three temperatures. Again, when the three temperature plots are separated (Figures 27-29), the relative impact of temperature is clearer. They suggest that the overall best growth is at 22°C, there is no isolate at the extreme temperatures that stand out as far as growth rate is concerned and a shift in climate wouldn't have a significant impact on the relative distribution of these isolates.

A Summary of Growth Rates for Alternaria Isolates at 22°C Sorted Highest to Lowest

30,0
25.0
20.0
15.0
10.0
5.0
10.0
5.0
Isolate

Figure 27: Alternaria isolate growth rates at 22℃





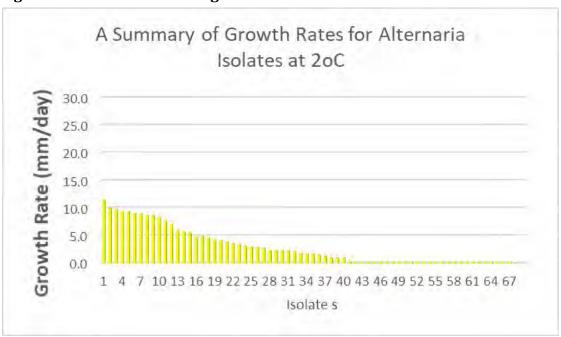


Figure 29: Alternaria isolate growth rates at 2°C

Taken together it appears a shift in climate to hotter temperatures could enhance the advantage of Mucor over Alternaria in the area.

#### **Sporulation Patterns**

The sections on fungicide screening, geographical distribution and growth rate evaluations presented above indicate opportunities for pathogen variations to respond to temperature shifts and for their distribution patterns to change. However, other characteristics of pathogens can be instrumental in their survival and aggressiveness. The ability to reproduce infection units (spores) and their survival are essential to their success in the field. Sporulation is a characteristic associated with pathogens that can impact their survival and pathogenicity.

As a monitoring tool, sporulation by fungi can be utilized more readily than other techniques. Fruit rot fungi, such as Alternaria, Botrytis, Monilinia, Penicillium and Mucor, produce masses of surface spores in the field and in the laboratory, and are the pathogens used for this segment of the project. A total of 198 tests were carried out and are summarized in Figure 30, which illustrates that spore production is dramatically different between pathogens. The average spore production on PDA plates (yellow bar at the end of the group) in Figure 30 demonstrate this. This again reinforces variability in

functional characteristics and is more evidence for potential competitive advantage.

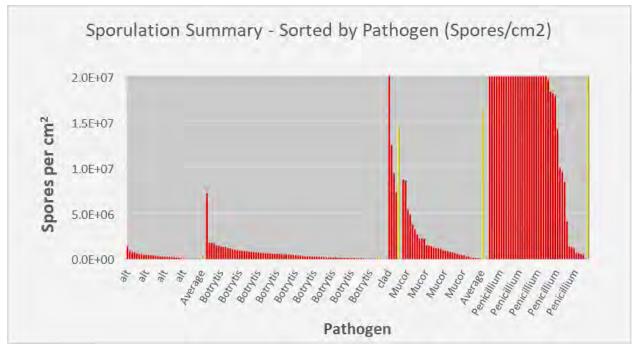


Figure 30: Sporulation by pathogen type

Evidence of variability within populations is directly related to the fungicide influence. Figures 31 and 32 show potential areas where normal sporulation in the field can be altered by the use of fungicides. Sporulation density is variable for Penicillium isolates but spore density (SD) in the isolates on unamended PDA do not appear to be overshadowed by the colonies that originated as transfers from fungicide amended PDA Plates.

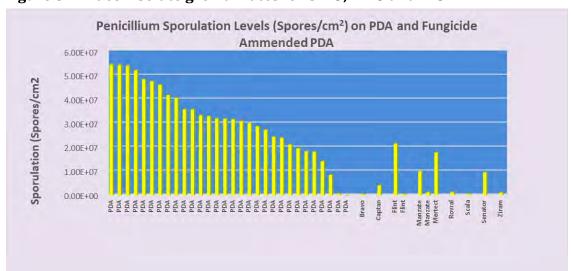


Figure 31: Mucor isolate growth rates for 32°C, 22°C and 2°C

With the exception of Mertect and Flint, colonies growing on the fungicide amended plate sporulated relatively poorly and exhibited no capacity to out-compete non-treated isolates. Sporulation by colonies grown on Mertect and Flint did not appear to be suppressed, but neither did they show any aggressive spore production.

Figure 32 shows some variability between isolates, however, in general each fungicide stimulated sporulation to some degree in most isolates even though linear growth for most of these isolates was retarded by the fungicide. Enhanced sporulation is one way for a pathogen to compensate for reduced growth.

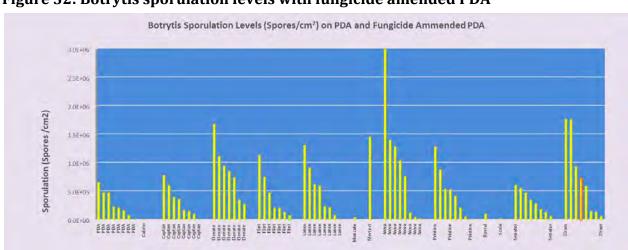


Figure 32: Botrytis sporulation levels with fungicide amended PDA

Another example of character change from outside influence is the data on Elevate treated Botrytis isolates (Figure 33). Sporulation for each isolate was enhanced by this fungicide. The average spore density more than doubled as a result of treatment, a factor which increases the isolates presence in the general population. Analysis of growth control by the fungicide Elevate (Figure 34) shows that sporulation enhancement would compliment poor growth control for the seven isolates controlled by the fungicide.

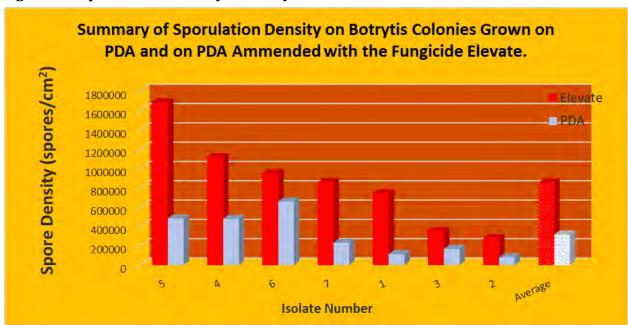
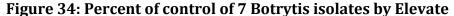
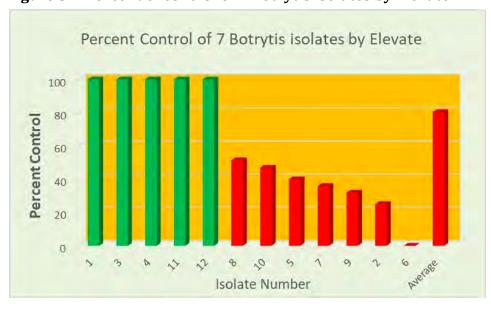


Figure 33: Sporulation density on Botrytis colonies with Elevate treatment





Looking specifically at isolate 7, growth control is 36%, but sporulation has more than tripled. This influence on survival capability could become an added advantage. There is no temperature-growth rate data for this specific isolate, but if favoured by a shift in weather pattern it would have an inherent advantage in the field under an Elevate control program by virtue of its increased sporulation.

#### **Tree Cankers and Sporulation**

Cytospora, a pathogen which causes tree cankers, is less convenient to collect and bring to the lab. However, they do sporulate heavily in the orchard and can be utilized directly from the field as a measure of the impact of weather. A shift from Cytospora *cincta*, being the prevalent species in the valley, to the species *leucostoma* may be influenced by the gradual increase in average valley temperature.

These two species have a distinctly different growth optimum, which introduces temperature as a significant factor in competition. C. *leucostoma* grows best at 25 - 30°C, while C. *cincta* grows best at 18-20°C. C. *leucostoma* is also able to grow at 37°C.

Figure 35, summarizing the Cytospora data, shows a complete continuum of growth rates for this pathogen. The pattern shows on average, the growth at 32°C and 22°C are similar. However, in general, the 22°C temperature supports most isolates better than the 32°C temperature and all isolates grow poorly at the 2°C temperature. For isolate 67, its growth rate values are shown on the graph for each temperature. Growth is fastest at 32°C, but relatively poor at both 2°C and 22°C compared to all other isolates. This isolate would have a distinct disadvantage at all temperatures except 32°C and possibly higher.

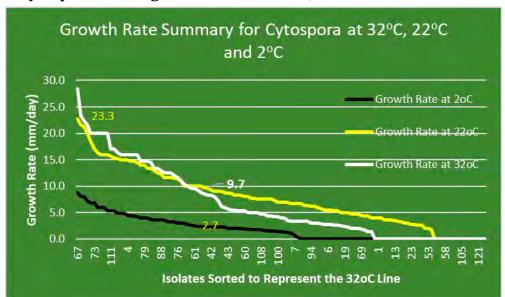


Figure 35: Cytospora isolate growth rates for 32°C, 22°C and 2°C

# **Spore Germination**

Because fungal diseases are spread mainly by spores, and probability of an infection occurring from an individual spore is very low, spore production is overwhelming in order to compensate. However, increases in spore production are only meaningful if those spores are viable. A colony of penicillium on a discarded apple on the orchard floor or on a single cherry left hanging in a tree, for example, can harbour millions if not billions of spores and the germination test is one assessment of the viability of those spores. Germination capability varies with each isolate and affects the isolates relative competitiveness. Suppression of germination by any means would reduce competitiveness.

Any modification in pathogen pattern attributed to climate change will likely be in concert with other modifications in the general characteristics of that pathogen. Defining germination efficiency is therefore important to the "process" associated with climate change impact on fruit diseases. Sixty isolates from the fruit rot group have been examined for germination efficiency and came from all the areas shown in Table.

Table 3: Environmental conditions and number of isolates by study region

Region Designation	Region Description	General C Latitude E	oordinates Boundary	General Environment	# isolates /region	
SS	The Osoyoos north area running from the US border to Rd 19	nning from the US border		Hot, dry	68	
S	Running from Rd 19 north to Vaseau Lake	49.1108	49.2521	Same as SS with more rain, later maturity	85	
С	OK Falls to just south of Peachland	49.2521	49.8051	Semi moderate from SS and S	42	
N	Westbank north to the Kelowna Airport	49.8051	49.9809	Wetter than south, fewer growing days than NN	219	
NN	South end of Ellison Lake to Vernon north	49.9809	50.3378	Wetter and colder than N	143	
К	Keremeos Cawston region	49.223	49.1344		80	
CR	Creston Valley				9	
IMP	Isolates from outside the interior region, includes Fraser Valley to Mexico					

Information on the isolates subjected to a germination test are provided in Appendix 7. Figure 36 shows the results for four fruit rot pathogens: Alternaria, Botrytis, Monilinia and Penicillium. The comparison between the four shows a distinct pattern difference. The general pattern seen is an "all-or-nothing" for three of these pathogens, but is distinctly different for Alternaria.

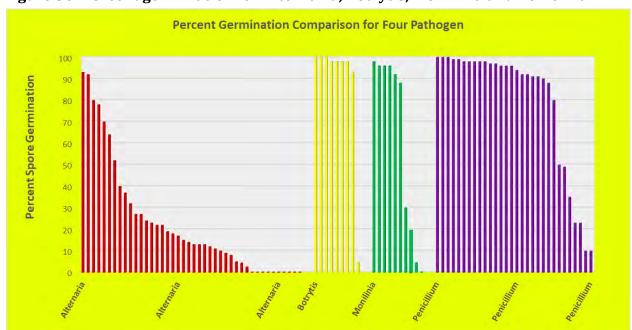
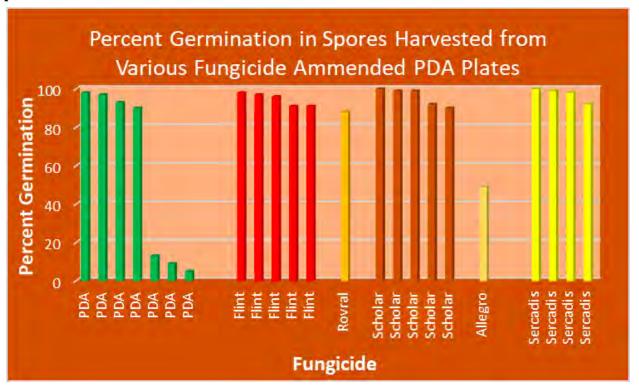


Figure 36: Percent germination for Alternaria, Botrytis, Monilinia and Penicillium

For Alternaria there is a small percentage of isolates to the left that may have an advantage based on spore germination, while for Botrytis, Monilinia and Penicillium the percent germination seems to fit into two distinct categories, strong or weak. Generally, spores, even those stored for extended periods, germinate readily when water is available and temperatures are moderate.

Alternaria spores are more complex than the spores of the other three pathogens shown. Alternaria sporulation (Figure 36) is much less dense, thus, it is difficult to rationalize that temperature or humidity would change the relative competitiveness for this pathogen. However, Alternaria is most competitive at blossom. It establishes within the early development of the fruit and remains dormant or develops slowly and internally until later in the season. Therefore, in the case of Alternaria, a climate advantage may be related more strongly to the impact of external temperature and moisture on the infection process than on any aggressive characteristic that competes against other pathogens.

Figure 37: Percent germination in spores harvested from fungicide amended PDA plates



The figures in the Appendix indicate that this pattern is consistent for all groupings of the data with the exception of the central region (Penticton, Summerland and Westbank). As mentioned above, the four general areas (south, central, north and Keremeos) represent four general climate types, i.e., warm dry south, the central area with milder temperatures, and the cooler, wetter north and the hot dry Similkameen. Generally, the germination pattern shows two groups, one with poor germination (averaging 20% and a second with over 80% germination). The isolates out of the central area (Penticton to Westbank) are an exception, having a percent germination distributed across the entire range from 10% to 90+%. This may be an area where the most concentrated variations in microclimates exist. The general pattern, showing two plateaus, is consistent for each of the other four geographical areas suggesting no climate influence on this characteristic (see Appendix 1).

# Morphology

Past experience has shown that there are a number of morphologically different isolates associated with the various fruit rot pathogens common to tree fruits. These have been documented in the valley for the pathogen Botrytis, but establishing their distribution during the fungicide efficacy testing, and an attempt to type these isolates according to their differences, has been time consuming. Over the years we have documented close to two dozen stable variants of Botrytis *cinerea*.

Appendix 2 is a small sample of the photos taken for the two key fruit pathogens Botrytis *cinerea* and Alternaria spp. showing the variation in morphology. Different variants can be assigned a numerical value and entered into the database so values can be plotted on a regional map. Establishing the distribution of these types within the region to show regional patterns is another marker to follow when attempting to assess changes in population make-up.

The two separate pictures illustrate one colony with heavy sporulation, which develop in a ring pattern, and a second colony with no visible sporulation, but with numerous sclerotia scattered about the surface of the agar. They are hard, usually dark, masses of dormant hyphae with differentiated rind and medulla and thick, hard, cell walls which permit survival in adverse environments.

In Appendix 2, the sclerotia appear as black spots on the surface of the Botrytis plates. The production of sclerotia is variable both in number and pattern and has proven in the past to be a distinguishing characteristic between isolates. Identification and quantification of sclerotia is an important marker for categorizing the isolates because they demonstrate a measurable level of survival.

This morphology is easily recorded by field monitors with minimum lab facilities. Spores can be transferred directly from infected fruit to PDA plates and kept aseptic long enough for classification without contaminant interference.

### Tree Cankers

Up to this point the discussion has been mainly about defining fruit rot pathogen characteristics. The other two main areas of assessment were tree cankers and root pathogens. The first of these, cankers, has been a neglected area of orchard disease study in this region. Identification of these diseases over the two-year study period has been accomplished mainly through the work of Dr. Hirkala at the tree fruit diagnostic lab in Winfield. Most isolates were collected by field monitors, brought to the lab and recorded in the data bank associated with the ArcGIS program. The one exception was Cytospora canker that was actively collected as part of the project sample program and subjected to characteristic assessments, such as growth rate, sporulation, etc.

As mentioned above, there are two species of Cytospora commonly found in the area. They are C. cincta and C. leucostoma. C. leucostoma survives better at higher temperatures and has become the most common species found, a shift from the situation commonly experienced a couple of decades ago. There has been a major change in commodity varieties in both pomme fruit and stone fruit in the valley and some of the shift may be a result of this change. However, C. leucostoma now appears to be much more aggressive than it was in the past, and much more aggressive than the C. cincta that was common in cherries several decades ago.

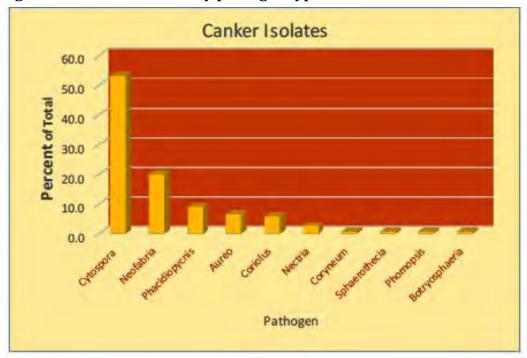


Figure 38: Canker isolates by pathogen type

Eighty-one Cytospora samples were collected over the two years and were evenly distributed between north and south. This was the most common canker found and evaluated. The other cankers were identified as part of the routine diagnostic program at BCTF lab (Figure 38).

More than any other individual pathogen, Cytospora has caused major losses to the industry through tree decline, limb death and removal. As discussed above, there is evidence that climate change may be a contributing factor. Growers and field personnel are becoming more and more capable of identifying the disease in the field and, as the distribution pattern becomes better defined, the link to sub-area microclimate will be clearer. There are other Cytospora species and other canker diseases, such as Botryosphaeria, Phomopsis, Stereum purpureum, etc. These have all been found in the area. They are relatively rare, but have the potential to have a significant impact.

Specific environmental conditions favouring one of these diseases over another are extremely variable. The approach supported in this project is to monitor pathogen distribution and look for coincidental climate related changes utilizing the ArcGIS mapping program. When pattern shifts are suggested by the data, additional information on growth characteristics can be further clues as to how permanent the change may be. Diseases such as perennial canker, Nectria canker, Coriolus canker, etc., have been endemic in the valley for decades and generally more prevalent in the north because of the wetter conditions. Cytospora canker, however, has always been more common in the south until recently. Table 4 summarizes the distribution of the isolates within the Okanagan by region and shows 64% of the canker samples came from the area of Kelowna northward, while only 24 (2%) came from the area of Oliver southward (32.8 if Keremeos is included), which is what experience has shown in the past.

Table 4: Distribution of canker isolates by region

Pathogen	SS	S	K	С	N	NN	TOTA L
Aureobasidium	1	1			7	1	
Coriolus	2	3		1	2		
Cytospora	5	11	7	6	26	17	
Botryosphaeria			1				
Nectria	1	1			2		
Neofabria	3	4	1		15	3	
Phacidiopycnis					12	1	
Phomopsis						1	
Coryneum		1					
Total number	12	21	9	7	64	12	136
Total percent	8.8	15.4	6.6	5.1	47.1	16.9	100

The table shows 64% of the canker samples came from the area of Kelowna north while only 24.2% came from the area of Oliver south.

As climate change occurs and the weather patterns change, particularly increases in precipitation, the incidence of cankers may become a serious problem. Following the incidence and distribution in relation to temperature changes is a key part of the canker segment.

# **Root Disorders**

The root system is critical to tree health and roots of all plants are subject to attack by pathogens. In the Okanagan there has been little effort to deal with this problem, so there are few guidelines for growers to follow. The potential for variations in soil microbial populations are virtually infinite.

Documentation of root pathogens in local orchards has not been well defined at the extension level. There are broad recommendations within the tree fruit production guide

for control in replanting soils, but these are critically inefficient. The decline of trees in mature and maturing orchards is widespread, causes significant loss of production, and is widely misdiagnosed. The vulnerability of the root system has been routinely seen by observation of the roots and condition of the trees, such as in Images 1 and 2.



**Image 1** (left): Often a tree at this stage of decline will contain canker disease or insect damage when examined and be diagnosed as succumbing to such. However, this tree has no above ground symptom and is clearly dying from a root disorder.

**Image 2** (right): Typical root "die-back", which is a common early symptom preceding the above ground decline symptom and a routine root evaluation to establish pathogen activity.

Root isolations, when identified to genus, are entered into the database, their frequency summarized in Table 5. Cylindrocarpon is the most common soil pathogen isolated from declining trees. Data on fungicide efficacy for Cylindrocarpon and other root pathogens are also entered into the database to be used to support treatment options.

Table 5: Root isolations and pathogen frequency

Pathogen	Frequency	Pathogen	Frequency
Cylindrocarpon	66	Unknown 1	13
Trichoderma	33	Unknown 2	6
Verticillium	17	Unknown Bla	5
Ilyonectria	10	Unknown 4	4
Alternaria	6	Unknown 3	3
Fusarium	6	Other	11

The importance of proper diagnosis in disease control strategies is obvious. The work carried out in this project was intended to document the presence (or absence) of a group of pathogens that have been suspected in the root dieback that has been observed. Soils were primarily chosen for screening based on the occurrence of decline symptoms in the orchard. Soil screening was carried out in the spring and fall in 2016 and 2017 and results entered in the database.

In the summer of 2016 further work was carried out to isolate pathogens directly from the roots. These data are shown in Appendix 7. A total of 211 isolates were successfully transferred from roots into culture and identifications attempted. The technique used was designed to allow for quick extraction of the pathogen from infected roots onto PDA plates. This could be carried out with minimal laboratory facilities. The technique is outlined in the Appendix 4.

The Okanagan is semi-arid and the tree fruit industry is dependent on irrigation. Soil moisture conditions, therefore, are more or less controlled and not likely to impact the soil microbe populations unless area precipitation increases significantly. If it does, however, it could increase the influence of the Phycomycetes in the soil. Soil temperature on the other hand does influence root development, especially in timing of root initiation. Soils tend to hold their heat, and temperature fluctuation is dampened, compared to air temperatures. This may result in higher average soil temperatures throughout the summer.

# Soil Microbiology

The importance of soil microbiology in the common occurrence of decline in fruit trees is often missed. There are significant losses in vigor due to the attack of roots by these pathogens and little has been done locally to address the problem. The main attack occurs on the feeder roots, which are so critical to the trees capacity to survive in our arid conditions. The early flush of these feeder roots is vulnerable to attack by several different pathogens. Some of the common ones identified in this area are: Cylindrocarpon, Verticillium, Fusarium, Pythium and Phytophthora.

There were 42 soil microbiological analyses carried out in 2016, and 31 in 2017 (Tables 6 and 7). The purpose of gathering this data was to establish some baseline for the distribution of certain soil pathogens in the valley. In 1960-80 there was a full-time researcher at the federal research station in Summerland working on the pathogen Phytophthora *cactorum*, which was the causal agent of crown rot. This disease was common in local orchards at that time, and the main rootstocks being used were susceptible. In the 1980's the importance of this disease was being questioned in light of the fact that root disorders were being implicated more and more.

Treatment of tree decline associated with root disease is almost non-existent. A recent biological product has been widely researched and shown to have a measurable effect on the levels of the root lesions. However, although there is some decline in soil pathogen numbers between spring and fall, there is no definitive result indicating the biological product worked to control every pathogen.

Table 6: Pathogen frequency in spring and fall, 2016

Pathogen	Spring Turffix	Fall Turffix	Spring Check	Fall Check
Trichoderma	2,000	0	7,371	800
Fusarium	28,125	60,200	24,714	103,000
Phytopthora	13,250	160	44,620	333
Rhizoctonia	2,500	6,600	19,429	3,333
Pythium	20,033	5,000	19,429	3,333
Cylindrocarpon	57,500	4,620	53,000	13,667
Verticillium	2,500	600	3,014	5,333
TOTAL Fungi	78,750	82,800	255,714	209,167

Table 6: Pathogen frequency in spring and fall, 2017

Pathogen	thogen Spring Turffix Fall Turffix		Spring Check	Fall Check
Fusarium	Not detected (Nd)	784	Nd	3,600
Phytopthora	Nd	.03	Nd	.01
Rhizoctonia	Nd	Nd	Nd	Nd
Pythium	.05	.15	.01	.2
Cylindrocarpon	57,500	4,620	53,000	13,667
Verticillium	Nd	400	Nd	1,920

As the industry shifted to higher density farming and the use of dwarfing rootstocks the association between root disorders, tree decline, and replant became more obvious. The shift from concern about crown rot to concerns about other soil pathogen that attack roots directly is finally taking place. Much of the shift is a new recognition of an old problem, a change in varieties, rootstocks and orchard management, and possibly a change in climate.

The semi-arid conditions in the region give the industry control over water supply, which has a major impact on the development of root and crown diseases. The impact of climate change, which may bring with it increased precipitation, can modify the pathogen populations in the soil. The extent of the replant problem and tree decline in all commodities in the valley is a clear indication that the problems are not well understood and are likely transitioning.

#### **Tree Assessments**

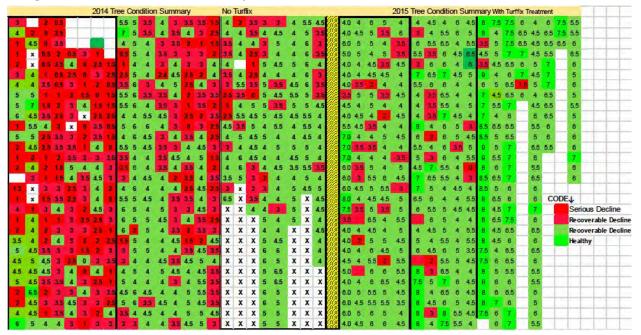
In order to assess the impact of any parameter, be it a management strategy or environmental change, it is necessary to document characteristics of the test area that will be a measurement of the impact of the change. Scoring individual tree condition within a block has been a technique used in the past, and can give a pictorial overview of a block of trees.

An assessment is made by scoring individual trees on a 1-10 scale measuring at 0.5 increments. The technique sets an upper score for the best trees in the block based on a subjective rating for their commercial acceptance. This value should be somewhere

between 7 and 10 considering 7 as the minimum commercially acceptable rating. All trees are compared to the top-level score. The value 0 represents a dead tree or missing Tree.

A single number, obtained by averaging the block scores, can go into the data summary sheet and can be searched and analysed with respect to location within the ArcGIS program. Schematic 2 clearly shows a change in general condition of a test block in the Kelowna Belgo area. The technique is time consuming but invaluable as a way to visually demonstrate the impact of a treatment or of a weather event.

**Image 3: Orchard Score for 2015/2016** 



# Possible Next Steps

A separate control centre would be ideal for the future tracking of climate change within the pathogen populations, but this would need to be supported by a neutral third-party in order to address the inherent competition between the cooperative segment of the tree fruit industry and the independents.

The project plan was to coordinate activities through the cooperative owned commercial lab, which solicits pay-for-service work from non-coop growers. However, the commitment is not feasible because of the conflict between the routine diagnostic work and the added work related to defining pathogen characteristics. Fungicide efficacy screening had an obvious and immediate impact on control strategies and could easily be justified in a routine monitoring program. Other evaluations, such as growth rates, sporulation, germination, soil microbiology, soil chemistry, weather data, etc., were activities with no short-term gains and could not be justified in a commercial setting.

The new ArcGIS computer program (and the app that allows field personnel to access the database in real time, extract specific data and download the information on a map of the area) creates an incentive to bring diagnostic problems from the field to the lab and enhances the database. Use of the ArcGIS system will continue through the Cooperative at their expense, but this will not necessarily include the characterization of the individual pathogen or relate the information to specific weather data.

# Conclusions

1) This project proposed to establish a baseline for this distribution of three key disease areas, tree cankers, general fruit rot of pome and stone fruits, and soil diseases and this was accomplished by virtue of the data in the database and the fact that data will continue to expand into the future.

Although the emphasis in data collection within this project is related to pathogens, the spin off for the tree fruit industry is the capacity to assimilate other data that is routinely collected by the various segments of the industry. Physiological disorders, such as sunburn and bitter pit in apples, firmness in cherries, and information important for replant disorder control, are examples of additional information that can be imported into the ArcGIS program and analysed in relation to climate patterns. The

significance of these spin-offs is evident in the recognition that the program has received, particularly in unifying independent growers and the cooperative.

The diagnostic reporting system initiated by the BCTF Winfield extension lab is an example of the long term viability of the program beyond the life of this project. The potential scope of on-going work is illustrated by the possibility to expand the matrix of data already accumulated.

2) The use of fungicides to control fungal infections is the primary defence growers have against diseases, but one important side effect of fungicide control is the influence these controls have on the characteristics of fungal populations. Resistance to fungicides is a common phenomenon related to population dynamics. In the lab, one characteristic that can be monitored (and that reflects a possible survival advantage) is any alteration in spore production that is consistent with the use of fungicides.

Typically, linear growth control is used to assess the efficacy of fungicides against various chemicals. However, linear growth is only one aspect of the influence a fungicide may have on the viability and morphology of a pathogen. Some of the other characteristics that are observed in these tests, besides linear growth, are pigment alteration, change in colony shape, enhancement or reduction in aerial mycelia on the colony, and colony ability to produce spores. The importance of the latter is clear. If the fungicide controls linear growth on a test plate this may be a false advantage if a second effect is to enhance the sporulation capacity of the retarded colony.

Based on the ability of pathogens to develop resistance to fungicides their potential to change due to climate changes is high. There was an attempt in the research to demonstrate this potential by describing specific temperature response patterns.

- 3) A number of stable variants of both Alternaria and Botrytis have been documented in the past, but were never captured in a registry. Once these stable variants are defined, then DNA sequencing could determine genetic variability and their distribution within the sampling area could also be determined and compared to fungicide efficacy response, growth rate, pattern etc. A record of DNA fingerprints of the various isolates would advance the trace potential of changes that are occurring. This is a logical extension of this type of approach, but is more suited to pure research than to applied extension.
- 4) The project aimed to make geographic labelling of information immediately accessible to field personnel and allow them to target control programs to specific high potential

areas for disease problems. This also facilitates storing of data in a mapping system that will allow a visual display of the distribution of the disease, its relative severity and its relationship to other factors such as microclimate, soils and management techniques. This has been completed and data can now be queried and plotted and access to the program is available in real-time to field persons.

- 5) In order to continue the work initiated with this project an ongoing commitment is required to maintain and continue entries into the database, as well as routine assessment of the data. This need is partially fulfilled by the BCTF Coop lab in Winfield that has adopted the ArcGIS program into their routine extension program. However, additional resourcing is required to maximize the value of ArcGIS system, to dedicate sufficient time/effort to data analyses and distribution, as well as allocate effort to identification and characterization.
- 6) Direct contact with growers has been considered one of the most important elements of project success and the contact made with both co-op and independent growers through the project shows the potential for groups to unify and work toward accomplishing shared objectives. A gap remains in bridging and unifying the cooperative and independent segments of the industry and this is critical for regional prevention strategies.

# Appendix 1 Contact Table

S	Summary of Individual	Contacts	Made During 2016
			ct personal contact with growers, cooperators, BCTFC specialists and independent packer/owners
			rower and independent packer contact involved discussions on climate change and the goals of
this project in	n relation to the projected clima	ite change impa	ct on orchard production.
Date	Grower Contact	Location	Reason for Visit
Jan. 4/16	M. Haithwaite	Cawston	Discuss project goals and arrange for orchard and soil sampling,
Jan. 6/16	R. Stewart	Cawston	Discuss project goals and arrange for orchard and soil sampling.
Jan. 20/16	R. Dhaliwal	Osoyoos	Discuss project goals and arrange for orchard and soil samples. Discussed financial support responsibilities.
Jan. 26/16	B. Witzke		Discussions with grower supporter in an attempt to solicite BCFGA support,
Feb. 25/16	B. Witzke	East Kellowna	Discussions with grower support person in an attempt to solicite BCFGA support. Discuss orchard and soil sampling.
		ı	
Mar. 17/16	M. Haithwaite	Cawston	Vist to initiate canker isolations in this orchard.
March 2/16	S. Tangaro	Winfield	Orchard sam[pling of soil .
March 17/16	D. Barker	Cawston	First organic grower. Discussed replant options and importance of soil temperatures relative to spring management.
April 5/16	S. Tangaro	Winfield	Visit to discuss project goals and to arrange for routine sampling.
April 6/16	G. Norton	Oliver	Root assessments and discussion with grower regarding root management.
April 6/16	J. Campbell	Osoyoos	Phone contact with grower regarding adding a soil probe to orchard weather station. Check soil temperatures
April 11/16	Rod King	Penticton	First grape grower with interest in project FI15. Collected fruit rot samples for fungicide assessment.
April 15/15	R. Dhaliwal	Osoyoos	Discussed soil temperatures in relation to root development and control strategies in valley and mountain or chards.
April 20/15	B. Witzke	East Kellowna	
April 20/15	S.Bader (for R. Bailey)	Oyama	Met with field person s. Bader and gave instructions on identifying Cytospera canker in cherry blocks
April 21/16	R. Stewart	Cawston	Discussion on soil temperature and root development. Discussed root condition after soil ammendment treatment.
May3/16	R. Dhaliwal	Osoyoos	Grower discussions and root evaluation training.
	R. Stewart	Cawston	Grower discussions on root evaluation-training and root management.
	M. Haithwaite	Cawston	Discussions on canker problems plus root evaluations.
May4/16	G. mundh	Westbank	Training on canker identification and control. Root and soil assessment.
	Halwood Orchards (S. Bader)	Oyama	Training for Shauna Bader (Halwood orchards) on canker identification and assessing root health.
May 13/16	D. Kruger	Vernon	Evaluate canker problem and collect isolates from infected wood for lab assessment.
MAY24/16	M. Haithwaite	Cawston	Assessing canker problem and collecting isolates from infected wood.
May 26/16	M. Haithwaite	Cawston	Assessing canker problem and collecting isolates from infected wood.
June3/16	G. Mundh	Westbank	Follolwup with grower and collected canker samples.
June 7/16	J. Campbell	Osoyoos	Root assessment and soil and root collection.
	G. Kunz Grapes	Osoyoos	Another grape block with decline problems. Spoke to the grower and indicated root problems.
	F. McLennan	Oliver	Discussion about project FI15. Attempt made to relate the project to current orchard management.

Appendix 1 1

### **Summary of Individual Contacts Made Sept 15-April 15**

The table itemizes orchard visits involving direct contact with growers, cooperators, BCTFC specialists and independent packer/owners within the valley. Each involved discussions on project goals in relation to the projected climate-change impact on orchard production.

within the ve	aney. Lacirinvolved discussio	nis on project goals ii	n relation to the projected climate-change impact on orchard production.
	Grower	<u> </u>	
Date	Grower Contact	Location	Reason for Visit
	D. C.		Visit farm to discuss soil amendments and treatment strategies in
Sept. 22/16	F. Guiiii	Kelowna, Belgo	
20/16	C Tangaro	Winfield	relation to soil temperatures.
Oct. 28/16	S. Tangaro	winiteta	Project FI15 discussions. Discussed decline problem its control and
Nov. 24/16	Tangaro Orchards P Gurm	Belgo	control timing in relation to soil temperatures.
Nov. 24/16	P Guilli	ветво	Discuss soil amendments and importance of early management as determined by soil
			temperatures.
		Cantacte wi	th Other Cuerne in the Industry
		Contacts wi	th Other Groups in the Industry
		Dfsianala	
		Professionals	
	Judie Steeves, BC Grower	Associate Editor	Meeting to discuss publications for magazine.
Nov. 16/16	Dr. M. Mazzola	USDA Res. Scientist	Discussions on his soil amendment research. Spring soil temperatures?
	Plant Pathologist	Wenatchee, WA	
		Dackers Own	and Durainest annumentance ato
Date	Contont		ers Project supporters etc.
Date	Contact	Location	Visit Detail
Oct. 13/16	R. Machial,	Oliver	Discussed FI15 project and the advantages to Fairview Orchards and their packing
	Fairview Orchards		business.
Oct. 24/16	D. Thomas,	Winfield	Training session on root isolation transfers.
<u> </u>	Independent Scout		
Oct. 26/16	S. Bader, Field Scout	Oyama	Training session on root isolation transfers.
20/10	Kalwood Orchards		
Oct. 28/16	Tangaro Orchards	Winfield	Discuss root health and spring treatment plans (soil temp. effect)
Mar. 27/17	CFP (Star Group Marketers)	Kelowna	Discuss fruit quality issues and potential climate change impact.
		Meetings	
Date	Meetings	Location	Details
Sept. 28/16	Meeting with Grower supply	Oliver	Overview of the FI15 project. Emphasis on expanding the ArcGIS program
			into the area of field controls, ordering logistics and marketing.
Oct. 19/16	Lab session -	Winfield	Participants - UBC students, growers, field persons and lab staff.
	root Isolation technique		
	Met with D. Hirkala re-ArcGIS		Discussed need for advanced training with Esri support seminars and related costs.
Dec. 8/16	Met with Molly Thurston	Kelowna	Discussed the ArcGIS program and current difficulties in utilization. Molly is a senior
			horticulturist with BCTFCo, realizes the power of the program and has some mapping
			experience. She has indicated a willingness to assist in
Jan. 19/17	Dr. Hirkala	Winfield	Update on ArcGIS program. Discussions on funding for online tutorial
	Winfield lab meeting		
Jan. 24/17	P. Schwinghammer	Kelowna	Checking on probes inserted last summer. Tested their operation.
	Irrigation Specialist		
	Grower Supply Co.		
Mar. 3/17	Dr. Hirkala	Winfield	Continued discussions on ArcGIS program and standards for field data reporting
	Winfield lab meeting		
Mar. 23/17	Short Course	Kelowna	Included: UBC Students (5), BC Crop Insurance Group (10), Field Service personnel (3),
			Private field monitors (2), Researcher (1), SIR Technician (1), Smld Varieties Corp. Field
Appendix	(1		Technicians (4), Lab Staff (3).
	1		

#### Summary of Individual Contacts Made April 16.17-Sept. 15.17 The table itemizes orchard visits involving direct contact with growers, cooperators, BCTFC specialists and independent packer/owners within the valley. Each involved discussions on project goals in relation to the projected climate-change Grower Date Contact Reason for Visit April 17/17 T. Van Kalkeren (12) Osoyoos, Discuss Peach maturity, Nectarine decline and look for disease carry over. April 17/17 Paul Kalkat (7) Osoyoos, Assess replant problems in the SW corner of a 9 acre Lapin block. April 20/17 Roger Borrett (14) East Kelowna Assess orchard, collect ground samples of grapes for signs of disease carryover. April 20/17 East Kelowna Assess tree decline in Golden Del. Block. Assess peach block for disease samples Dave Ross (7) April 20/17 Kevin Day (1) Kelowna storages. Checking for over-wintering or storge room rot samples. April 20/17 Allen Reid (3) Kelowna Check pear trees for twig dieback. May 2/17 Blair Wilson (8) Kelowna Check apple trees for cankered wood. May 12/17 Lance Issac (6) Oliver Look at decline in cherries. Also check for canker samples. May 24/17 Iqubal Gill (3) Osoyoos Look at cherry tree decline and check for canker samples. May 24/17 Deepank Brar (3) Summerland Assess cherry tree decline. Check cherries for cankered wood. May 30/17 Raj Dhillon (8) Osoyoos Assess Peach and Nectarine tree decline and check orchards for canker samples. June 5/17 Mike Beulah (5) Summerland Assess cherry tree ddecline. June 7/17 Narinder Gill (6) Cawston Assess (a) apples and (b) peaches for replant and leaf chlorosis. June 14/17 Jasvis Sandhu (8) Osoyoos Assess cherry block for decline. June. 15/17 Ravi Dhaliwal (1) Osoyoos Check cherry replant block for root condition. June 29/17 Rick Duarte (4) Oliver Check cherry tree decline. July 7/18 Bikaramjit Sandhu (1) Summerland Check cherry block with decline problems. July 10/19 Gary Klassen (4) Oliver Check on cherry tree and apple nursery problems. July 11/20 Kalwood Farms S. Bader (3) Oyama Check on cherry decline problems. July 13/21 Gurmail Dhaliwal (2) Cawston Check on cherry tree problem. July 21/22 Nirmil Brar (4) Oliver Check on Coronation grapes for fruit condition. July 27/17 Ken Witzke (1) Okanagan Centre Check on cherry fruit condition. Aug. 17/17 B. Dhanoa (1) Vernon Assess decline and winter injury problems. Aug. 22/17 Al Decosta (3) Assess grape maturity. Contacts with Other Groups and Packers in the Industry April 28/17 A&M Orchards (3) Check cherries for canker wood and fruit rot carryover. (Packinghouse) Keremeos June 19/17 Check cherry decline problem. (Packinghouse) Sun City Orchards (3) Kelowna July 4/17 Tony Dimelo Lual Packers(4) Osoyoos Check on cherry decline/replant problem. (Packinghouse) July 25/17 Dr. D. Hirkala Penticton Discussion to update matrix size and the success of the associated data app. Aug. 17/17 T. Dimaria - BCTF Vernon Orchard discussion on canker outbreak. Sept. 12/17 B. Sandhers - SFP -Ellison area Kelowna Aware of our work as part of the climate change project this group has sought help for their extension problems. Volume wise they represent a major segment within the industry. (Packinghouse)

Note: The number in brackets indicates the number of individual visits to this grower. These are not visits directly related to the climate change project but in most cases indicate the spin-off resulting from the initial climate change visit.

Appendix 1 3

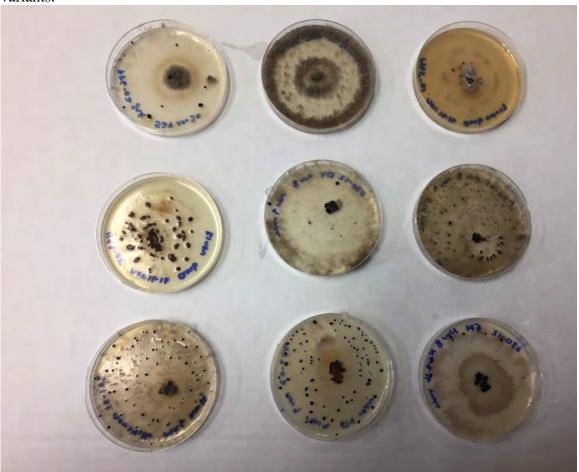
#### Summary of Individual Contacts Made Sept. 15.17 to Present The table itemizes orchard visits involving direct contact with growers, cooperators, BCTFC specialists and independent packer/owners within the valley. Each involved discussions on project goals in relation to the projected climate-change impact on orchard production. Contact Date Grower Reason for Visit Oct. 19-17 G. Klassen Assess Replanting site and take soil sample for microbiological analysis. Oct/Nov. S. Tangaro (4) Tangaro Orchards Visits Tangaro orchards 1 of 6 main orchards in the project. Check tree decline -cause and relation to soil conditions. Discussed potential for support for independent continuation of project. **Extension Personnel** Sept. 14-17 Shauna Bader Kalwood Orchards This group of independent field extension persons were contacted and arragment made to meet to Diana Thomas Ochc Consulting creating a formal group for technology exchange cooperastion. This will be done around the "climate Gayle Krahn Jealous Fruit change " theme. There is interest in creating a program similar to the one installed at the cooperatiive Tamara Richardson Cornucopia Consulting give this group more independence. Jan. 29/18 Dr. Hirkala **BCTFC** Discussions on the ArcGIS program and requests for data related to the project. Packinghouse Manag't 15-Sep Gary Schiek Sandhers Packers Contact for assistance in maturity assessment. Discussion on potential impact climate change could have on maturity timing. Meetings BCFGA Hort Symposium Feb. 8/18 Kelowna Attended on behalf of Turffix (Paloverde Env.) in support of their organic soil ammendment. Feb. 8/18 BCFGA Hort Symposium Kelowna Meeting with Horticulturists from Grower Supply Co. (discussions on use of soil ammendments for soil pathogens).

Appendix 1 4

# Appendix 2 Photo Samples

#### Botrytis types:

These are a number of typical isolates of botrytis obtained from the field, including post harvest. The number classifications are given arbitrarily and all isolates will eventually be compared and the numerical value entered into the data base. As sorting is done by region or degree day value etc. the distribution of type will accompany the sort information if requested. Past experience has shown that there can be 20 + different variants.



Type 1-3 (left to right) top row. Type 4-6 middle row. Type 7, 7 & 8 the bottom row. Type 6 looks similar to type 7 with random and abundant sclerotia but, unclear in the photo, Type 7 has no sporulation while type 6 does have surface spores

The typing is based on the presence and pattern of the sclerotia and spore, which are both important in pathogen survival. Sclerotia are hard, dark masses of dormant hyphae with differentiated rind and medulla and thick, hard cell walls, which permit survival in adverse circumstances such as severe drought, heat and cold. Spores are a reproductive body, which can initiate infection. The abundance of spores is important in the potential for an infection event to occur.

The combination of these characteristics and the variant type's interaction with climate will have a significant influence on the pathogens virulence.

Appendix 2

Picture 1 shows heavy sporulation on the surface and no visible sclerotia. In this isolate sporulation is ringed at the colony periphery and centre.

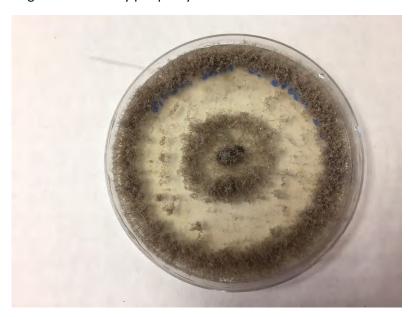


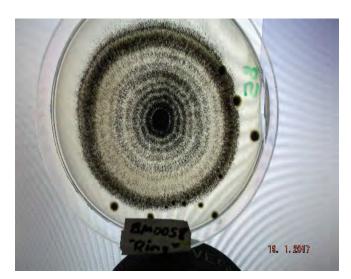
Figure 2 shows a typical isolate with no surface sporulation but with extensive sclerotia development.



Both these examples represent a stable type that occurs frequently and is consistent in form on subsequent transfers onto PDA plates.

Appendix 2 2

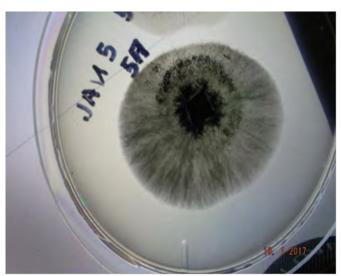
Similarly, with *Alternaria alternata*, which, like Botrytis, is a major market problem in fruit, there appear to be stable variants and, as with Botrytis, there may be survival differences related to climate.





Type 1 Type 2

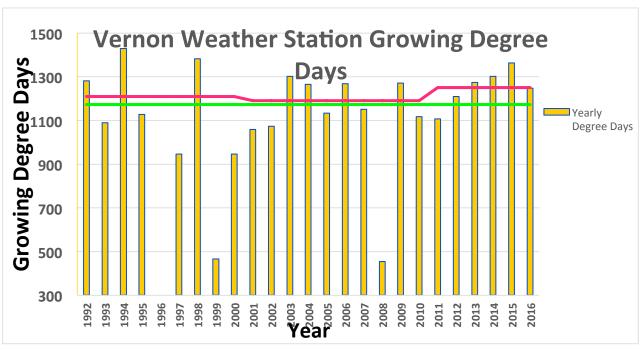
Type 3

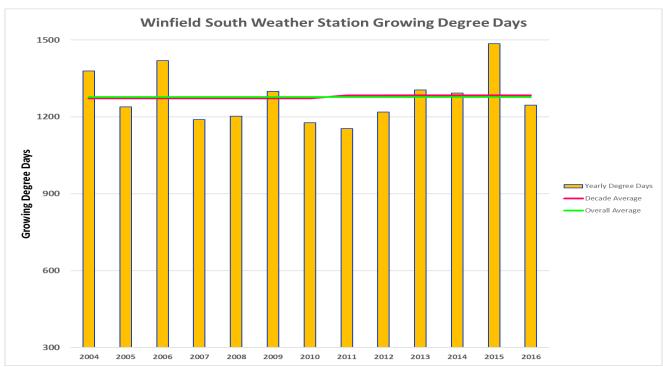


Appendix 2 3

# Appendix 3 Growing Degree Day Accumulation

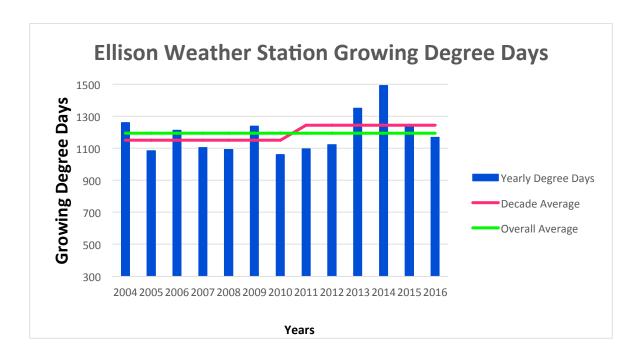
#### North-North Area

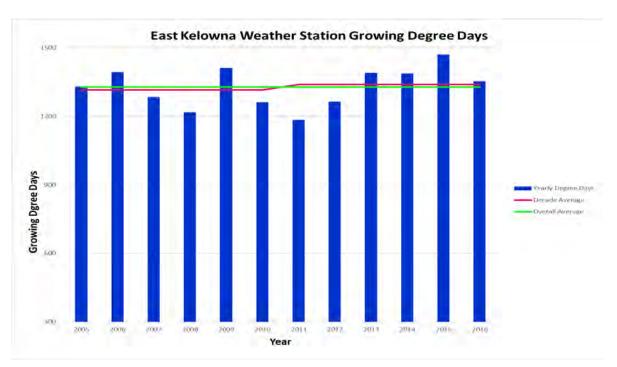




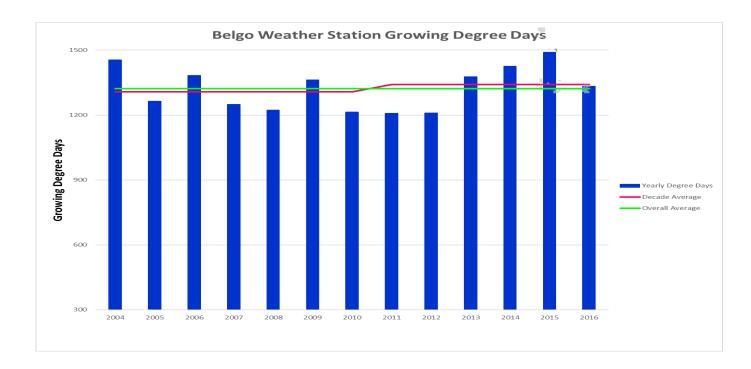
Appendix 3

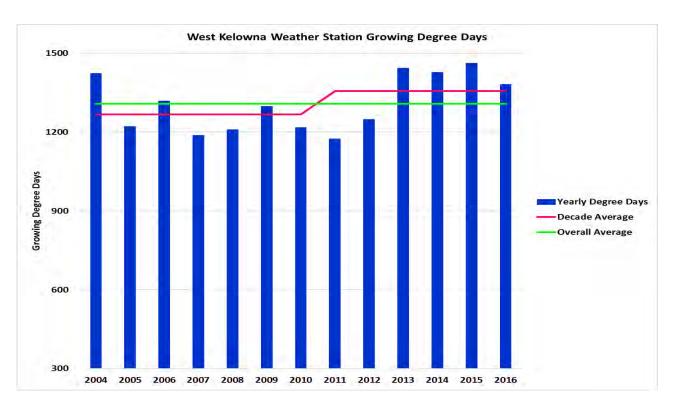
### North Area



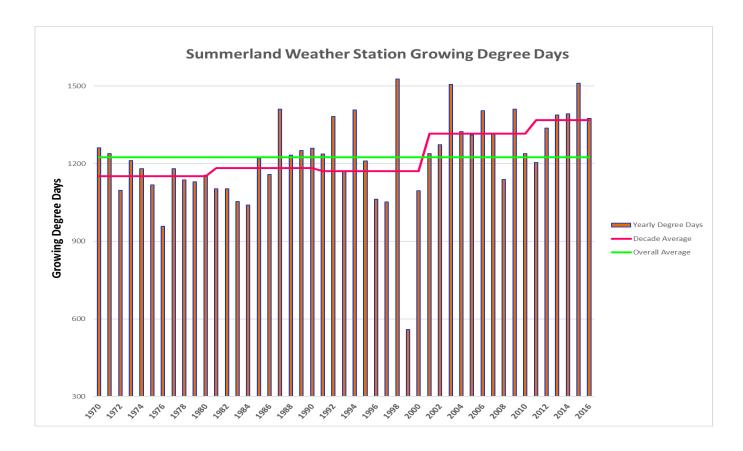


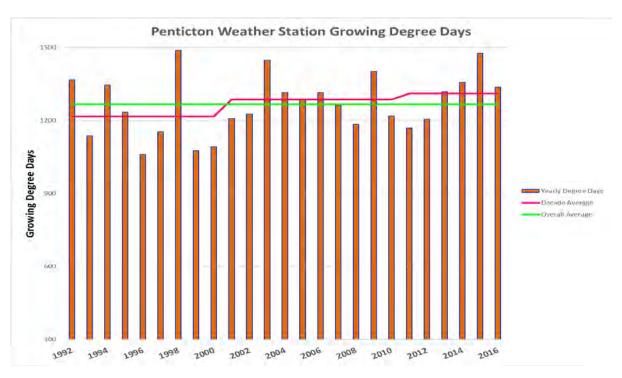
### North Area



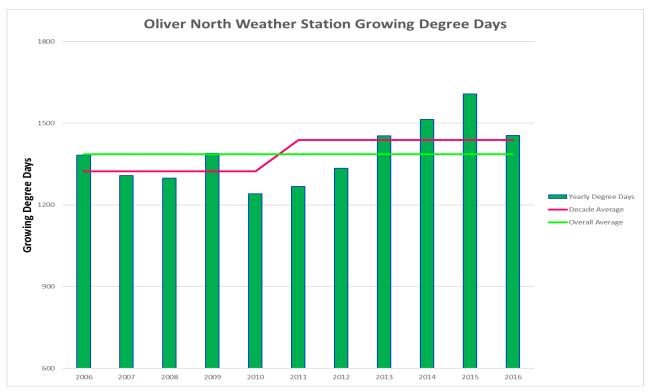


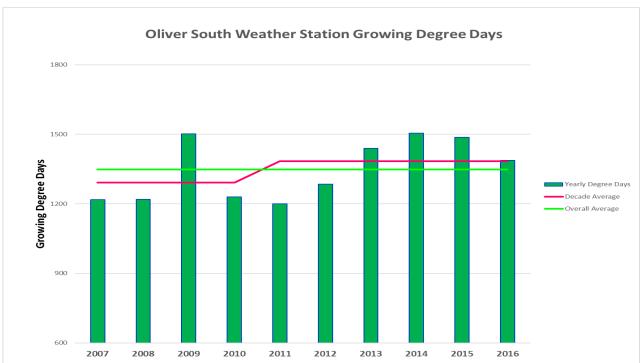
### Central Area



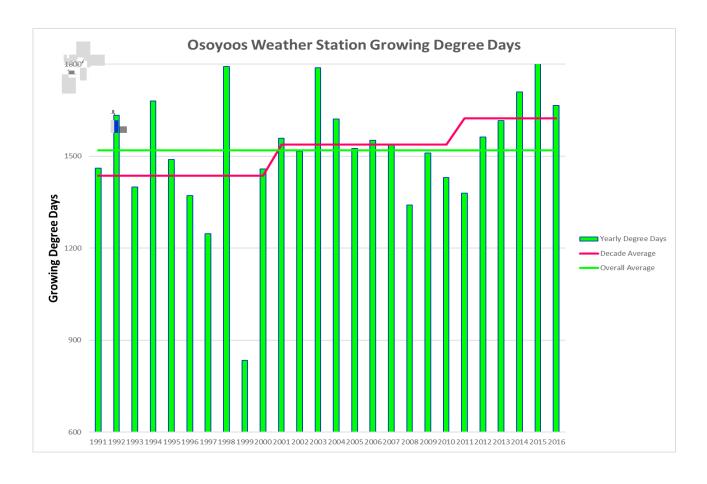


### South Area

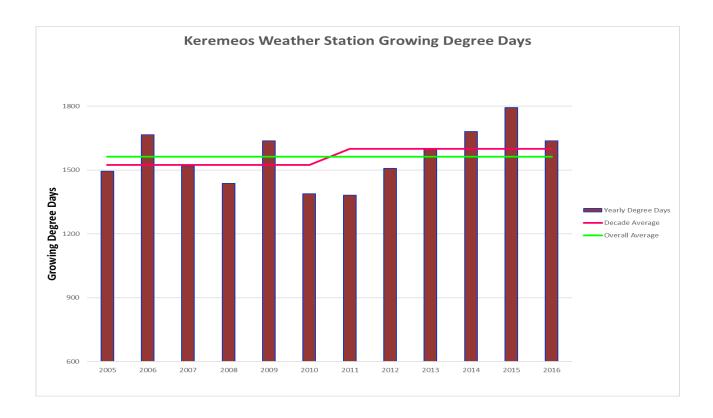


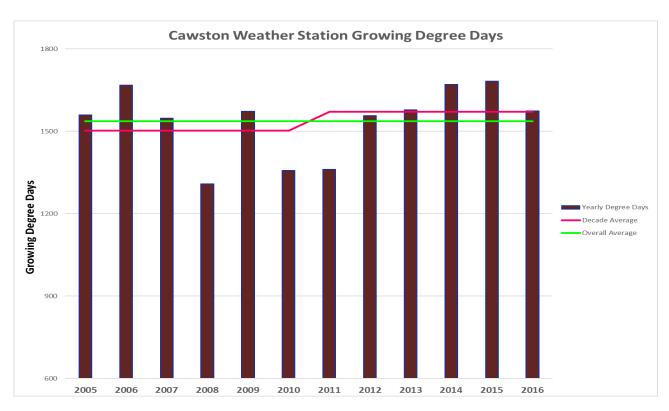


## South South Area



### Similkameen Area





# Appendix 4 Methods

#### **Spore Germination Test:**

This test was carried out by depositing a sample of spores directly on a petrie plate and observing the percent germination that occurs over time. This method allows field monitors to carry out the germination test on many fruit rot samples isolated directly in the field.

Spores are transferred directly from pathogens sporulating on fruit in the field or from a petrie plate colony of a field isolation. This is easily done by using a clean cotton swab and sterile or tap water, touching the swab to a spore mass and gently touching the swab to an agar plate. The spores will normally disperse on the agar surface or, if a dry pickup is used, a drop of water on the "transfer" will disperse the spores sufficiently that they can be clearly distinguished as individuals under magnification.

Several fields are observed and the ratio of germinating spores to non-germinating spores is recorded. The time required for germination to be initiated is also recorded.

#### **Growth Rate Tests:**

The growth rate for any pathogen can enter into its survival chances. The relative growth rate at different temperature levels can also impact the ability of a pathogen to out compete competitors.

Pure isolations were made from fruit samples onto PDA plates. A small core was taken from the isolate and placed on a fresh PDA plate face down. The colony diameter was determined at regular intervals to determine the "growth rate" as mm/day increase in colony diameter.

Expansion of the colony is suppressed as the colony approaches the limits of the petrie plate. Also, growth rates varied greatly from isolate to isolate. Readings were taken as frequently as necessary in order to capture the maximum growth, which generally occurred between 1/3 and 2/3 of the plate diameter.

#### **Colony Sporulation:**

Isolates on PDA that produced surface spores were used to assess the number of spores per square cm. The total area of the colony on the plat was calculated by measuring the colony diameter. Spores were harvested by flooding the plate with water containing a drop or two of Tween 80, used to allow the spores to be wetted. The surface of the colony that was flooded with the Tween-water was agitated with a glass rod to free the spores. The suspension of spores was poured into a 100 ml volumetric flask. The rinsing was done at least three times or until the washings indicated that there were no spores remaining\* and each timer the washings added to the flask. The flask was filled to volume with water.

Appendix 4 1

<sup>\*</sup>The water appeared clear. The test is intended to assess major differences in spore numbers and a visual assessment of the water is sufficient. A simple microscopic assessment can confirm the absence of spores in the final washing.

From the flask a serial dilution was made to bring the spore number to a level that the spores could be conveniently counted under the microscope. A drop of the diluted solution (0.05ml) was fed under a cover slip on a slide. The drop filled the space under the slide. The area visual through the microscope (either at 100 or 400 power) was assessed and the number of spores per field counted (usually between 20 and 50). From this the number of spores per 0.05 ml was calculated (the cover slip 20X22mm square = 4.4square cm) and from this the total number of spores in the volumetric flask was calculated. This gave the number of spores per plate. Dividing the total number of spores by the colony area in square cm gave the number of spores per cm<sup>2</sup>.

Spore numbers per plate varied within the pathogen population but also from pathogen to pathogen. Alternaria spores were relatively large and Penicillium spores relatively small. The number of spores per plate were much higher for Penicillium isolates than for Alternaria. The dilution facture after harvesting the spores was altered to accommodate the counting under the microscope.

#### **Orchard Assessment Procedure:**

In order to assess the impact of any parameter, be it a management strategy or environmental change, it is necessary to document characteristics of the test area that will be a measurement of impact of the change. Scoring individual tree condition within a block has been a technique used in the past and can give a pictorial overview of a block of trees. It is a subjective assessment but very useful for measuring treatment improvements or general decline patterns over time that can be matched to significant weather changes.

An assessment is made by visually scoring individual trees on a 1-10 scale measuring at 0.5 increments. The technique sets an upper score for the best trees in the block based on a subjective rating for their commercial acceptance. This is related to tree size in relation to age, tree vigour and general leaf size and colour.

This value should be somewhere between 7 and 10 considering 7 as the minimum commercially acceptable rating. All trees are compared to the top-level score. The value 0 represents a dead tree or missing tree.

When all trees in a selected area have been scored a single number, obtained by averaging the individual tree scores, can go into the data summary sheet and can be searched and analysed with respect to location within the ArcGIS program.

This is a time-consuming process and obviously cannot be done for all blocks. However, it is useful for specific blocks that require detailed treatments in order to remain commercially viable.

#### **Root Assessments:**

Root development in spring is soil temperature sensitive. Evaluation of roots at this time and throughout the season is an indication of the general health of the tree and of the presence of various pathogens. A scoring system has been developed that assesses three categories within the root system:

Appendix 4 2

- 1. The system score assesses the total root development in relation to tree size. This indicates the normality of the root development over the years of growth.
- 2. The feeder root system assesses the extent of the new feeder roots developing as the tree roots are reactivated in spring (or after a reactivation treatment).
- 3. The die-back assessment is a measure of the level of attack by soil borne pathogens.

Each level is scored on a 1-10 scale. The score-impact and responses are outlined in the following table.

## Scoring System & Relevance to Tree Health

(These numbers are based on several years of field observations in healthy and decline orchards)

Syste	System Score		Feeders		Die-back		
<7 7+	Usually indicates early management problem. Commercially acceptable		∢4	Serious shortage of feeders necessary for nutrient and water uptake. Usually equat- ed to poor tree growth.	<4	A die- back score of less than 4 is acceptable.	
NOTE	The root system in the field is predetermined by replant and post plant	4	1-5	Poor feeder condition. Usually indicating that the tree is in decline	4 - 5	When the die-back damage reaches a level of 4-5 this should trigger some concern. The normal response would	
	management. The segment of the root system assessed in a	5		Acceptable but with concern.  Roots in this condition need to be monitored closely.		be to monitor feeder root condition to determine if this die-back level continues to increase.	
	field-assessment is very limited. In the pot test, the results represent the entire root system and	7	7 - 8	Generally associated with good healthy tree if die-back is low.		When the die-back level becomes greater than 5, corrective procedures should be initiated.	
	the "system" value be- comes more meaningful.		>8	Excellent feeder roots but dependent on die-back being low.			

This system does not accurately do direct comparisons between blocks because of the tremendous variability is root development from soil to soil. However, the assessment in conjunction with the tree health is a guideline to the necessity for root management changes.

#### **Fungicide Efficacy Test:**

Standard commercial test by BCTFC lab using fungicide amended PDA plates to control growth.

#### **DNA Identification Procedure:**

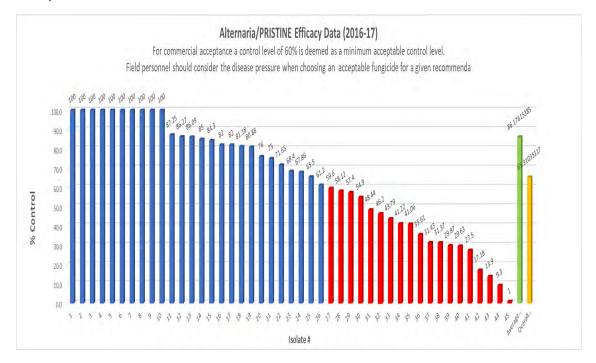
Standard electrophoresis system with identification markers.

Appendix 4 3

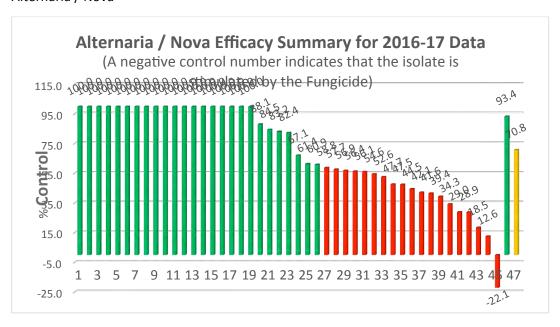
### Appendix 5 Fungicide Graphs

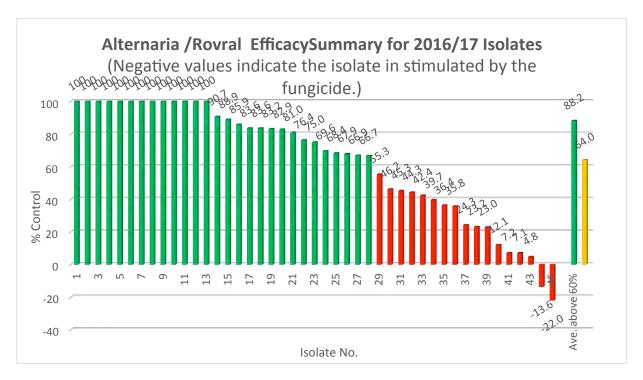
There is an enormous amount of fungicide efficacy data attached to the database. These are a few of the typical efficacy graphs that have been made. A full set of up-to-date graphs will be prepared by early spring covering the major tree fruit rots. These will be made available to all field personnel including the independents.

#### Alternaria / Pristine

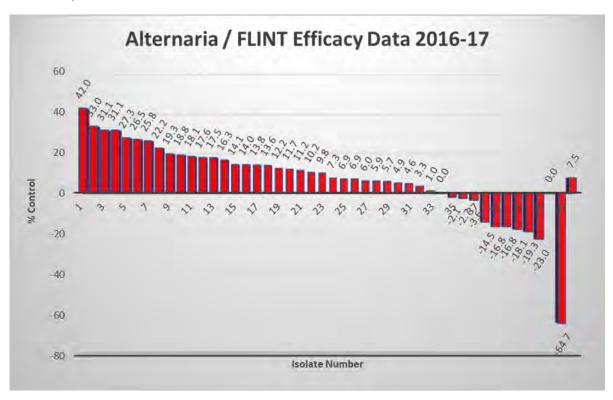


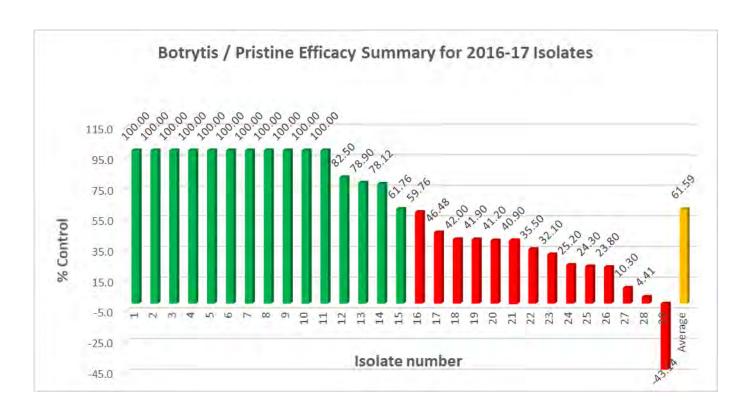
#### Alternaria / Nova

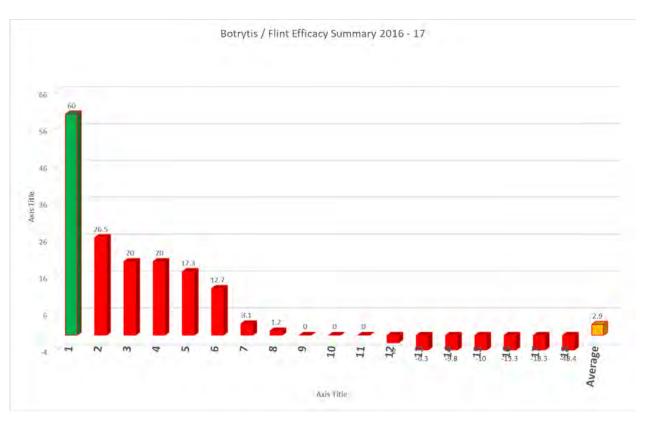


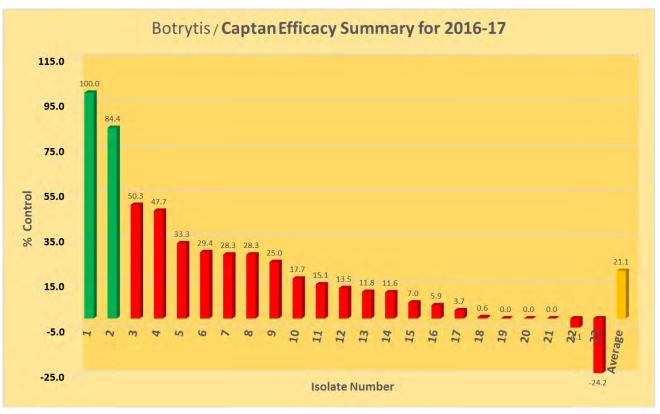


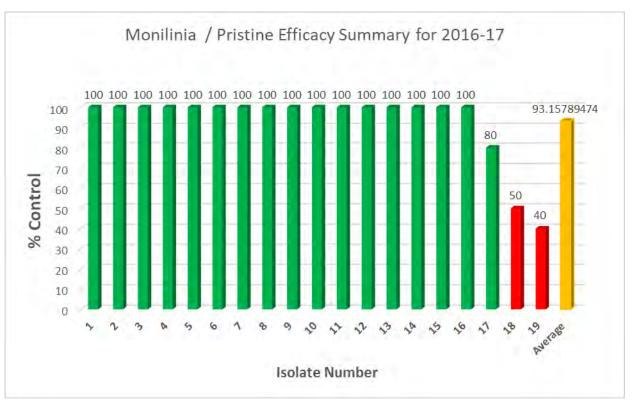
#### Alternaria / Flint



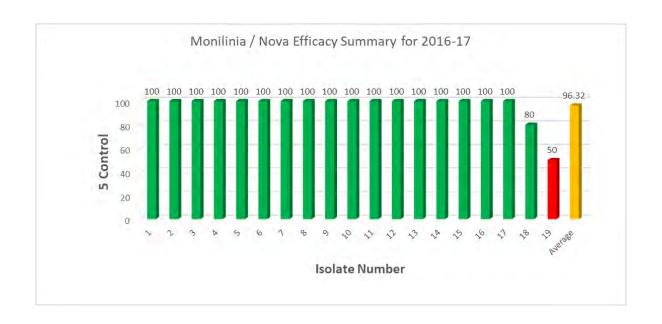


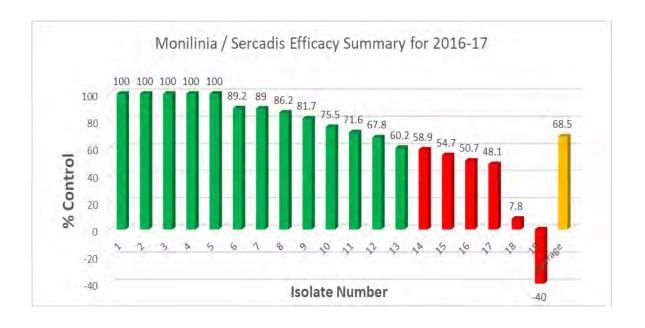






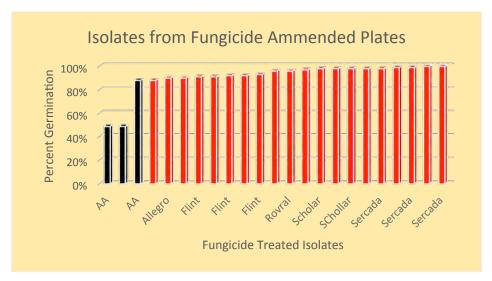
Appendix 5 4

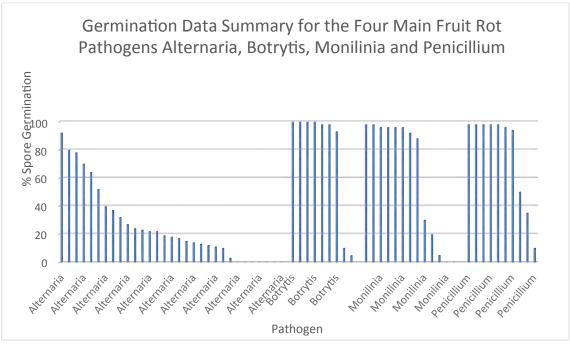


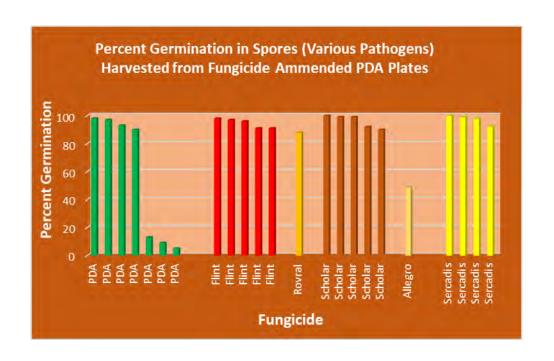


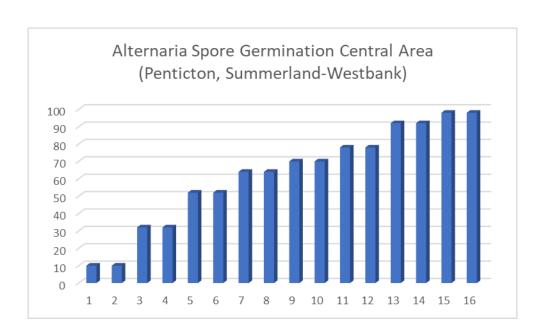
Appendix 5 5

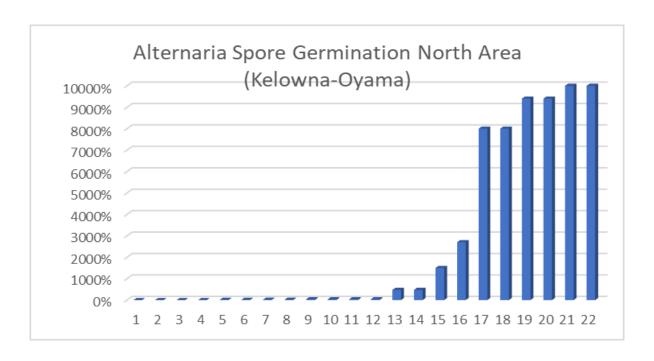
## Appendix 6 Germination Graphs

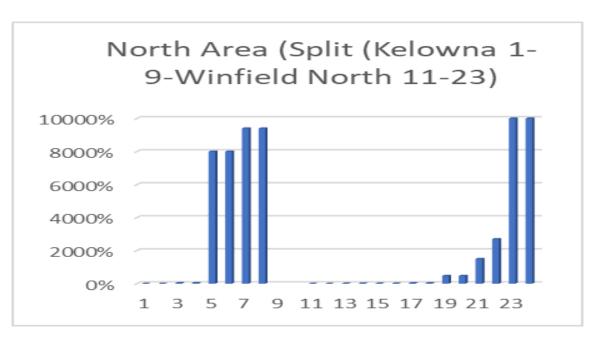


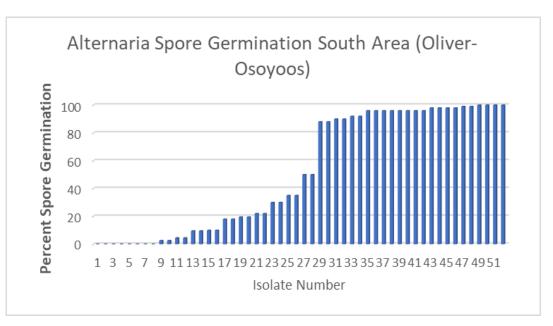


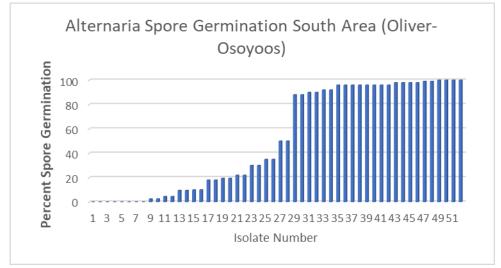


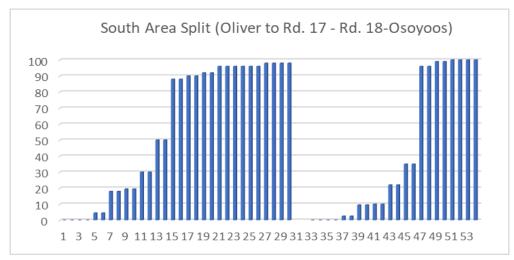


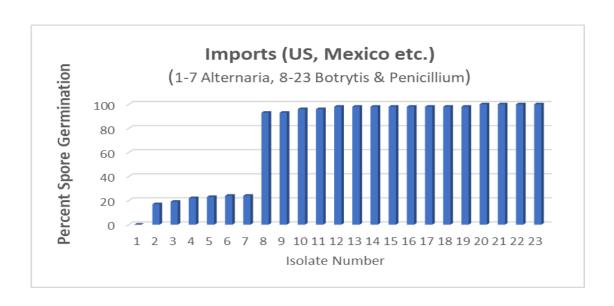












# Appendix 7 Root Isolations Data 2016

			Appendix	- Root				
	A. Sid SOIL Trich 1	?	85 mm		Soil	Vernon	Apple	trich
	A.Sid SOIL Trich 2	?	85 mm		Soil	Vernon	Apple	trich
1 ISO's	A.Sid Sunrise 2T 2nd	4-Jul	53mm		Tip	Vernon	Apple	Unknown black
2 ISO's	A.Sid Sunrise 3L	28-Jun	40mm		Lesion	Vernon	Apple	Unknown black
3 ISO's	A.Sid Sunrise 3T 2nd	4-Jul	40mm		Tip	Vernon	Apple	Unknown black
4 ISO's	A.Sid Sunrise 4Ta	28-Jun	40mm		Tip	Vernon	Apple	Alternaria
5 ISO's	A.Sid Sunrise 4Tb	28-Jun	53mm		Tip	Vernon	Apple	Alternaria
6 ISO's	Bailey Repl. SOIL - 3 #1	?	68mm		Soil	Oyama	Cherry	Vert
7 ISO's	Bailey Repl. SOIL - 3 #2	?	70mm		Soil	Oyama	Cherry	Vert
	Bailey Repl. SOIL 1 trich	?	85 mm		Soil	Oyama	Cherry	trich
	Bailey Repl. SOIL 1 vert	12-Jul	85 mm		Soil	Oyama	Cherry	vert
	Bailey Repl. SOIL 2 trich	?	85 mm		Soil	Oyama	Cherry	trich
	Bailey Repl. SOIL 2 vert	12-Jul	85 mm		Soil	Oyama	Cherry	vert
	Campbell Amb 4T #1	?	85		Tip	Osooyos	Apple	vert
	Campbell Amb 4T #2	?	85		Tip	Osooyos	Apple	Cylindro
	Dhaliwal SOIL Trich 1	?	85 mm		Soil	,	Soil	Trich
	Dhaliwal SOIL Trich 2	?	85 mm		Soil		Soil	Trich
	Garth 1T	Aug. 11			Tip	Osoyoos	Apple	Cylindrocarpon
	Geen Glengrow SOIL 1Lo	,	85 mm		Soil	Winfield	Cherry	Trich
	Geen Glengrow SOIL 2Lo	***********	85 mm		Soil	Winfield	Cherry	Trich
	Geen Home CH SOIL #1	?	85 mm		Soil	Winfield	Cherry	Trich
***************************************	Geen Home CH SOIL #2	?	85 mm		Soil	Winfield	Cherry	Trich
	Geen WE Re CH 1L	5-Aug	53mm	~13mm/		Winfield	Cherry	Trich
	Geen WE Re CH 1L	3-Aug	85	~21mm/		Winfield	Cherry	Trich
	Geen WE Re CH 1T	3-Aug	85	~21mm/	Tip	Winfield	Cherry	Mucor/Rhizo
	Geen WE Re CH 6T	3-Aug	85	~21mm,	Tip	Winfield	Cherry	Mucor/Rhizo
	Geen WE Replt. Turf 1L	1-Aug			Lesion	Winfield	Cherry	Cylindrocarpon
	Geen WE Replt. Turf 1T #1	21-Jul	30 mm		Tip	Winfield	Cherry	Cylindrocarpon
	Geen WE Replt. Turf 1T #2	21-Jul	32 mm		Tip	Winfield	Cherry	Cylindrocarpon
	Geen WE Replt. Turf 2L	1-Aug				Winfield	Cherry	Cylindrocarpon
	Geen WE Replt. Turf 2T	1-Aug			Tip	Winfield	Cherry	Cylindrocarpon
	Geen WE Replt. Turf 3L	1-Aug				Winfield	Cherry	Cylindrocarpon
	Geen WE Replt. Turf 3T	1-Aug			Tip	Winfield	Cherry	Cylindrocarpon
	Geen WE Test Tree 2T	5-Aug	53mm	~13mm,	Tip	Winfield	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	Mucor/Rhizo
	GLC SOIL 1	?	85 mm		Soil		soil	trich
	GLC SOIL 2	?	85 mm		Soil		soil	trich
	GLT Trich SOIL 1Lo	?	85		Soil			Trich
	GLT Trich SOIL 2Lo	?	85		Soil			Trich
	Gord not treat 1T 2nd	20-Jun	32mm		Tip	Westbank	Cherry	Cylindro
	Gord not treat 3T 2nd	20-Jun	53mm		Tip	Westbank	Cherry	cylindro
	Gord not treated 1L 2nd		20mm		lesion	Westbank	Cherry	? Multiple
	Gord treat 1L 2nd	20-Jun	35mm	5mm/da	lesion	Westbank	Cherry	Cylindro
	Gord Treat 1T #1	?	85 mm		Tip	Westbank	Cherry	Unknown 2
	Gord Treat 1T #2	?	85 mm		Tip	Westbank	·	Unknown 2
	Gord treat 1T 2nd	20-Jun	53mm		Tip	Westbank	·····	cylindro + Unk2
	Gord Treat 2L 1	ıne 15/1		4mm/da		Westbank	3	
	Gord Treat 2L 1	une 15/1	85 mm	4mm/da	lesion	Westbank	Apple	<u> </u>

Gord Treat 2L 2	?	85 mm		lesion	Westbank	Apple	
Gord Treated 1L 2nd	#####	40mm	?????	Legion	West Ke	/	
Gord Treated 1T 2nd	#####	46mm	******	····	West Ke		
Gord Treated 2L			:	}	West Ke		
Gord treated I 3rd	20-Jun	43mm			Westbank	Cherry	Cylindrocarpon
Gord Treated   3rd	#####	44mm	?????	Root	West Ke	Iowna	
Gord Untreated 1L 2nd	#####	14mm	?????	Legion	West Ke	Iowna	
Gord Untreated 1T 2nd	#####	34mm	?????	Root tip	West Ke	Iowna	
Gord Untreated 2T			?????	Root tip	West Ke	Iowna	
Gord Untreated 3T 2nd	#####	44mm	?????	Root tip	West Ke	Iowna	
Gord Untreated 4T		<u> </u>	?????	Root tip	West Ke	lowna	
HA BI3S 4T 3rd	une 8/10	6	7mm/da	Tip	Cawston	Apple	Cylindrocarpon
HAB12S+H 1H	#####	46mm	4mm/d	Root tip	Cawston	apple	
HAB12S+H 2H	#####	40mm	4mm/c	Root tip	Cawston	apple	
HAB12S+H 2S 2nd	16-Jun		13mm/c	·		Apple	Unknown 1b
HAB12S+H 3H	#####	42mm	,		Cawston	apple	ļ
HAB12S+H 3S	#####	68mm	6mm/c	day		apple	
HAB12S+H 4H #1	6-Jun	85mm		?	Cawston	Apple	alternaria
HAB12S+H 4H #2	7-Jun	·	4mm/da	·····	,	Apple	alternaria
HAB13S 1T Second		10mm	,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	Cawston		Trich
HAB13S 2T		18mm	·		Cawston		Trich
HAB13S 3Ta	6-Jun	<del>,</del>	3mm/da		·	Apple	Cladosporium
HAB13S 3Ta		24mm	*		Cawston		Trich
HAB13S 4T 2nd	6-Jun	<del>,</del>	7mm/da	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	,	Apple	Cylindrocarpon
HAB13S 4T Third		68mm	·····	·····	Cawston	····	Trich
HAB13S 5T Second		66mm	·····	·····	Cawston	f	
HMS4x Fuji 1L	,	38mm 46mm	4mm/0	m	Cawston	munum	
HMS4x Fuji 3T Second		<del> </del>	<u> </u>	\	Cawston	f	
HMS4x Fuji 3T Second HMS4x Fuji T2a		38mm 82mm	,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	Cawston Cawston	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	Aureobasid
HMS4x Fuji T2b		82mm	,i	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	Cawston		Aureobasid
HMS4x SOIL #1	##### 12-Jul	0.5	Jiiiii/(	Soil			trich
HMS4x SOIL #2	12-Jul 12-Jul			Soil	Cawston Cawston	Apple Apple	trich
HMSCH Fuji 1L	12 JUI		3mm/c	<del></del>	Cawston		Cylindro
HMSCH Fuji 1T			<del>,</del>	·	Cawston	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	Cylindro
HMSCH Fuji 2T				·····	Cawston	·	, -,
HMSCH Fuji 2T 2nd	16-Jun	Transfer		Tip		Apple	llyonectria
HMSCH Fuji 3T		42mm	7mm/c	}	Cawston	·	
McLennan D1 1L	Aug. 9	·	·	Lesion	·	Cherry	Fusarium
McLennan D1 2L	Aug. 11	·		Lesion		Cherry	Unknown 1a
McLennan D1 3L	Aug. 16	•	20 mm/	Lesion		Cherry	Unknown 1a
Norton check 2L	24-Jun	•		lesion	Oliver	Cherry	Pythium
Norton check 3T	24-Jun	53mm		Tip	Oliver	Cherry	Cylindro
Norton Turf 1L #1	?	85		lesion	Oliver	Cherry	Cylindrocarpon
Norton Turf 1L #2	?	85		Lesion	Oliver	Cherry	Unknown 1
Norton Turf 1T	28-Jun	42mm	4mm/da	Tip	Oliver	Cherry	cylindro?
Norton Turf 2L 2nd	4-Jul	43mm	9mm/da	lesion	Oliver	Cherry	Trichoderma

Appendix 7 2

Norton Turf 2T	28-Jun	85mm		Tip	Oliver	Cherry	Unknown 1b
Norton Turf 2T #1	?	85 mm		Tip	Oliver	Cherry	Trichderma
Norton Turf 2T #2	?	85 mm		Tip	Oliver	Cherry	Cylindrocarpon
Norton Turf 3T	28-Jun	85mm	6mm/d		Oliver	Cherry	cylindro?
Norton Turf 3T 2nd	27-Jun	40mm	5mm/da	Tip	Oliver	Cherry	Cylindrocarpon
Norton Turf 4T	28-Jun	85mm	3mm/da	Tip	Oliver	Cherry	Cylindro
Norton Turf 5T 2nd	4-Jul	53mm		Tip	OLiver	Cherry	Illyonectria
Norton Turf 5T 2nd	27-Jun	39mm	3mm&5	Tip	Oliver	Cherry	Cylindrocarpon
Norton Turf 6T 2nd	27-Jun	36mm	4.5mm/	Tip	Oliver	Cherry	Cylindrocarpon
Ouchi blue 10T 3rd	7-Jul	53mm		Tip	vernon?	Gala	trich
Ouchi Blue 1L	28-Jun	53mm		Lesion	vernon?	Apple	Cylindrocarpon
Ouchi Blue 2T 2nd	4-Jul	40mm		Tip	vernon	Apple	Ilyonectria
Ouchi Blue 3T 2nd #1	4-Jul	53mm	3mm/da	Tip	vernon	Apple	Ilyonectria
Ouchi Blue 3T 2nd #2	4-Jul	53mm		Tip	vernon	Apple	Ilyonectria
Ouchi Blue 4T 2nd	4-Jul	53mm		Tip	vernon	Apple	Ilyonectria
Ouchi Blue 6T trans 3rd	11-Jul	53mm		Tip	Vernon	Apple	Verticillium
Ouchi Blue 8T	28-Jun	53mm		Tip	vernon?	Apple	Unknuwn 3
Ouchi Blue 9T	28-Jun	53mm		Tip	vernon?	Apple	Unknuwn 2
Ouchi Check 1T 2nd	4-Jul	53mm		Tip	Vernon	Apple	Unknown
Ouchi Check 2T 3rd	4-Jul	50mm		Tip	Vernon	Apple	Unknown black
Ouchi Check 3T	28-Jun	44mm		Tip	Vernon	Apple	Alternaria
Ouchi Check 4T	28-Jun	40mm		Tip	vernon?	Apple	Unknown black
Ouchi Orange 1T #1	?	56mm		Tip	Vernon	Apple	Verticillium
Ouchi Orange 1T #2	?	58mm		Tip	Vernon	Apple	Verticillium
Ouchi Orange 2T 3rd	7-Jul	50mm		Tip	vernon	Apple	Illyonectria
Ouchi Orange 4T	24-Jun			Tip	Vernon	Apple	Ilyonectria
Ouchi Orange 6T	24-Jun			Tip	Vernon	Apple	llyonectria
Ouchi Pink 3L#1	?	85 mm		Lesion	Vernon	Apple	Cylindrocarpon
Ouchi Pink 3L #2	?	85 mm		Lesion	Vernon	Apple	Cylindrocarpon
Ouchi pink 4L	24-Jun	53mm	5mm/da	lesion	vernon?	Gala	trich
Plate Label/Grower	Date	Current	·	Isolated	Location	Fruit Typ	Our ID
RB Home 1T #1	16-Jun		7.5mm/		Oyama	Cherry	Trichoderma
·····	une 17/1	·	5.5mm/	Tip	Oyama	Cherry	Trichoderma
			8.5mm/	Tip 	Oyama	Cherry	Cylindrocarpon
		85 mm		Tip	Oyama	Cherry	Cylindrocarpon
· · · · · · · · · · · · · · · · · · ·		85 mm	8.5mm/	Tip 	Oyama	Cherry	Cylindrocarpon
RB Home 3T #2	?	85 mm	0.5	Tip 	Oyama	Cherry	Cylindrocarpon
			8.5mm/	Tip	Oyama	Cherry	Cylindrocarpon
	une 20/1		7,000	Tip	Oyama	Cherry	Cylindrocarpon
RB Home 4T 2nd	20-Jun		7mm/d		Oyama	Cherry	vert? Unknown 1
RB old vern 1L 2nd	17-Jun		2mm/dy		Oyama	Cherry	unknown 4
RB old vern 1L 3rd RB old vern 3T 2nd	20-Jun		0 F /		Kelowna	Cherry	Unknown 1
RB old vern 3T 3rd	17-Jun	~~~~~~	8.5mm/		Oyama	Cherry	Fusarium
RB old vern 31 3rd	20-Jun	30mm 53mm	1E mm/	Tip	Kelowna	Cherry	Illyonectria
RB old vern. Replant 4T	15-Jun #####		15 mm/	<del></del>	Kelowna Kelowna	Charry	mucor
			711111/(			·	Unknows
Reena Lapin 4TA 2nd	20-Jun	39mm	i	Tip	S'land	Cherry	Unknown

Reena lapin 4Tb 2nd	20-Jun	53mm		Tip	S'land	Cherry	Unknown 5
Reena Mac 2T	15-Jun	3311111	5mm/da		Summerlar		Verticillium
Reena Mac 2T #2	10-Aug	53	Jiiiii/u	Tip	Summerlar		Trichoderma
Reena Mac T3							
Reena Macs 1L 2nd	10-Aug	35mm		Tip	Summerlar S'land		Trichoderma
Reena Macs 1T 2nd	20-Jun 20-Jun	85mm		lesion Tip		Apple	Unknown 5
Reena Macs 2T 3rd A/B		85mm			S'land	Apple	Unknown 2
Reena Macs 4T 2nd	20-Jun	50mm		Tip	S'land	Apple	Unknown 1b
Reena Macs 5T 2nd	20-Jun 20-Jun	53mm		Tip	S'land	Apple	Cylindrocarpon
Rob S 2y T3 Third			21mm/s	Tip	S'land	Apple	Trichoderma
Rob S. 2nd yr. graft T1 3r	21-Jun	85mm	21mm/c	Tip	Cawston	Apple	Unknown 1a
Rob S. 2y graft T2	17-Jun	53mm	17mm	Tip	Cawston Cawston	Apple	Unknown 4
Rob S. 2y T4 3rd	23-Jun	85mm	±//			Apple	
Sick 1D 6T 2nd	19-Jul	53mm		Tip Tip	Cawston Winfield	Cherry	Verticillium Cylindrocarpon
Sick 1D 8L 2nd	19-Jul	53mm			Winfield	····	Cylindrocarpon
Sick 2A 1L 2nd	22-Jul	53mm		Lesion	Winfield	Cherry Cherry	vert ?
Sick 2A 1E 2Hd Sick 2A 1T	22-Jul	53mm	53Mm	Tip	Winfield	Cherry	vert?
Sick 2A 2T 2nd	22-Jul	53mm	53Mm		Winfield	Cherry	vert?
Sick 2A 3L	19-Jul	53mm	. 33141111	Tip Lesion	Winfield	Cherry	Cylindrocarpon
Sick 2A 3L 2nd	23-Jul	53mm			Winfield	Cherry	Cylindrocarpon
Sick 2A 3T 2nd	22-Jul	53mm	53Mm	Tip	Winfield	Cherry	trich
Sick 2A 4L	19-Jul	53mm	. 33141111	Lesion	Winfield	Cherry	Cylindrocarpon
Sick 2A 4L 2nd	22-Jul	53mm	53Mm		Winfield	Cherry	cylindro?
Sick 2A 4T	19-Jul	30mm	Joiviiii	Tip	Winfield	Cherry	
Sick 2A 4T 2nd	22-Jul	53mm		Tip	Winfield	Cherry	Cylindrocarpon cylindro?
Sick 2B 1L	21-Jul	64mm		·····	Winfield	Cherry	Cylindrocarpon
Sick 2B 3L	22-Jul	53mm	53Mm		Winfield	Cherry	cylindro?
Sick 2B 4L	21-Jul	30mm	Joiviiii		Winfield	Cherry	Cylindrocarpon
Sick 2B 4T	21-Jul	50mm		Tip	Winfield	Cherry	cylindro?
Sick 2B 5T	21-Jul	53mm		Tip	Winfield	Cherry	cylindro?
Sick 2C 3T	9-Aug	transfer		Tip	Winfield	Cherry	Cylindrocarpon
Sick 2C 4T		transfer		Tip	Winfield	Cherry	Cylindrocarpon
Tangara 1B 3T	19-Jul	trunsier		Tip	Winfield	Cherry	Neonectria
Tangaro sent 2T	24-Jun	48mm		Tip	Winfield	Cherry	Alternaria
Tangaro sent 3T	24-Jun	35mm		Tip	Winfield	Cherry	Epicoccum
Tangaro Sent 4T	24-Jun	30mm		Tip	Winfield	Cherry	Epicoccum
Tangaro Sick 2A 2L	19-Jul			Lesion			
Tangaro Sick 11B 3T	19-Jul			Tip			Cylindrocarpon
Tangaro Sick 1A 1T	19-Jul			Tip			Cylindrocarpon
Tangaro sick 1A 2T	19-Jul	53		Tip	Winfield	Cherry	Fusarium
Tangaro sick 1C 1T	15-Jul	38mm		Tip	Winfield	Cherry	vert
Tangaro Sick 1C 3L A	19-Jul			Lesion			Cylindrocarpon
Tangaro Sick 1C 3LB	Aug. 3			Lesion			Cylindrocarpon
Tangaro sick 1C 4L	19-Jul	32			Winfield	Cherry	
Tangaro sick 1D 1L	15-Jul	34mm			Winfield	Cherry	cylindro
Tangaro sick 1D 1T	15-Jul	53mm		Tip	Winfield	Cherry	Unknown 4
Tangaro Sick 1D 7L 2nd	19-Jul	21.71.1		Lesion		1	

Appendix 7 4

,					,	<del>,</del>
Tangaro Sick 2C 1L	21-Jul	44mm	Lesion	Winfield	Cherry	cylindrocarpon
Tangaro Sick 2C 2T	21-Jul	42mm	Tip	Winfield	Cherry	Cylindrocarpon
Tangaro Sick 2C 5L	21-Jul	40mm	Lesion	Winfield	Cherry	cylindrocarpon
Tangaro Sick 2C 6T	21-Jul	42mm	Tip	Winfield	Cherry	Fusarium
Tangaro Sick 2D 2T	21-Jun		Tip	Winfield	Cherry	Cylindrocarpon
Tangaro Sick 2D 4T	21-Jun		Tip	Winfield	Cherry	Cylindrocarpon
Tangaro Tim lot 1T 2nd	4-Jul	53mm	Tip	Winfield	Cherry	Cylidrocarpon
Tangaro tim lot 2T	28-Jun	53mm	Tip	Winfield	Cherry	?
Tangaro Tim Lot 3T 2nd	4-Jul	53mm	Tip	Winfield	Cherry	cylindrocarpon
Tangaro Tim Lot 4T 2nd	4-Jul	53mm	Tip	Winfield	Cherry	Unknown 2
Tangaro Tim lot 4T 3rd	11-Jul	53mm	Tip	Winfield	Cherry	Cylindrocarpon
Tangaro UPS 1TA	24-Jun	53mm	Tip	Winfield	Cherry	Unknown 1
Tangaro UPS 1Tb	24-Jun	53mm	Tip	Winfield	Cherry	unknown 4
Tangaro UPS 3T 2nd	29-Jun	85mm	Tip	Winfield	Cherry	Unknown 1a
Tangera sick 1A 3T	10-Aug	32	Tip	Winfield	Cherry	Fusarium ?
Tangera sick 1A B	10-Aug	53		Winfield	Cherry	Trichoderma
Tangera sick 1B 5Ta	10-Aug	30	Tip	Winfield	Cherry	Fusarium ?
Wit ori HC #2	#####	68mm	expon Root tip	Kelowna	apple	Trichoderma
Witzke Ori HC 1L 2nd	16-Jun	85mm	Lesion	Kelowna	Apple	Unknown 3
Witzke Ori HC 1T 2nd	16-Jun	85mm	Tip	Kelowna	Apple	Verticillium
Witzke ori HC 2La	#####	70mm	10mm, Root tip	Kelowna	apple	
Witzke ori HC 2Lb	#####	70mm	8mm/(Root tip	Kelowna	apple	
Witzke Ori HC 2T 2nd	16-Jun	60Mm	Tip	Kelowna	Apple	Verticillium
Yosh replant 2L 2nd	4-Jul	53mm	lesion	vernon		Unknown 1
•						

Appendix 7 5

## Appendix 8 Sporulation Graphs

