

THE RISK OF *PYTHIUM APHANIDERMATUM* IN  
HYDROPONIC BABY-LEAF SPINACH PRODUCTION

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# THE RISK OF *PYTHIUM APHANIDERMATUM* IN HYDROPONIC BABYLEAF SPINACH PRODUCTION

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*Pythium aphanidermatum* has been identified as the main obstacle/risk in the production of hydroponically grown babyleaf spinach. This organism is so prevalent that even crops grown in fresh nutrient solution will often show signs of *Pythium* damage at harvest time. Previous studies have identified nutrient solution temperature as a key factor in determining the growth and development of *Pythium*.

The goal of this study was to estimate the risk of *Pythium* outbreaks under different hydroponic production strategies. To provide these estimates, a greenhouse simulation model was coupled with a spinach growth and *Pythium* disease model. Through Monte-Carlo simulation, estimates of the time between and the seasonality of outbreaks were determined. The strategies modeled included using nutrient solution temperatures of 18, 20, 22, 24C and unregulated temperature. Within these temperature regimes different harvesting schedules were examined. Assuming supplemental lighting control to a daily light integral of  $17 \text{ mol m}^{-2}$ , crop durations of 12, 14 and 16 days in pond were examined. To quantify the importance of supplemental lighting, crop durations were replaced with target harvest biomasses. The target harvest biomasses were then achieved without supplemental lighting. A further strategy examined within the fixed duration conditions was the use of a two pond system where crop cohorts older than half the total crop duration were moved to a separate pond.

A nutrient solution temperature of 18C with a 12 day crop duration resulted in an expected frequency of *Pythium* outbreak of 0.032 per year, compared to 1.47 outbreaks per year for a 16 day crop grown without nutrient solution temperature control. The same conditions without supplemental lighting resulted in outbreak frequencies of 7.9 per year and 16 per year respectively. Under a two pond system the expected frequency of outbreak was approximately halved at 18C and increased with increasing temperature until there was no difference between the one and two pond systems for the uncontrolled temperature conditions.

## BIOGRAPHICAL SKETCH

Timothy Shelford was born in Vancouver, BC in 1974 to Helen and James Shelford. He graduated from Point Grey Highschool in 1992. He then did his undergraduate studies at the University of British Columbia in the department of Bioresource Engineering, and graduated with a B.A.Sc. with distinction in 1997.

Timothy then completed a Masters of Applied Science in Bioresource Engineering at the University of British Columbia, in 2000. His project investigated different ways of scheduling irrigation in greenhouse tomato production, with the aim of conserving water and nutrients. Following his Masters degree he worked as an environmental engineer at Entech Environmental Consultants in West Vancouver, BC. Projects included contaminated site investigations and powder post beetle assessment and abatement in historic wooden structures.

During his youth he worked at a variety of agricultural jobs from picking raspberries and selling sweet corn on his uncle's farm, to an animal technician on the Agriculture Canada greenbelt research farm in Ottawa ON, and as a research associate on a fish farm research project on Quadra Island, BC.

In 2002 Timothy was accepted into the Cornell Controlled Environment Agriculture program in the department of Biological and Environmental Engineering at Cornell University. During his time at Cornell he worked as a teaching assistant in several courses including, Introductory computer programming, Heat, Mass and Momentum Transfer, and Bioinstrumentation. He also worked on a variety of projects related to hydroponic spinach production. In 2007 he began working for CEA Systems Inc, developing a supplemental light, shade and CO<sub>2</sub> control algorithm developed by his advisor Professor Louis Albright.

His hobbies include collecting tools for home improvement projects, keeping various ancient computers running and travel in general.

This research is dedicated to my father who passed away in 2002, and my mother whose unquestioning love and support have made everything I've done possible.

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A listing of all my friends, who provided encouragement, support and sanity preserving distraction would fill volumes two and three of this dissertation and would still not be complete.

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## CHAPTER 1: INTRODUCTION

### 1.1 BACKGROUND

#### *Why hydroponic baby-leaf spinach?*

The Controlled Environment Agriculture (CEA) Group at Cornell University has focused on improving production techniques for existing greenhouse crops, and developing production procedures for new crops. Through manipulating parameters such as quantity and quality of light, CO<sub>2</sub> concentration, aerial and root temperature, and nutrient solution characteristics and composition, production times have been decreased, productivity increased, and new production strategies have been passed on to growers. With the success of producing greenhouse lettuce, focus has shifted to looking at other similar crops that can benefit from greenhouse production, particularly spinach.

A number of factors have driven the development of greenhouse hydroponic spinach production. First is the increasing demand for baby spinach due to the popularity of salad greens. Spinach is a crop rich in vitamins and nutrients, and baby-leaf spinach is more tender than mature bunched spinach. Currently, spinach is primarily produced through field production; however, there are a number of drawbacks to such systems. The increasing cost of diesel has made shipping from traditional production locales such as California and Florida more expensive. In addition, leafy greens such as lettuce and spinach do not store and travel as well as other produce such as tomatoes, cucumbers and peppers. Though lettuce and spinach can be shipped, it is a race against time to get them to the consumer as quickly as possible. Coupled with a public that wants to “eat local” this has led to the desire for locally produced crops. To provide year round production in population dense areas such as the Northeastern United States, greenhouses are the only option for local production, due to the severity of winters.

Hydroponic greenhouse production makes more efficient use of valuable water and nutrients. In field production, excess water and nutrients need to be applied, which can lead to contamination of both aquifers and surface water. In addition, the 2006 recall of field grown spinach due to *E.coli* contamination highlighted the inherent dangers of field production. Pathogens such as *E. coli* can be spread either through animal or insect vectors as well as direct runoff and in aerosols of dust, and it is extremely difficult to completely eliminate their threat from the field. Another potential infection avenue is the washing stage which is necessary to remove the dirt, grit and other residues including pesticides from the crop. Greenhouse spinach does not need to be washed as it is grown in soilless culture and pesticides are not required. An added benefit of removing the washing stage is increased shelf life as well as removing another common avenue of food borne pathogen spread.

The demand for greenhouse hydroponic babyleaf spinach has led to considerable research on growing techniques. However, to date there is no commercial production in the United States, and there is limited production overseas. The primary reason for this lack of production is the water mold *Pythium aphanidermatum* (further referred to as *Pythium*). This organism is present in the natural environment and attacks roots in a variety of field-grown crops. However, it is particularly virulent in hydroponic production systems due to its ability to spread through the nutrient solution. Its zoospores possess flagella that allow them to move from root to root. This spread of disease is less of a problem in field conditions where direct water paths are more limited. However, a highly mixed nutrient solution provides ideal conditions for spread. *Pythium* zoospores attack the roots of a plant and form mycelia that can release further zoospores or directly spread to other roots. In addition to stunted growth, plants with *Pythium* damaged roots can wilt and even die depending on the severity of the attack. In her doctoral dissertation, Katzman (2003)

investigated some of the factors that influence the progress of *Pythium* infections. To expand on this work and to continue the development of hydroponic spinach production, NYSERDA (New York State Energy Research and Development Authority) commissioned a study (Albright et al., 2007) to investigate techniques for controlling *Pythium* in the nutrient solution, focusing on traditional and novel water sterilization techniques. As was found in this study, and previously recommended by Katzman, the key to producing spinach is to manage, rather than attempt to completely eliminate the disease. This dissertation seeks to apply the techniques of Risk Analysis to arrive at recommendations for successfully producing hydroponic babyleaf spinach in the presence of *Pythium*.

#### *Why use Risk Analysis?*

According to the Society of Risk Analysis (SRA, 2009), risk analysis is defined as “Detailed examination of a facility, process, or materials that includes risk assessment, risk evaluation, and risk management alternatives performed to understand the nature of unwanted, negative consequences to human life, health, property, or the environment; an analytical process to provide information regarding undesirable events; or the process of quantification of the probabilities and expected consequences for identified risks.” Risk analysis is a systematic means of identifying, assessing, and managing risk. The overall goals are to minimize losses, and to maximize opportunities. These are precisely the goals of any farmer, and particularly greenhouse growers who are faced with an organism such as *Pythium*.

The major steps involved in a risk analysis entail first identifying the risks that face a particular endeavor, evaluating the likelihood of encountering these risks, and then developing and implementing a management plan to minimize the impact of the risks. This is also the procedure that the CEA program has taken in attempting to

develop hydroponic babyleaf spinach production. Following initial difficulties in achieving consistent production, Katzman identified *Pythium* as the “root” cause. This was backed up by other experiences described in the literature. Further experiments and contact with other groups attempting to produce spinach hydroponically quickly determined that the threat of *Pythium* is a problem of when, and not if, an outbreak will happen. This assumption then led to the NYSERDA sponsored work to evaluate techniques for dealing with *Pythium*, which relates to managing the risk. This dissertation seeks to build on all of this work, to develop and test, in simulation, management plans/production guidelines, to reduce the risk a grower faces when they produce babyleaf spinach hydroponically.

## **1.2 RESEARCH OBJECTIVES:**

The overall objective of this dissertation is to develop and use quantitative methods to compare the risk of *Pythium aphanidermatum* infection in different hydroponic baby spinach production systems and identify the least risky alternatives. Such quantitative comparisons could then be used by a grower to determine which production system suits their particular needs, and/or by a researcher interested in further investigating/validating the risk of *Pythium*.

To achieve this objective a number of tasks were necessary:

1. to investigate current and non-traditional techniques for the control of *Pythium aphanidermatum* in hydroponic nutrient solutions,
2. to apply the findings of the solution treatment techniques in the development of production strategies,
3. to develop a linked greenhouse, crop, and disease model that can be used to better understand the dynamics of a *Pythium aphanidermatum* infection, and to simulate and evaluate different production strategies,

4. to simulate these production strategies under the greenhouse crop disease model with crop damage metrics, in order to develop expected frequencies of *Pythium* outbreak for each strategy.

### **1.3 RESEARCH APPROACH**

*Pythium aphanidermatum* has been identified as the main obstacle/risk in the production of hydroponically grown babyleaf spinach. This organism is so prevalent that even crops grown in fresh nutrient solution will often show signs of *Pythium* damage at harvest time. If this nutrient solution is subsequently reused for another crop, the damage only multiplies.

In chapter two of this dissertation, new and existing water treatment technologies were evaluated at a benchtop scale in an effort to manage infections by destruction of *Pythium* zoospores. The results of this work were presented at the 2005 ASABE (American Society of Agricultural and Biological Engineers) annual conference, and the paper was submitted to the conference proceedings (Shelford et al., 2005). This work was funded by NYSERDA, and is also a part of the report made to NYSERDA.

An experiment was then set up to test the promising technologies identified in the benchtop work, and evaluate the performance of these technologies in actual continuous production. The results of this work were presented at the 2006 ASABE annual conference (Shelford et al., 2006), and make up chapter three of this dissertation. This work was funded by NYSERDA and is also included in the final report to NYSERDA.

The primary finding reported in the third chapter is that managing the disease through crop cultural practices, rather than attempting to destroy *Pythium* zoospores in solution, is the best way to deal with *Pythium*. Through a combination of reducing the

nutrient solution temperature to slow the reproduction rate of *Pythium*, and removal of infected roots through short harvest cycles, it was shown that infections do not begin, and that systems can recover from severe infections. Reducing nutrient solution temperatures, and placing strict restrictions on the amount of time a crop can spend growing, limits the flexibility available to a grower and imposes costs which may or may not be acceptable. To determine the true limits of the system, a massive experimental effort would be required, the results of which may or may not apply to situations found outside of the Cornell greenhouses. It was decided that a simulation of the greenhouse, crop and disease system would at least indicate the production strategies which merit further investigation.

The fourth chapter of this dissertation provides a greenhouse thermal environment model that uses the climate of the greenhouse location as input. Of particular concern is the temperature of the nutrient solution, and the effects of supplemental lighting and shading for crop daily light integral control on the temperatures within the greenhouse, and the quality and timing of the light. A portion of this program was used to assist in the evaluation of the LASSI2 (Light and Shade System Implementation with CO<sub>2</sub> supplementation) algorithm undertaken as a USDA SBIR-funded project, in conjunction with CEA Systems Inc. In this dissertation the greenhouse simulation model serves as an input to the spinach crop, and *Pythium* disease models.

The fifth chapter uses data collected during the NYSERDA project, but not included in the final report, of the growth of a spinach crop over time. Using this data, the mechanistic growth model SUCROS (Goudriaan and van Laar, 1994) was adapted to baby spinach production. SUCROS was selected as a framework for the growth model because it differentiates the growth into root and shoot portions, and determines the growth in these organs as a function of photosynthate production. It was felt that

such a model would be adaptable to modeling the effects of root damage due to *Pythium* on the production of photosynthate, and hence on the growth of the entire plant.

The sixth chapter of this dissertation uses the data Katzman collected to model the effects of temperature, zoospore concentration, and time of inoculation on the growth and spread of *Pythium* in a spinach crop. From this work, the parameters and form of model necessary to describe an infection of *Pythium* were developed. Because Katzman's work was in single crops of spinach, data collected during the NYSERDA disease project was used to validate the model in multi-cohort common nutrient solution use.

The seventh chapter ties the previous chapters together. The production strategies suggested by the work of the third chapter are investigated through simulation using the greenhouse model of chapter four, the crop growth model of chapter five, and the *Pythium* disease model of chapter six. Thirteen years of measured climate data for Ithaca, NY were used as the input for the greenhouse simulation, and assumed distributions for the values of the crop growth, and *Pythium* disease model parameters were used as input to a Monte-Carlo simulation of the different production strategies. The results of the simulation are expected frequency of *Pythium* outbreak for each production strategy. This information will provide insight into production strategies to investigate further, before commercial adoption.



## CHAPTER 2: BENCH-TOP EVALUATION OF MANAGEMENT OPTIONS

### 2.1 INTRODUCTION

*Pythium aphanidermatum* is arguably the largest hurdle to be overcome in the development of commercial hydroponic spinach production systems. This ubiquitous root disease organism can quickly spread through a crop, killing roots and devastating production. Because this pathogen is waterborne, recirculating hydroponic systems are ideally suited for quickly spreading the infection.

To suppress the dispersal of the organism, various treatment technologies have been tried. Pasteurization and ultraviolet systems have demonstrated their ability to successfully eliminate *Pythium* (Zhang and Tu, 2000, Runia et.al., 1988, Stanghellini et al., 1984). However, there are drawbacks associated with their operation. Even with the use of efficient heat exchangers, pasteurization is an energy intensive operation that may not be feasible for large scale spinach production. Ultraviolet radiation, though requiring less energy to achieve adequate *Pythium* destruction, has been demonstrated to destroy the organic chelators which are so important in maintaining iron solubility. Chemical means of control such as metalaxyl and other fungicides are not an option, as spinach is a food crop. New technologies that do not suffer from these drawbacks would make a commercial spinach production system more viable.

A preliminary step in evaluating a new treatment technology is to determine its effectiveness in pathogen destruction. Ultimately the best test of a system would be to construct it to scale and operate on a growing spinach crop; however, this is certainly not always feasible. Because of the dilute nature of the pathogen, it is not feasible to directly attempt to count zoospores with the use of a hemocytometer. Indirect counting techniques such as serial plating on selective agar, with colony counting are required. However such a system requires a relatively small (~1 ml) sample size,

which, if the sample is not well mixed, can lead to erroneous results. Plating also requires the use of expensive and perishable selective antibiotics. In this research we tried a simpler, inexpensive alternative that utilized seedlings of the spinach crop itself in the form of a bioassay.

We used this bioassay to determine the effectiveness of two new treatment technologies as compared to treatments known to be effective against *Pythium*.

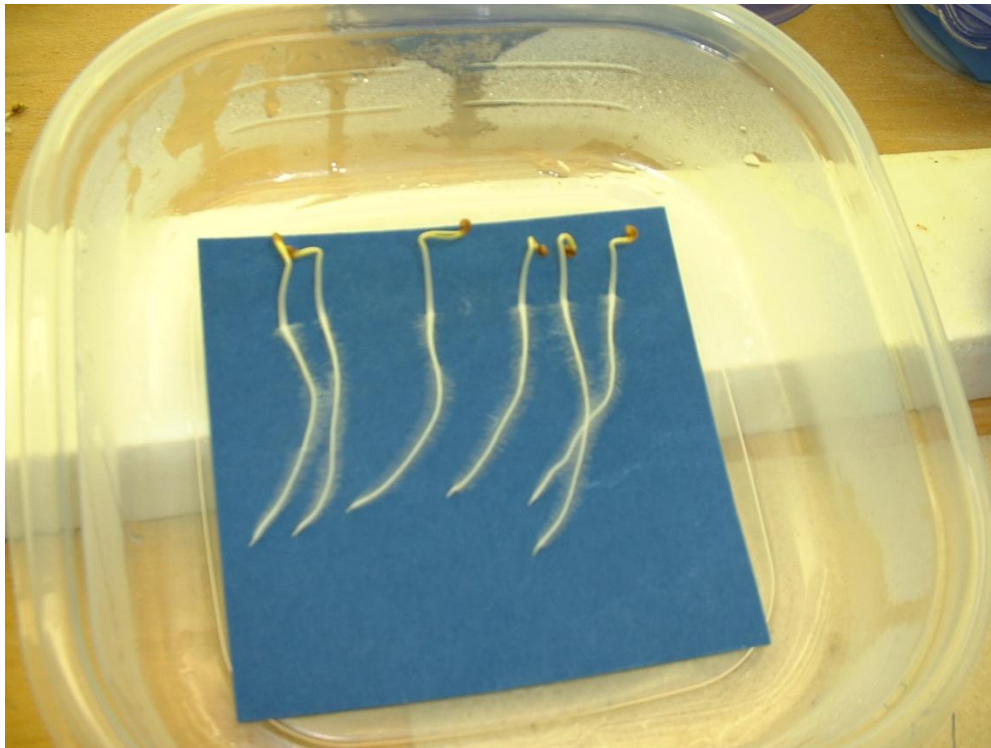
## **2.2 METHODS AND MATERIALS**

### **2.2.1 *Pythium* bioassay**

The concept of using seeds or seedlings in the study of *Pythium* is not new. Watanabe (1984) used cucumber seeds to determine the presence of *Pythium aphanidermatum* in soil, and to get a crude estimate of the number of propagules per gram. Because of reported difficulty in isolating *Pythium* from nutrient solution in an NFT system, Vanachter (1995) also used cucumber seedlings to bait *Pythium*.

To quantify the damaging effects of *Pythium* on spinach seedlings a modified technique was developed. Each bioassay consisted of a sandwich sized Ziplock™ container which contained a 100 cm<sup>2</sup> (4" x 4") piece of blue blotter paper (Hoffman Manufacturing) presoaked in deionized water. Ten spinach seeds (*cv.* Alrite) were then placed 2 cm in from one edge of the blotter with the radicles aimed away from the edge. This alignment prompts the roots to grow down the blotter paper and attempts to keep the roots parallel with each other. To hold the seeds in place and create a uniformly humid environment, a 2 x 10 cm strip of wetted germination paper was placed on top of the seeds. The containers were arranged in a growth chamber on a 30 degree slope with the edge containing the seeds at the high end. Following incubation at 25C for 48 hours in darkness, the seeds developed shoots of approximately 2 cm and roots of approximately 5 cm (Figure 2.1). The covering piece

of germination paper was then removed and the assays tubs were filled with 175 ml of the solution of interest. The angle of the germination box was adjusted to ensure the entire root was submerged in the solution and that the shoot was not. Following inoculation, bioassays were grown for an additional 48 hours at 25C and 100  $\mu\text{moles m}^{-2} \text{s}^{-1}$  of continuous light. Bioassays were then drained of solution and examined under a dissecting microscope (Bausch & Lomb Stereo Zoom 4) at a magnification of 1.5X.



**Figure 2.1** Spinach seedling bioassay before application of solution of interest.

Damage to the roots was classified into four categories: lesions, superficial exterior streaking, light interior streaking and dark interior streaking. Lesions appeared as distinct brown or yellow discolorations on the roots. Superficial exterior streaking was seen as a continuous discoloration with or without the presence of definite lesions. Light interior streaking was a discoloration interpreted as lesions that

had penetrated the root and begun to spread. Dark streaking was an indication of more serious damage. Lesions were recorded as numbers per root, and streaks were recorded as length in cm per root. To allow comparison between roots, damage from streaks and lesions was weighted to provide a single measure of damage for comparison between treatments. Though somewhat arbitrary, the weighting coefficients used attempted to reflect the relative severity of the damage each category represented.

To determine the efficacy of the treatment systems and to determine the levels of damage associated with differing concentrations of *Pythium* zoospores (zoos), bioassays were infected with known levels of zoospores in a dilution series. The dilutions used were 0 (control), 1 and 10 zoospores ml<sup>-1</sup>. Because the experiments were conducted on multiple occasions these dilution series were repeated with every experiment to allow comparison, and to allow accounting for any differences in *Pythium* concentration. This is necessary because the initial concentrations of *Pythium* solution, following removal from the mycelium, are determined with a hemocytometer and then diluted several times to achieve the target concentrations.

### 2.2.2 Treatment Systems

Four treatment systems, Electrochemical (EC), Pasteurization, Sonication and Ultraviolet (UV) sterilization were evaluated for their effectiveness in reducing root damage due to *Pythium aphanidermatum*. Treatment conditions of pasteurization and UV sterilization that have been demonstrated to be effective against *Pythium* were used to verify the effectiveness of the recommendations and allow comparisons with the new technologies.

Nutrient solution (1/2 strength Hoaglands) was infected with *Pythium* to a dilution of 10 zoospores ml<sup>-1</sup>, before the various treatments were applied to it. Control

solutions with concentrations of 0, 1 and 10 *Pythium* zoospores ml<sup>-1</sup> were used to provide a comparison with the solutions collected from the four systems post treatment. All conditions were replicated with two bioassays used per sample.

### *Electrochemical Treatment*

Spinu et al. (1998) described a system that uses electrodialysis to continuously adjust the pH of the nutrient solution to a desired level for regular pH control. The electrochemical treatment system uses the same principles, but takes the pH and ORP to greater extremes.

The electrochemical treatment unit consisted of two compartments 10 cm x 10 cm x 15 cm separated from each other by a cation exchange membrane. In the cathode compartment a stainless steel electrode measuring 9.5 cm<sup>2</sup> was placed against the side of the compartment farthest from the membrane (Figure 2.2). In the anode compartment, a titanium plated electrode of identical size was placed on the opposite side. One liter of the solution with a concentration of 10 zoos ml<sup>-1</sup> was placed in each



**Figure 2.2** Electrochemical pH adjustment apparatus.

compartment, and 100 VDC was applied across the electrodes. During treatment, solutions in both compartments were constantly mixed with glass stirring rods. Treatment times were 1, 2, 5, 15, and 30 minutes. The final pH values in the cathode compartment corresponding to these times were: 4, 3.5, 3, 2.3 and 2. Following the treatment duration, 400 ml of solution was removed from the cathode chamber and then brought to a pH of 5.8 with 1M KOH. Current through the electrodes was measured with a Greenlee multimeter (model # DM-200).

### *Pasteurization*

Pasteurization has been used for many years as a means of removing pathogens from nutrient solutions, and considerable research has been conducted as to its efficacy. Several authors recommend a temperature of 95C for 30 seconds, (Runia et al., 1988, Rey et al., 2000), however others have demonstrated that lower temperatures and longer durations such as; 55C for 2 minutes, (Tu and Zhang 2000), and even 51C for 15 seconds (Runia and Amsing, 2000) can be effective as well.

In the pasteurization treatment, two liters of nutrient solution was heated on a Thermolyne hotplate (Type 2200) until reaching a target temperature of 60C and 95C (measured with a Digi-Sense digital thermometer model # 8528-20). At this time enough zoos were added to bring their concentration to 10 zoos ml<sup>-1</sup>. The solution was then thoroughly mixed with a glass stirring rod and at set time intervals 400 ml of solution was removed, placed in an Erlenmeyer flask, and run under cold tap water until the solution reached ambient temperature. Set times of 30 seconds, 1 and 2 minutes were used for the 60C solution, and 15, 30 and 60 seconds were used for the 95C solution.

### *Sonication*

Sonication employs the phenomenon of cavitation to lyse cells, rendering them harmless. Cavitation is the formation of pockets of vapor in the solution due to a reduction in the pressure in the liquid. A probe vibrating at a high frequency (generally 20 to 40 kHz) causes cavitation in the solution. The rapid formation and collapse of the pockets of vapor create extreme conditions under which cells are destroyed. Tu and Zhang (2000) examined the use of sonication for eliminating *Pythium* in nutrient solution in a bench top test. They found that after 1.5 minutes of sonication 100% of zoos and cysts were destroyed. They applied their treatment in a batch system where a probe vibrating with an amplitude of 120  $\mu\text{m}$  and frequency of 20 kHz was inserted into a beaker containing 150 ml of solution.

To test sonication on a flow through or continuous basis we utilized a Misonix flowcell (model # 800B), that attached to our sonicator probe. Sonication was applied with a Misonix 500W ultrasonic generator (Model #2020XL), and converter (model CL4) with a 13mm (1/2") probe. The probe was threaded allowing it to screw into the flowcell. Flow was directed up through the base of the flowcell through a 3.2 mm (1/8") orifice plate where it contacted the tip of the probe. Solution then flowed around the probe and exited the flowcell at the highest collection port. The solution was pumped through the flowcell with a Procon pump (model # C01607AFV) at a pressure of 700 kPa (100 psi) and at flowrates of 200, 400, and 600  $\text{ml min}^{-1}$ . At each flowrate the sonicator was set at amplitude setting seven and nine, corresponding to amplitudes of 120 and 150  $\mu\text{m}$ .

Following the experiment the equipment was run with the power turned off to the sonicator to determine whether passage through the pump and system had any effect on zoospore infectivity.

## *Ultraviolet*

Ultraviolet radiation is a very popular method to disinfect of recirculating nutrient solution. Stanghellini (1984) found that a dose of  $90 \text{ mJ cm}^{-2}$  provided adequate disinfestation, and Tu and Zhang (2000) found that  $80 \text{ mJ cm}^{-2}$  was sufficient to kill 100% of *Pythium* zoos and cysts.

Ultraviolet treatment of the nutrient solution was achieved with an 8W flowthrough ultraviolet reactor (model Aqua Ultraviolet 8W) which utilized a low pressure bulb to produce UV-C at a wavelength of 253.7 nm. UV dosage was regulated by varying the flowrate through the reactor. UV doses of 120 and  $240 \text{ mJ cm}^{-2}$  were examined. To ensure uniform dosing of the solution, approximately 3 liters of nutrient solution was passed through the reactor before sampling. Between runs, the outlet of the reactor was disassembled and cleaned to prevent cross contamination.

In normal operation UV sterilization equipment requires filtration to remove particles that prevent the transmission of the UV light. Because we were working with new nutrient solution no such sediment was present, and so filtration was not used.

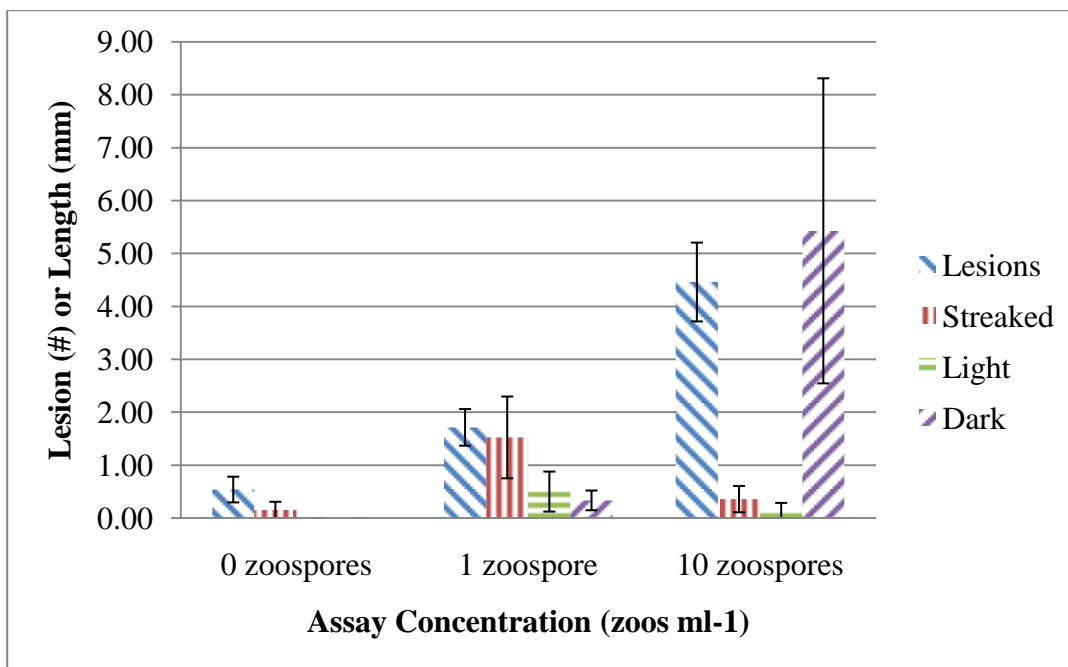
## **2.3 RESULTS AND DISCUSSION**

### **2.3.1 Bioassay**

To determine the level of damage associated with a particular concentration of zoos a dilution series was conducted. Figure 2.3 illustrates the average damage visible on the root of each plant in the bioassay. In the case of dilutions of 0 and  $1 \text{ zoos ml}^{-1}$  we can see that most of the damage is fairly minor, and constrained to exterior lesions and streaking. In the  $10 \text{ zoos ml}^{-1}$  case we can see considerably more damage as evidenced by the relatively large amount of dark interior streaking. Presumably the infection has moved into the roots and advanced further than in the other two dilutions. Low level damage in the  $0 \text{ zoos ml}^{-1}$  treatment was unexpected, and was



most likely not due to *Pythium*, but other organisms or mechanical damage. In a side experiment to try and determine the cause of the damage, it was still present even after surface sterilization of the seed with chlorine bleach.



**Figure 2.3** Average root damage due to varying concentration of *Pythium* (Error bars are  $\pm 1$  SE)

Because care was taken to minimize the chances of biological contamination, other factors such as mechanical damage or a natural reaction of the root going from a humid environment to being submerged, may have contributed to the discolorations on the root which were then interpreted as lesions and streaking. Kinking of the roots was observed in the 10 zoos ml<sup>-1</sup> and to a lesser extent in the 1 zoos ml<sup>-1</sup> dilution and seemed to correspond with damage due to lesions. The roots of the uninfected dilution were straight, without the abrupt directional changes visible in the more heavily damaged roots.

### 2.3.2 Treatment Systems

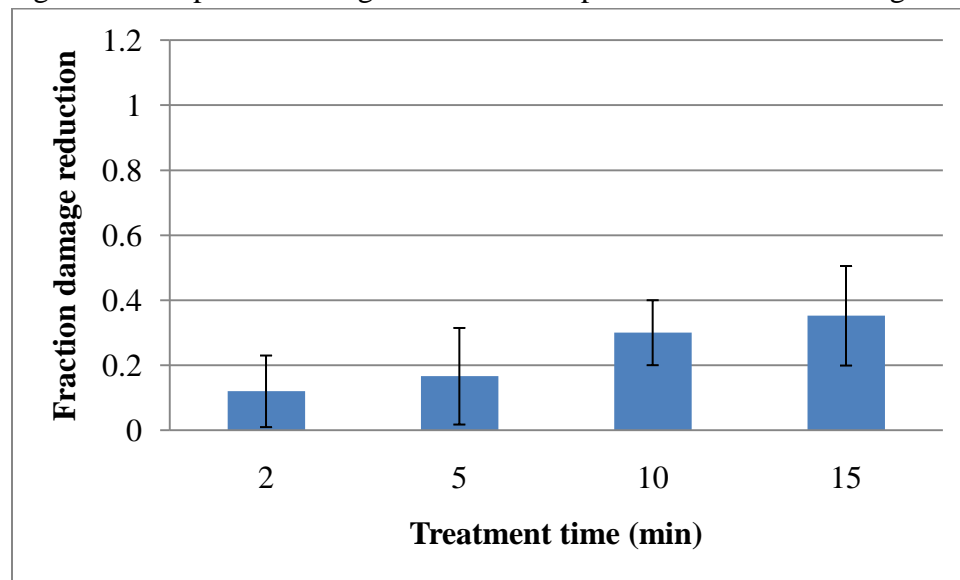
To facilitate comparing the damage present on the roots following treatment an overall damage score was calculated by weighting the four categories. As lesions are the most minor damage each lesion contributed a value of one to the overall damage score. Exterior streaking was considered to be relatively minor damage and every cm of streaking contributed two to the damage score. Light interior streaking was an indication of more serious damage and so each cm was considered to contribute a value of four. Dark interior streaking was a sign of major root damage and was given a value of eight per cm. Summing up the damage on each root from each category provides an overall level of damage for the root. The damage present in the uninfected control condition was subsequently treated as due to other factors and the average damage of the control was subtracted from the other conditions so that the remaining damage would be due solely to *Pythium*. The coefficients used to apply damage values to the roots are somewhat arbitrary, however adjusting them moderately does not substantially affect the relative overall damage.

Because we are primarily interested in reducing damage from *Pythium*, the percentage reduction of damage on each root was calculated by subtracting the damage present on the treatment roots from the average damage of the control solution infected at 10 zoos ml<sup>-1</sup> and then dividing by the average damage of the control solution infected at 10 zoos ml<sup>-1</sup>. Besides giving us the percentage reduction in damage, this procedure allows us to compare treatment systems that were run at different times, where it would be expected that the concentration of the initial solution might be slightly different from 10 zoos ml<sup>-1</sup> and/or other factors might influence the amount of damage caused on a particular day.

### Electrochemical

Minute gas bubbles started to evolve from each electrode (Hydrogen gas at the cathode and Oxygen at the anode,) when current was applied across the electrodes. The solution in the anode compartment began to turn cloudy after a few seconds, as the pH rose and some of the mineral salts began to precipitate out. Precipitate was not formed in the cathode compartment. During operation of the electrochemical unit an average 700 mA of current was drawn by the electrodes, and with a supply voltage of 100 VDC resulted in a power usage of 70 W. In the 15 minute condition the temperature increased to approximately 35C. Figure 2.4 presents a comparison of the percent reduction in damage at each Electrochemical duration.

Electrochemically reducing the pH and ORP does not appear to have a large impact on *Pythium*, or at least at the durations we examined. This technique might be more successful if durations were greatly increased to the order of several hours. Current could be turned off when a target pH is achieved, and then the solution allowed to react. However, long treatment durations would correspond to the need for very large retention ponds making this treatment option unfeasible on a large scale.



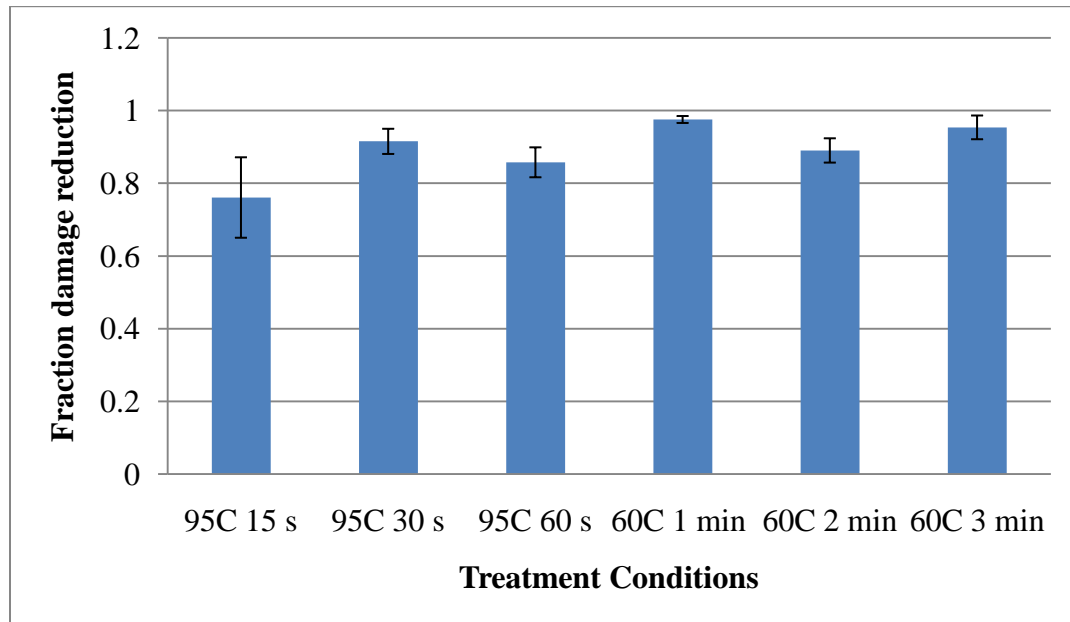
**Figure 2.4** Fraction damage reduction due to Electrochemical Treatment

### *Pasteurization*

Concentrated *Pythium* solution was added to preheated nutrient solution to bring the concentration of zoos to 10 zoos ml<sup>-1</sup>. We felt this was important because Tu and Zhang (2000) demonstrated that *Pythium* can be affected at temperatures above 45C if the duration is long enough.

After addition of the *Pythium* concentrate and the allotted treatment time had passed a sample was cooled using a 2 liter Erlenmeyer flask run under cold tap water. This caused an initially rapid temperature drop, and the solutions reached ambient temperature within 5 minutes. Figure 2.5 presents a comparison of the two treatment temperatures and the various treatment durations at these temperatures.

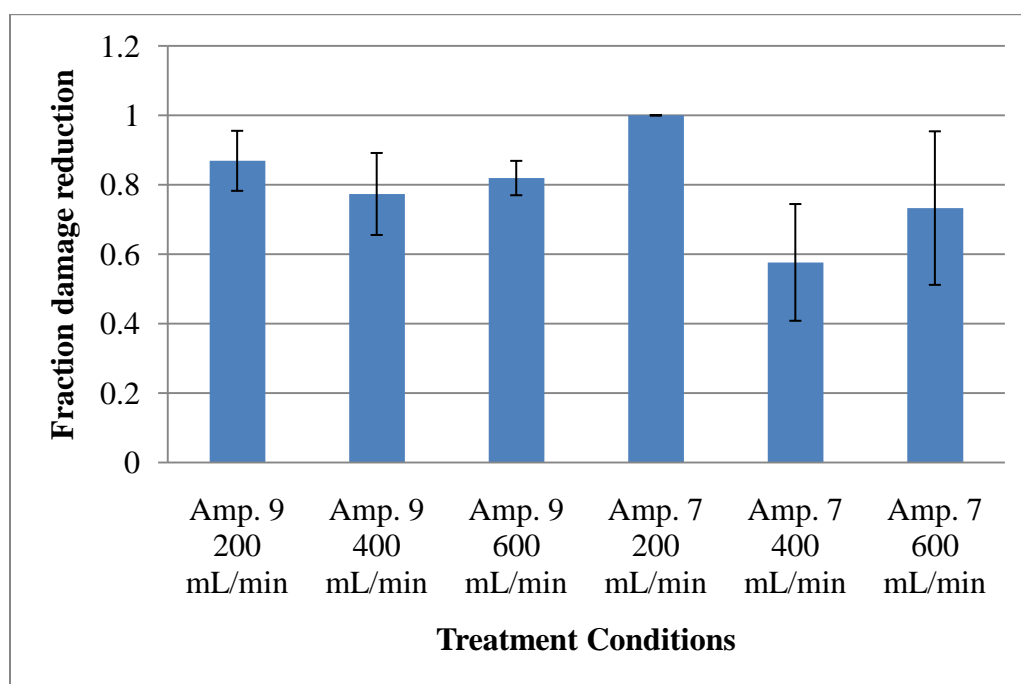
Both treatment temperatures seem capable of achieving a relatively good reduction in the level of damage. Though requiring a longer dwell time and subsequently a larger volume in the treatment system, a temperature of 60C is easier to attain and work with than 95C.



**Figure 2.5** Fraction damage reduction due to Pasteurization

### Sonication

The Misonix XL2020 used to generate the signal that causes the high frequency vibrations in the converter can also display instantaneous power usage as a percent of maximum. Larger amplitude corresponds to higher power consumption. At amplitude 9 the power consumption was approximately 70% of maximum (500W) corresponding to a value of 350 W. At amplitude 7 the power consumption was approximately 50% or 250 W. Figure 2.6 illustrates that continuous sonication is capable of successfully eliminating *Pythium*. The best results were achieved with an amplitude of 120  $\mu\text{m}$  and the lowest flowrate tested, 200  $\text{ml min}^{-1}$ , though satisfactory results were achieved with an amplitude of 150  $\mu\text{m}$ . However, to deliver an effective dose of sonic energy, our results indicate that flows greater than 200  $\text{ml min}^{-1}$  are not recommended for this size of generator.



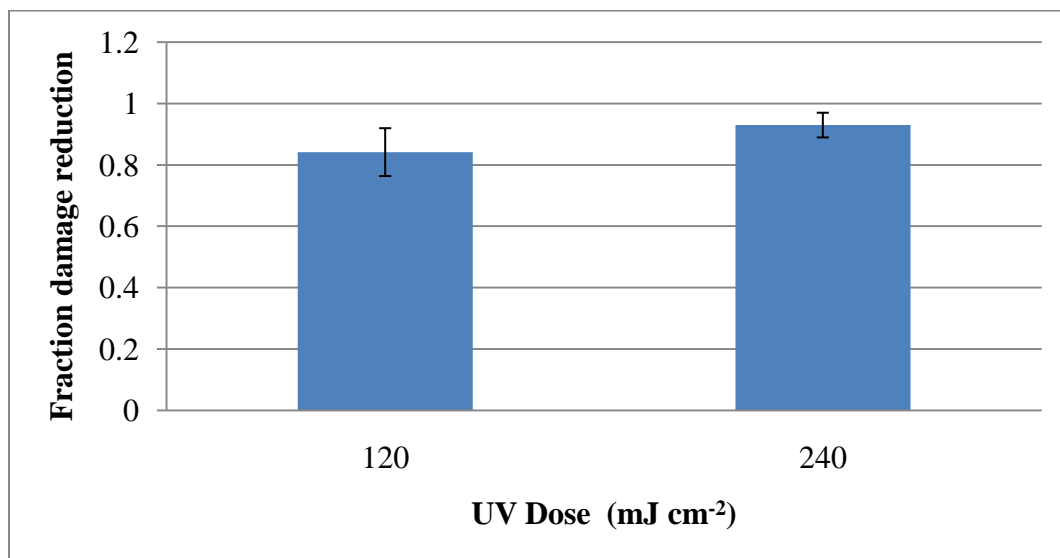
**Figure 2.6** Fraction damage reduction due to Sonication

Passage of *Pythium* containing nutrient solution through the pump and floccell with the sonicator turned off had little effect on the damage caused by the *Pythium*, indicating that any reduction in *Pythium* activity was due to sonication and not pressurization and turbulence.

The stated maximum recommended flowrate for the floccell is 0.66 liters/min, and it was developed for use with the XL2020 and other generators of a similar size. Larger generators capable of handling greater flows exist, so scaling up the flowrate is possible.

#### *Ultraviolet Sterilization*

Control of the flow through the UV treatment unit was with a ball valve mounted immediately before the inlet to the reactor. Closing of the ball valve caused more of the flow to divert through a bypass back to the reservoir containing the *Pythium* infected solution. The action of the bypass also served to keep the solution well mixed and prevented *Pythium* from settling on the bottom of the tank. Figure 2.7 shows the damage reduction due to UV-C doses of 120 and 240 mJ cm<sup>-2</sup>.



**Figure 2.7** Fraction reduction due to Ultraviolet Sterilization

Ultraviolet sterilization worked quite well, especially considering the energy input was only 8 W.

## 2.4 CONCLUSIONS

Though the uninfected control bioassays sometimes displayed symptoms of low levels of damage, the concept of utilizing spinach seedlings to measure *Pythium* levels appears valid. Though a single bioassay is not able to provide a precise count of the number of *Pythium* propagules present in a solution, when used in conjunction with a dilution series, a reasonable estimate can be obtained. The spinach seedling bioassay is well suited to comparing the efficacy of different treatment systems, and whether or not they are worth developing further. The seedling bioassay has the advantage of being very simple, which allows the testing of many conditions quickly, without expensive or complicated laboratory procedures, or expertise requirements.

Because the ultimate goal of the research project is to develop a viable commercial hydroponic spinach production system, testing on spinach gives a better indication of the actual damage causing ability of the organism. Tests were performed on the roots during the initial period where roots are most susceptible to *Pythium* damage. In the production systems under evaluation the crop is floated after 48 hours (germination time) and presumably this is when new roots would come into contact with *Pythium* in the nutrient solution. The bioassays are infected at age 48 hours to correspond to this stage.

Further refinements of the bioassay are required to better quantify how root damage progresses. Repeated examination of the same bioassay over the course of time following infection could provide a better picture of how lesions form, grow and spread. This information could be used to better define the coefficients used to weight

the various types of damage. Additional studies could also compare the accuracy of the bioassay with more traditional plating techniques.

The presence of damage on the roots even after the solution has undergone treatment does not mean the crop is doomed to failure. In the short crop cycle of spinach, *Pythium* may not have enough time to produce zoospores to damaging levels.

Of the new technologies tested, sonication demonstrated an ability to successfully eliminate *Pythium*. Sonication in a continuous flow mode is effective provided the flowrate doesn't exceed the capacity of the generator. Unfortunately the electrochemical treatment was found to be largely ineffective at the durations we tested. Though the system is very effective at manipulating the pH of the nutrient solution and can achieve extreme levels, these levels don't appear to be extreme enough to eliminate *Pythium* to a point on par with other treatment techniques.

The next step in the evaluation and selection of a treatment system for spinach production is a comparison of the operational costs. Other factors besides energy usage will have to be considered as well. In the case of UV sterilization, chelator destruction, and bulb and filter replacement need to be considered. In the case of sonication, the generator and converters are relatively expensive, and their operational lifetime needs to be factored into the cost of operating such a system.

Another consideration is the microflora which develops in a normal nutrient solution. Not all the microbes present are detrimental, and many can benefit the crop. Ultraviolet sterilization tends to be more effective on smaller celled organisms which can lead to the elimination of useful bacteria (Zhang and Tu, 2000). According to Tu and Zhang (2000) sonication has the opposite effect and is more effective against larger organisms. It may then be possible to target the larger zoospores and preserve beneficial microorganisms.



Regardless of the technology selected, a successful spinach operation will require diligence when it comes to monitoring the health of the crop. Perhaps a spinach seedling bioassay will be a tool growers can utilize to ensure success.

## CHAPTER 3: CONTINUOUS PRODUCTION EXPERIMENTS

### 3.1 INTRODUCTION

*Pythium aphanidermatum* is a water mold that can cause severe damage to crops through destruction of their root systems. It is of particular concern in hydroponic production systems because *Pythium* zoospores are mobile in water, and can quickly spread. *Pythium* affects a variety of crops, but spinach has shown itself to be particularly sensitive to this pathogen (Bates and Stanghellini, 1984). It is a leading obstacle to the commercialization of hydroponic spinach production.

A proven technique for growing spinach hydroponically is to change out the nutrient solution between crop cycles. This production model has been utilized in Japan where the market for whole plant baby spinach is strong. Towards the end of the production cycle the crop is allowed to strip the nutrients from the solution in preparation for disposal. Careful cleaning of the growing system between crops is then required before new nutrient solution is made up, and the new crop is installed.

Unfortunately, this model for spinach production requires the added expense of disposing of the nutrient solution and disinfecting the growing surfaces. To allow the re-use of the nutrient solution, considerable research has been conducted into means of eliminating *Pythium* (and other nutrient solution borne pathogens). Technologies that have proven effective have been heat (pasteurization) (Runia and Amsing, 2000), Ultraviolet radiation (Stanghellini et al., 1984), Filtration (Tu and Harwood, 2005), and Sonication (Tu and Zhang, 2000). These systems have proven effective and could be implemented for a batch type production system where the nutrient solution required to grow a crop is treated after the crop is harvested, in preparation for re-use with the next crop. However, the use of these systems in a continuous production (multiple stages of crop present in the same pond) floating hydroponics system has not been presented. Pond systems have distinct advantages over other hydroponic

production systems including: increased reliability and performance during electrical or other failure, easy materials handling and high space use efficiency, uniformity of the nutrient solution, conservation of water and nutrients and the large buffering of many parameters. It is for these reasons we have chosen to investigate means to produce baby spinach in such a system.

As a part of her dissertation, Katzman (2003) examined the effect of *Pythium aphanidermatum* concentration on damage to a spinach crop. In her system she transplanted spinach seedlings into a pond system at day 9, and then inoculated the pond with varying concentrations of *Pythium* zoospores. She then took destructive harvests at periodic intervals and categorized damage to the roots and shoots. She found that low levels of zoospores could be tolerated, provided the crop was harvested early enough (typically before day 21). She suggested a hydroponic system that can maintain a low enough concentration of zoospores in solution may be viable.

In a study of the effect of nutrient solution temperature on the spread of *Pythium*, Katzman (2003) found the damage caused by the pathogen was reduced in cooler conditions. In her study Katzman transplanted healthy 9-day-old seedlings into ponds with temperatures of 18, 24 and 30C, and then inoculated with *Pythium* zoospores. Zoospore concentrations were monitored and shoot weights were taken in periodic harvests, until final harvest at day 28. Katzman found soon after inoculation the concentration of zoospores in solution dropped below detection level for a period of time, before recovering to significant concentrations. The period of time when zoospores could not be detected in the solution was a function of temperature, with colder solution temperatures corresponding to longer periods before re-appearance of zoospores. The period of zoospore absence was approximately 10 days at 30C and 24 days at 18C. However, no matter the temperature, at day 28 the spinach crops

suffered some damage from *Pythium*, the higher the temperature, the more severe the damage.

In production of baby spinach the length of time the crop spends in the ponds is typically 14 days or less, if a consistent daily light integral of  $17 \text{ mol m}^{-2}$  is maintained. In this experiment we sought to examine whether this shortened crop cycle combined with various treatment methods to counter *Pythium* would allow the continuous production of hydroponic baby spinach. The continual removal of the source of inoculum (infected roots) before they are capable of releasing zoospores to propagate the infection should allow such a system to work. We sought to examine and compare two strategies to accomplish this goal. The first was to use two active techniques (filtration and ultraviolet irradiation) to remove zoospores from the solution and maintain a concentration acceptable to the crop. The second approach was to use a reduced nutrient solution temperature to increase the amount of time required by *Pythium* to complete its lifecycle to a period greater than the crop duration in the pond.

### **3.2 METHODS AND MATERIALS**

For the experiment, eight identical temperature controlled benches were constructed (Figure 3.1). The interior dimensions of the benches were 235 cm long by 35 cm wide, and 12 cm deep. To set the depth of the nutrient solution (half-strength Hoagland) to a height of 11 cm, a stand-pipe was used. The benches were supported at a height of 80 cm above the level of the floor, and 150 cm below the lighting array, which provided an average intensity of  $200 \mu\text{moles m}^{-2} \text{ s}^{-1}$  PAR. Nutrient solution from the drain cascaded into a 50 liter insulated reservoir, and through a coarse filter. The temperature of the nutrient solution in each bench was monitored by RTD (Omega RTD-810), and controlled through MATLAB with a USB DAQ

(Measurement Computing USB 1208-LS) switching an immersion heater (Aquatic Ecosystems VT201, 200W), and a cold finger circulating chilled water. The nutrient solution was pumped up to the bench by positive displacement pumps (Little Giant 2-MCHD).



**Figure 3.1** Overview photograph of spinach growing benches used in the multi-cohort evaluation of filtration, UV sterilization and temperatures suppression in continuous production.

Intact spinach seed (cv. Eagle) was seeded into Rediearth® growing medium, corrected to a moisture content of 3.0 to 1.0, in 132 cell styrofoam plug trays of area c.  $0.1 \text{ m}^2$  (Beaver Plastic, modified 288 cell flats). Once seeded and covered, the trays were placed in darkness in a growth chamber set at 26C, to germinate for 48 hours. Following 24 hours in the growth chamber c.  $100 \mu\text{mol m}^{-2} \text{ s}^{-1}$  of continuous light was provided on to the emerging seedlings to prevent stretching. At the time of flotation, trays were placed in the growing bench at the end closest to the inflow of nutrient solution from the pump. Space for the new float was made by pushing the existing floats towards the drain of the bench. To provide a consistent plant density, floats

were randomly thinned to a final count of 72 plants because germination percentage was variable and only about 70%. Target daily light integral was set for  $17 \text{ mol m}^{-2} \text{ d}^{-1}$ , and the day/night temperature was set at 23C. pH and EC were monitored daily and adjusted to targets of 5.8, and  $1400 \text{ mS cm}^{-1}$ , respectively when necessary. To maintain a constant system volume, the reservoir was replenished daily with modified half strength Hoaglands solution. Solution flow rate was balanced across all benches, and set to produce an average velocity of  $1 \text{ cm s}^{-1}$  under the floats.

Three treatment methods, filtration, ultraviolet and temperature reduction were examined, and each had an inoculated, and un-inoculated condition requiring a total of six separate production systems or benches. An additional two benches were maintained at 27.5C with and without inoculation, and served as controls for all three of the treatment methods. The filtration system was installed inline after the pump and consisted of two 25.4 cm (10") opaque polypropylene filter units (Cole-Parmer C29820-32). The first filter provided filtration to a level of 5 microns (Cole Parmer C-01509-15), and the second to a level of 1 micron (Cole Parmer C-01509-14). Ultraviolet treatment was provided by Aqua ultraviolet aquarium treatment units (Aqua Ultraviolet Classic 8), and provided a dose of UV equivalent to  $100 \text{ mJ cm}^{-2}$ .

To prepare for the experiment, crops in all benches were grown at 27.5C. New flats were added to the bench every three days, until a full cohort of 5 flats was in the system. At that time enough *Pythium* zoospores were placed into the systems to be inoculated, to bring the concentration to a level of  $100 \text{ zoospores ml}^{-1}$  (c. 7 million zoospores per bench). Bioassays utilizing spinach seedlings were prepared beforehand and samples from the growing bench and reservoir were taken 15 minutes after inoculation and applied to the bioassays. The subsequent damage to the bioassays indicated *Pythium* had successfully spread throughout the system. The production systems were allowed to run as usual for 24 hours, without application of remedial

treatments, to give the *Pythium* the fullest opportunity to establish itself before power was applied to the Ultraviolet units, the filters put in place, and the nutrient solution temperatures dropped.

### 3.3 RESULTS AND DISCUSSION

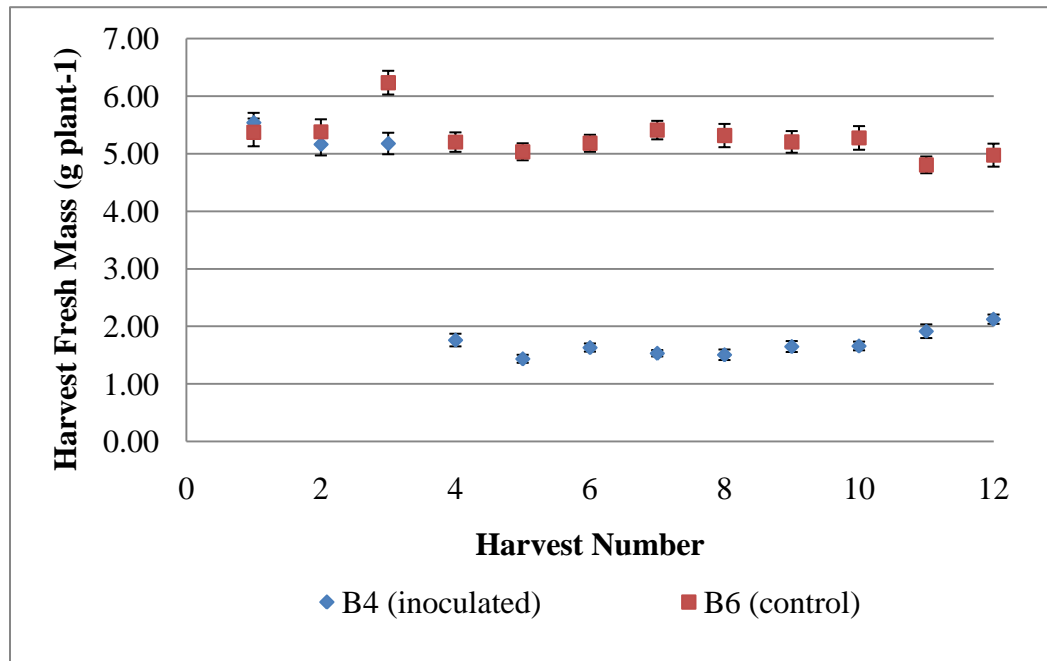
The experiment was run for 12 harvests (36 days) following inoculation and imposition of treatments. Identical procedure was followed for each stage of production, allowing comparisons to be made between cohorts over time. At the time of harvest the flat containing the oldest plants (taken from the end nearest the drain) was carefully removed and brought to a mini-pond where individual plant weights were taken for the entire flat. Plants in five central rows were retained for dry-weight measurement.

#### 3.3.1 Control

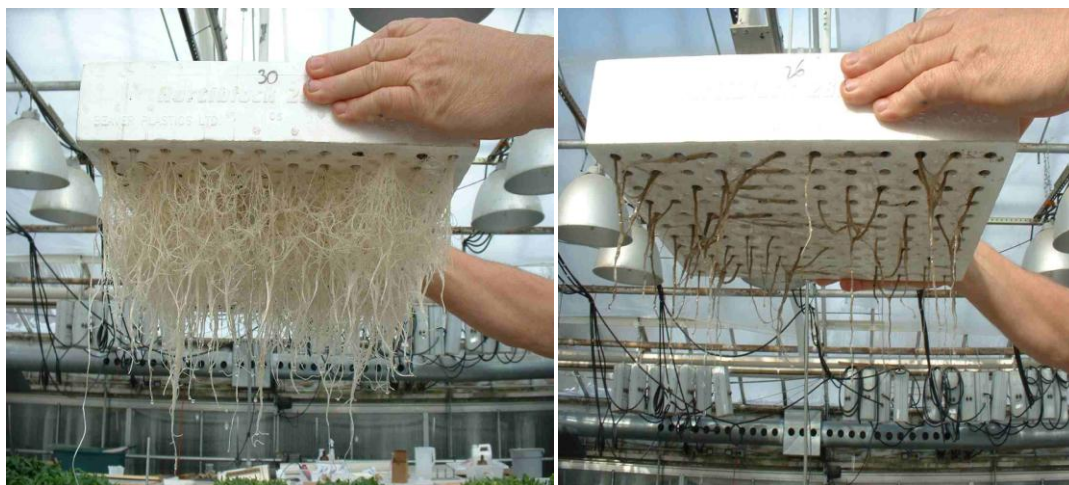
Figure 3.2 shows the mean plant fresh weight of both the inoculated and uninoculated benches that were untreated and maintained at 27.5C, for each of the 12 harvests of this experiment. As the experiment progressed, even heavily damaged flats showed a significant amount of growth from when they were first floated. This is likely due to the considerable root mass present in the flat that was not saturated with nutrient solution. Root material emerging from the bottom of the flat was attacked and destroyed quickly but the material in the root ball held out longer. However, even this material succumbed as the *Pythium* worked its way up the roots to a point where it caused already stunted seedlings to wilt.

There is little difference between the benches for the first two harvests and this is to be expected because *Pythium* had little time to attack the crop. The material in the first harvest was exposed to the zoospores for only 1 day, and the second harvest

for 4 days. At this point these flats had well established root systems and considerable shoot mass. By the third harvest the difference was more pronounced and at the fourth harvest dramatically different. Though the subsequent flats suffered extreme root damage, enough material was present to continue the infection. Figure 3.3 shows the difference in root appearance between the healthy and infected flats.



**Figure 3.2** Average plant fresh weights at time of harvest in control benches

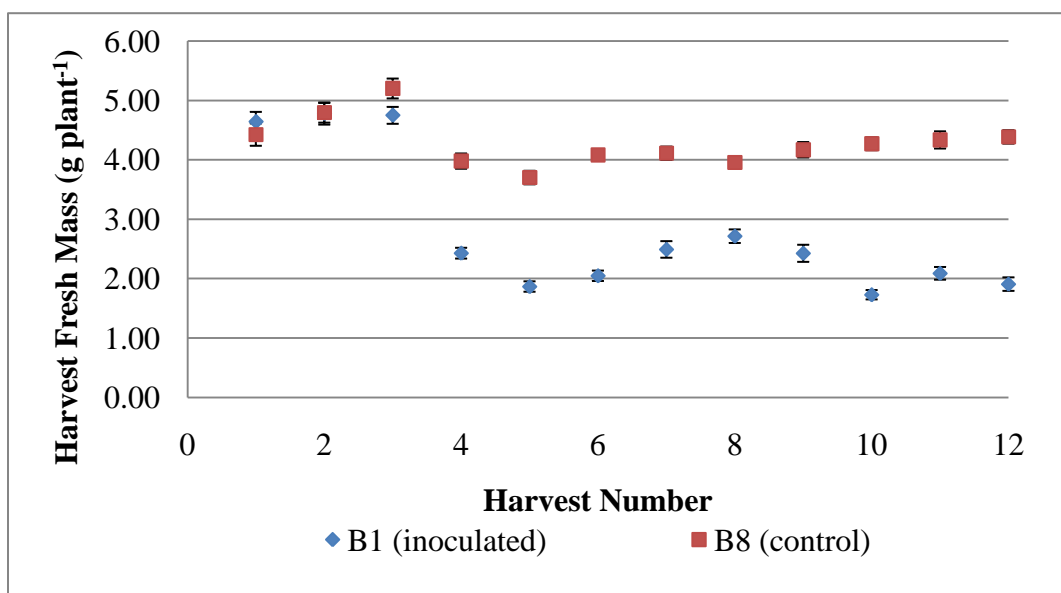


**Figure 3.3** Roots of uninfected (left) and infected (right) flats



### 3.3.2 Filtration

Figure 3.4 shows the mean plant fresh weight for the filtration systems. As in the case of the inoculated control bench damage at harvest 4 was severe. In subsequent harvests it appeared that the filtration system was allowing the infected bench to recover, but at harvest 8 the infection was able to reassert itself. It appears the filtration system was effective in removing free zoospores from the solution, however, not to a level that is tolerable by the crop.



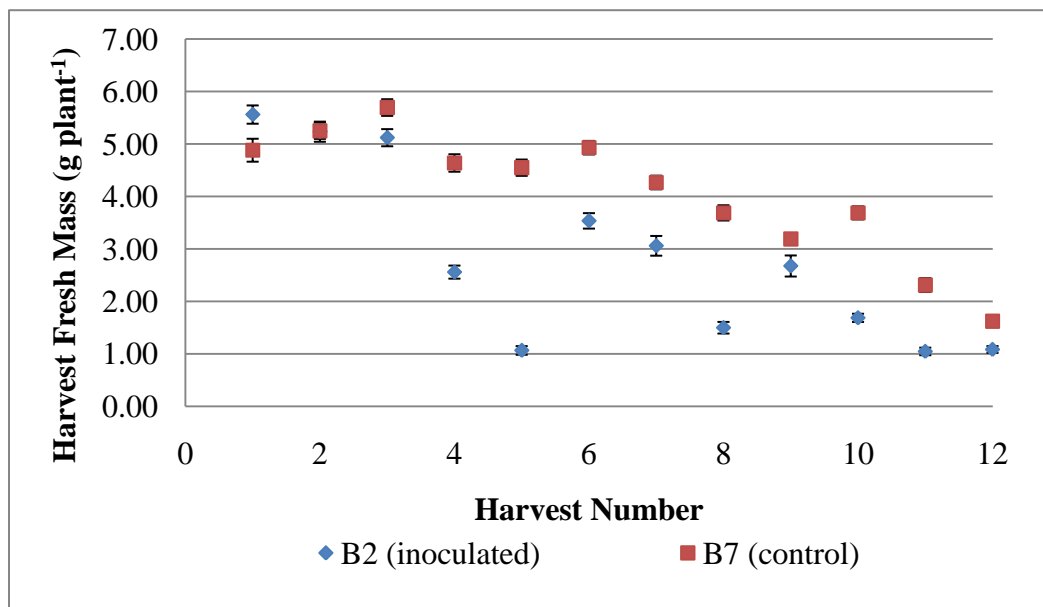
**Figure 3.4** Average plant fresh weight in the filtration, control (uninoculated) and inoculated systems, at time of harvest.

To improve the chances for active disinfection to succeed, a smaller volume of nutrient solution was used per plant than would be found in a typical pond system. A smaller volume was used, to decrease the turnover time of the pond, and to attempt to get directional bulk flow from the inlet to the outlet. It was thought that creating a velocity gradient would limit the migration of zoospores upstream towards the younger, more vulnerable plants. This also caused the roots to arrange themselves in the direction of flow which allowed the roots on the edge of a younger cohort to come into contact with older cohort root material. However, infection was not limited to

edge plants alone indicating zoospores were able to migrate upstream. This is not surprising as the flow velocity immediately beneath the floats was likely low enough to allow *Pythium* migration.

### 3.3.3 Ultraviolet Irradiation

Figure 3.5 shows the results of the ultraviolet irradiation systems. As in the case of filtration, the UV system was not able to prevent the *Pythium* infection from propagating. Again there appeared to be an initial recovery (even better than that observed with filtration) followed by a relapse. In addition, the control bench which was subjected to UV treatment but without inoculation also fared poorly. During this time, leaves gradually became pale yellow-green in color and failed to grow as well as the other benches. Ultraviolet radiation has been shown to destroy the chelator responsible for keeping iron soluble in a hydroponic solutions (Acher et al., 1997). Though new nutrient solution containing the complete mix of required elements was added daily through topping up of the reservoirs, iron content of the solution dropped to zero, and it is likely that the crop suffered from iron chlorosis.

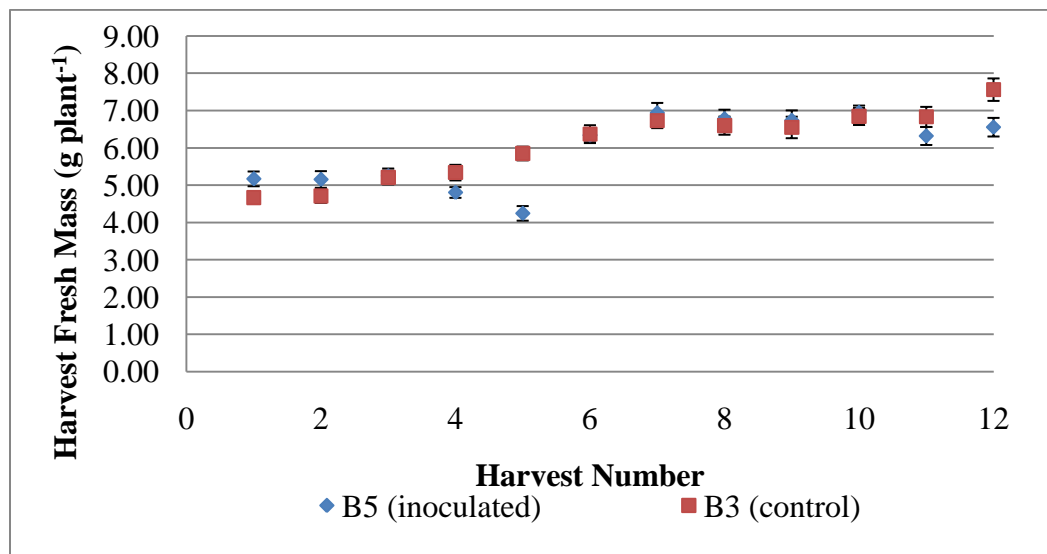


**Figure 3.5** Average plant fresh weight in the Ultraviolet systems at time of harvest

### 3.3.4 Root zone temperature depression

Of the treatment systems tested, temperature depression appeared to perform the best, as can be seen in Figure 3.6. The initial dosage of *Pythium* was enough to cause considerable damage to the crop, and we see this in reduced fresh weights in the fourth and fifth harvests. The harvest from the fifth cohort suffered the most damage which is to be expected, as it was the youngest material present at the time of infection and the zoospores added to the solution obviously set it back. However, the infection did not propagate and it seems damage was limited only to those plants directly exposed to the initial infection. Visual inspection of roots showed it took several additional harvests before all traces of root disease disappeared, but the remnant infection was so slight it did not manifest itself in shoot biomass reduction.

In comparing the average plant fresh weights among the controls it is apparent spinach performed better under cooler root conditions. A steady increase in average plant fresh weight until harvest 6 is apparent. This is to be expected as the crops spent more and more time under the more favorable temperature condition until harvest 6, at which point the entire crop cycle was at a root zone temperature of 20C.



**Figure 3.6** Average Plant fresh weight in the temperature depression systems at harvest.

### 3.4 CONCLUSIONS

Though it initially appeared active disinfection through filtration and ultraviolet irradiation would prove a viable means of suppressing *Pythium* for continuous pond culture, each of these systems was unable to eliminate the infection. It was clear they were having some positive effect as the crops in the inoculated control (no treatment) bench generally did not perform as well. However, the effect was not great enough to eliminate the infection or to be commercially useful.

Temperature depression appeared to work very well. We believe 20C was low enough a temperature to slow the reproductive cycle of *Pythium* to a point where it could not reproduce before the source of inoculum for the next generation was removed.

While temperature depression worked very well in eliminating the effects of a heavy infection of a virulent strain of *Pythium aphanidermatum*, there is no guarantee that more virulent and/or rapidly reproducing strains of *P. aphanidermatum* do not exist for which the method will not work. If such strains should be encountered, one recourse would be to use a yet lower solution temperature and shorter crop cycle. Other species of *Pythium* such as *Pythium disoticum* have temperature optima lower than *Pythium aphanidermatum* and could potentially thrive at 20C (Bates and Stanghellini, 1984), though Katzman (2003) was not able to isolate that species in nutrient solution taken from a commercial scale lettuce production system, located in Ithaca, NY.

## CHAPTER 4: GREENHOUSE TEMPERATURE SIMULATION MODEL

### 4.1 INTRODUCTION:

In Katzman's dissertation (Katzman 2003) and subsequent *Pythium* studies (Albright et al., 2007), the central role of nutrient solution temperature on the reproduction and spread of *Pythium* was identified. Before evaluating the performance of hydroponic babyleaf spinach production techniques in a commercial setting, a means of predicting the nutrient solution temperature in a greenhouse as a function of climate is required. In addition, because it is assumed that production will occur in a modern greenhouse with cooling and heating systems, supplemental lights and shade, the greenhouse simulation model must take into account the technologies available to a grower to optimize the growing environment, and the effects of these systems on the plant environment.

Many greenhouse models have been developed, including ones that incorporate shading systems; however, most models stop at either the crop canopy or the ground surface temperature. In the hydroponic babyleaf spinach system developed at Cornell, individual spinach plants are grown in Speedling type trays filled with a peatlite mixture. These styrofoam trays are then "floated" in a nutrient solution pond typically 20 to 30 cm deep. A similar system is used commercially to produce hydroponic lettuce; however, rather than use the densely packed speedling trays, the individual lettuce plants are spaced (and respaced depending on their stage of growth) on 2.5 cm (1") thick styrofoam boards. The need to model nutrient solution temperature as a function of climate is highlighted by the need to control the solution temperature in lettuce production ponds, even though this crop is less susceptible to *Pythium* than spinach. In the summer months pond temperatures can increase up to 30C if left uncooled; a temperature that would quickly lead to the rapid spread of *Pythium* and extreme damage to spinach production.

In one of the first forays into studying the commercial production of hydroponic spinach, Bates and Stanghellini (1984) identified seasonal effects of *Pythium* outbreaks in their greenhouse in Arizona. With a range of pond temperatures from 17 to 27C they found the dominant species of *Pythium* changed over the course of the year. Above 23C, *Pythium aphanidermatum* was the predominant species, and below 23C, *Pythium disoticum* dominated. In her dissertation, Katzman (2003) found that maintaining a nutrient solution of 18C slowed the development of *Pythium aphanidermatum* when she inoculated her spinach crops. In the NYSERDA spinach disease project (Albright 2007), temperatures of 20C were found to prevent the spread of *Pythium* infection from cohort to cohort grown in the same pond, by effectively breaking the reproductive cycle of *Pythium* through the removal of infected roots before they had the opportunity to release zoospores.

The key to successfully overcoming *Pythium aphanidermatum* in hydroponics is to reduce the nutrient solution temperature to the point where the growth and maturation rate of *Pythium* is slowed enough so the crop can complete its growth cycle, and to harvest before infection has a chance to spread. The modeling of the growth cycle of spinach is equally as important as the temperature of the nutrient solution. The growth cycle of spinach is essentially controlled by the amount of light that the crop receives. In the winter months in Ithaca, NY, natural sunlight is not adequate to produce a harvestable baby spinach crop in two weeks, whereas, in summer months, it may take considerably less time. To balance the production over the course of the year, supplemental lighting and shading is used. Lighting and shading not only have an important influence on the growth of the crop, they also significantly affect the thermal environment of the greenhouse. Supplemental lights are not 100% efficient and produce considerable amounts of heat as well. Shading systems not only block a portion of the light, they also essentially create two airspaces

within the greenhouse that can have quite different temperatures. Shading also reduces radiative heat loss during the night and can help insulate the greenhouse. Because of the important role these two controls have on both the greenhouse environment, and crop production, they were included in the greenhouse model.

The greenhouse model provides the input needed by the subsequent spinach growth and *Pythium* disease models. The greenhouse model provides temperatures and light levels (and light source) to the spinach growth model, which allows the prediction of how a spinach crop will develop. Nutrient solution temperatures and the status of the spinach crop from the spinach growth model are the input to the *Pythium* disease model.

The basis of the *Pythium aphanidermatum* simulation is the temperature of the nutrient solution and so the need for a model that will provide not only these values, but the lighting environment that leads to them.

*Objectives:*

1. to develop a greenhouse climate simulation model capable of predicting nutrient solution pond temperatures under standard controlled environment agriculture operating principles,
2. to validate the operation of the model with both simulated and real weather data.

#### **4.2 LITERATURE REVIEW:**

Greenhouse simulation models are usually developed for a specific purpose including control and production/operation optimization. Models are also used to better understand the complex interactions that occur within greenhouses, and to evaluate new crops, or ways of managing them (Jones et al., 1995). With the increasing ease and power of computer simulations, greenhouse models have become

very popular with researchers. One of the most referenced is by Bot (1983). In this work, Bot quantifies the fundamental physical processes at work within a greenhouse, focusing on ventilation, radiation and convective exchanges. In Bot's model, thermal and moisture interactions with the plant canopy and underlying soil are included.

A primary focus in greenhouse models is the temperature of the air within. Whether it is estimating the effect of a night curtain (Chandra and Albright, 1980), or intermittent fogging (Abdel-Ghany and Kozai, 2005), modelers are usually interested in the aerial temperature of the greenhouse. One of the main reasons for using a greenhouse is to moderate the temperature, making the conditions more conducive to plant growth. However, few models include the temperature of the nutrient solution in which a floating hydroponic crop is grown. This omission is likely because relatively few greenhouse crops (other than leafy greens) are grown in this fashion. Also the nutrient solution temperature may often be ignored as a simplification because, as a general rule, the nutrient solution tends to follow longer term aerial temperature trends. Usually, only when the nutrient solution temperature is a primary concern of the model, is it included. Such a case is outlined in Zhu et al. (1998) where greenhouses were used to cover aquaculture ponds, within which the temperature of water has powerful effects not only on the growth and development of the fish, but also on the solubility (and availability) of dissolved oxygen.

Though Bot included interactions with the plant canopy, crop response to the greenhouse climate was not a goal of the work. In other models however, crop response is a fundamental component. In the HORTISIM model Gijzen et al. (1998), developed a model encompassing seven submodels including: weather, greenhouse climate, crop, soil, greenhouse manager, crop manager and soil manager. The HORTISIM model is used for predicting both the production of tomatoes and sweet peppers, and the cost of that production in terms of energy, CO<sub>2</sub> and water in quite



different climactic locations (data from the Netherlands and Israel was used to validate the model). Not all models need to be so complex, and often assumptions can be made to simplify both the form of the model, and the computational requirements.

### **4.3 METHODS AND MATERIALS:**

#### **4.3.1 Model Formulation:**

To model temperature and moisture relations in a greenhouse, one approach is to divide the greenhouse into several layers. Assuming that there is adequate mixing/thermal conductivity, temperatures within each layer are considered to be uniform. Assuming uniform temperatures and properties within layers is called a lumped parameter approach. This approach was used to model heat flow through the layers of a green roof (Lambert 2006). Temperature changes within layers are due to conduction, convection, radiation and enthalpy exchanges, depending on the medium of which the layer is made and the layer with which it is exchanging energy. To determine the change in temperature of the different layers over time, the heat lost from a particular layer is subtracted from the heat input to that same layer and the difference is the storage. Dividing by the heat capacitance of the layer medium gives the change in temperature.

To account for climate seasonal and diurnal changes it is necessary to model how the greenhouse layers change temperature over time. The assumption is that temperatures within each layer are constant and uniform over the course of a set time step. A time step of one hour was selected as this is less than the period of diurnal change and greater than the thermal response time of a greenhouse. Climatic data is usually available on an hourly basis, and more important, an hour is usually sufficiently long for adequate convection and conduction to take place to ensure temperatures are relatively uniform within layers. Another reason for an hourly time

step is many greenhouse operational decisions are made on an hourly basis (temperature setpoints, lighting). This type of model is called a stepwise steady state model. Stepwise describes the time steps used, and steady state for the assumption that temperatures are uniform and constant within layers for the duration of each time step.

Another major simplifying assumption is that energy flows within the greenhouse are one dimensional and that energy exchange through the sides of the greenhouse is negligible, compared to magnitudes of other flows.

With the exception of radiation, the other heat fluxes, conduction, convection and exchange of air of differing enthalpies, can all be described by linear equations. The Stephan-Boltzman Law governs the exchange of energy between two surfaces at temperatures  $T_1$  and  $T_2$ , through radiation:

$$radiative\ flux = \varepsilon\sigma A(T_1^4 - T_2^4) \quad 4.1$$

where:  $\varepsilon$  is the emissivity of the material  
 $\sigma$  is the Stephan-Boltzman constant  
 $A$  is the surface area

This relation is not linear, but it can be linearized if the assumption is made that the temperature differences are not large, and do not change drastically between time steps (equation 4.2).

$$radiative\ flux \cong \varepsilon\sigma A \left( \frac{T_{1,t-1} + T_{2,t-1}}{2} \right)^3 (T_1 - T_2) \quad 4.2$$

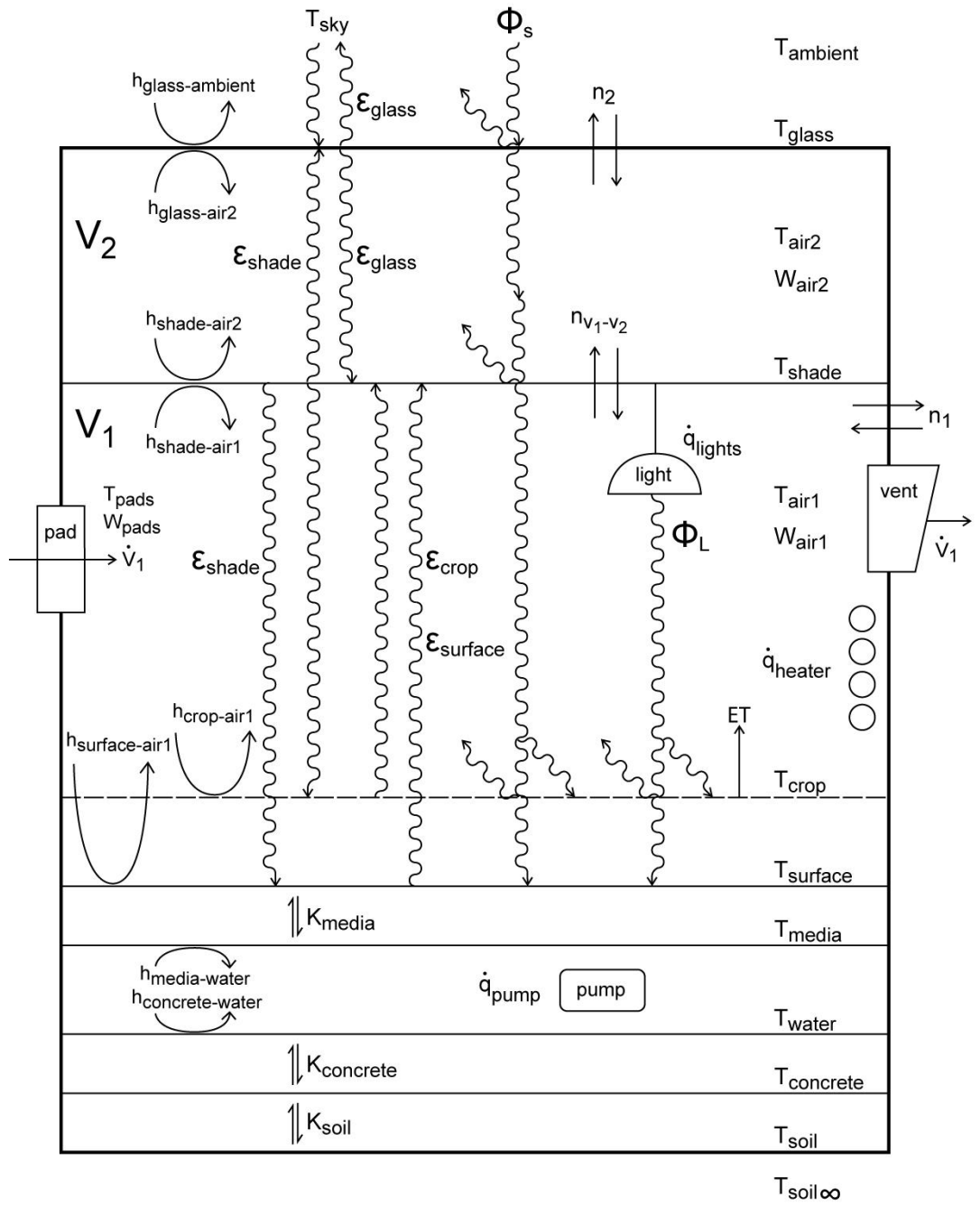
Using this simplification it is possible to develop a series of linear equations that describe the flow of energy between the layers which can then be solved simultaneously to give the temperature of each layer.

#### 4.3.2 Structure of the Model:

To account for the effects of the shade/thermal curtain, two similar versions of the model are used. When the shade is in place the air within the greenhouse is divided into two layers separated by the shade material. With the shade retracted, the previous model is simplified, as the greenhouse air is no longer segregated and the shade material is no longer a layer to be considered.

The layers of the model are illustrated in Figure 4.1. Also illustrated in Figure 4.1 are the energy and moisture exchange processes described in the model. Symbols are defined in Table 4.1.

In establishing the equations to describe the flow of heat and moisture between layers, the assumption is made that heat flows into a layer from the layer (or boundary) above it, and out of the layer into the layer (or boundary) beneath it. This assumption provides a consistent method of assigning signs to each term. Once all of the terms of the series of linear equations have been calculated (a and b terms are developed in the following text), they can be solved simultaneously for the temperatures by putting the terms into a 10 x 10 matrix (equation 4.3). Inverting the matrix and multiplying by the solution matrix ( $b_0, b_1, \dots, b_9$ ) yields the 10 temperatures.



**Figure 4.1** Diagram of thermal layers and energy flow processes within the greenhouse model (F. John Peters).

$$\begin{bmatrix}
 a_{00} & a_{01} & 0 & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\
 a_{10} & a_{11} & a_{12} & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\
 0 & a_{21} & a_{22} & a_{23} & 0 & 0 & 0 & 0 & 0 & 0 \\
 0 & 0 & a_{32} & a_{33} & a_{34} & 0 & 0 & 0 & 0 & 0 \\
 0 & 0 & 0 & a_{43} & a_{44} & a_{45} & a_{46} & a_{47} & 0 & 0 \\
 0 & 0 & 0 & 0 & a_{54} & a_{55} & a_{56} & a_{57} & 0 & 0 \\
 0 & 0 & 0 & 0 & a_{64} & a_{65} & a_{66} & a_{67} & a_{68} & 0 \\
 0 & 0 & 0 & 0 & a_{74} & a_{75} & a_{76} & a_{77} & a_{78} & a_{79} \\
 0 & 0 & 0 & 0 & 0 & 0 & a_{86} & a_{87} & a_{88} & a_{89} \\
 0 & 0 & 0 & 0 & 0 & 0 & 0 & a_{97} & a_{98} & a_{99}
 \end{bmatrix} \times$$

$$\begin{bmatrix}
 T_{\text{soil}} \\
 T_{\text{concrete}} \\
 T_{\text{water}} \\
 T_{\text{medium}} \\
 T_{\text{surface}} \\
 T_{\text{canopy}} \\
 T_{\text{air1}} \\
 T_{\text{shade}} \\
 T_{\text{air2}} \\
 T_{\text{glass}}
 \end{bmatrix} = \begin{bmatrix}
 b_0 \\
 b_1 \\
 b_2 \\
 b_3 \\
 b_4 \\
 b_5 \\
 b_6 \\
 b_7 \\
 b_8 \\
 b_9
 \end{bmatrix}$$

4.3

An additional component to the flow of energy necessary when modeling air layers is the enthalpy, which is a function of not only the temperature of the air, but the amount of moisture within the air. The enthalpy of air is defined in equation 4.4.

$$h = 1006t + W(2501000 + 1805t) \quad 4.4$$

- where:
- h is the enthalpy of the moist air (J kg<sup>-1</sup>)
  - 1006 is the specific heat of dry air (J kg<sup>-1</sup> K<sup>-1</sup>)
  - t is the temperature in Celsius (C)
  - W is the humidity ratio (kg water kg<sup>-1</sup> dry air)
  - 2501E3 is the Heat of Vaporization at 0 C(J kg<sup>-1</sup>)
  - 1805 is the specific heat of water vapor (J kg<sup>-1</sup> K<sup>-1</sup>)

When considering the case of bulk exchanges of air through venting and infiltration, it is assumed (for the purposes of developing the equations of the model) that energy flows into the layer from the source of the flow (whether that is ambient air or another air space) and that energy, in an identical mass of air, leaves the layer (the volume of air in the layer remains constant though the energy content of the air changes). It is necessary to mix the existing mass of air with the incoming mass to determine the values of its properties. Assuming one mass of air with enthalpy  $h_1$ , humidity ratio  $W_1$  and volume  $V_1$  mixes with a second mass of air with enthalpy  $h_2$ , humidity ratio  $W_2$  and volume  $V_2$ , the resultant air mass properties are defined in equations 4.5 and 4.6;

$$h_3 = \left( \frac{h_1 \frac{V_1}{V_{sa1}} + h_2 \frac{V_2}{V_{sa2}}}{\frac{V_1}{V_{sa1}} + \frac{V_2}{V_{sa2}}} \right) \quad 4.5$$

where:  $V_{sa1}$  is the air specific volume of air volume 1 ( $\text{m}^3 \text{kg}^{-1}$  dry air)

$V_{sa2}$  is the air specific volume of air volume 2 ( $\text{m}^3 \text{kg}^{-1}$  dry air)

$$W_3 = \left( \frac{W_1 \frac{V_1}{V_{sa1}} + W_2 \frac{V_2}{V_{sa2}}}{\frac{V_1}{V_{sa1}} + \frac{V_2}{V_{sa2}}} \right) \quad 4.6$$

Table 4.1 summarizes the variables and parameters used in the energy and moisture flow equations, and gives their symbols and assumed values. Parameter values were estimated or developed from data given in heat and mass transfer texts and ASHRAE (American Society of Heating, Refrigerating and Air-Conditioning Engineers) (2009) and ASABE (American Society of Agricultural and Biological Engineers) (2007) standards.

**Table 4.1** Symbols, definition and values of parameters and variables used in developing the energy flow equations of the lumped parameter stepwise steady state greenhouse simulation model.

$\alpha_{\text{crop}}$	Long Wave radiation Absorbance of the crop (unitless)	0.95
$\alpha_{\text{medium}}$	Long Wave radiation Absorbance of the medium (unitless)	0.9
$\alpha_{\text{shade}}$	Long Wave radiation Absorbance of the shade (unitless)	0.5
$\Delta$	Slope of the vapor pressure curve (kPa K <sup>-1</sup> )	variable
$\epsilon_{\text{crop}}$	Long Wave Emissivity of the crop (unitless)	0.95
$\epsilon_{\text{medium}}$	Long Wave Emissivity of the medium (unitless)	0.95
$\epsilon_{\text{shade}}$	Long Wave Emissivity of the shade (unitless)	0.5
$\epsilon_{\text{glass}}$	Long Wave Emissivity of the glass (unitless)	0.95
$\gamma$	Psychrometric constant (kPa K <sup>-1</sup> )	variable
$\rho_{\text{concrete}}$	Density of concrete (kg m <sup>-3</sup> )	2200
$\rho_{\text{crop}}$	Canopy reflectivity to solar and supplemental light (unitless)	0.25
$\rho_{\text{glass}}$	Glass reflectivity to solar radiation (unitless)	0.25
$\rho_{\text{medium}}$	Density of root medium (kg m <sup>-3</sup> )	100
$\rho_{\text{soil}}$	Density of soil (kg m <sup>-3</sup> )	1800
$\rho_{\text{water}}$	Density of nutrient solution (kg m <sup>-3</sup> )	1000
$\phi_{\text{l}}$	Radiation from supplemental lights, (J m <sup>-2</sup> hr <sup>-1</sup> )	variable
$\phi_{\text{solar,glass}}$	Solar radiation through the glass ( $\phi_{\text{solar,outside}}\tau_{\text{glass}}$ ), (J m <sup>-2</sup> hr <sup>-1</sup> )	variable
$\phi_{\text{solar,outside}}$	Solar radiation outside the greenhouse glass, (J m <sup>-2</sup> hr <sup>-1</sup> )	variable
$\phi_{\text{solar,shade}}$	Solar radiation through the shade ( $\phi_{\text{solar,glass}}\tau_{\text{shade}}$ ), (J m <sup>-2</sup> hr <sup>-1</sup> )	variable
$\tau_{\text{glass}}$	Transmittance of the greenhouse glass to solar radiation (unitless)	0.7
$\tau_{\text{shade}}$	Transmittance of the shade to solar radiation (unitless)	0.3
A	Area of greenhouse (m <sup>2</sup> )	10000



**Table 4.1** (Continued)

$c_{p_{soil}}$	Heat capacity of soil ( $J\ kg^{-1}\ K^{-1}$ )	1217
$c_{p_{concrete}}$	Heat capacity of concrete ( $J\ kg^{-1}\ K^{-1}$ )	882
$c_{p_{water}}$	Heat capacity of nutrient solution ( $J\ kg^{-1}\ K^{-1}$ )	4184
$c_{p_{medium}}$	Heat capacity of medium (including Styrofoam float) ( $J\ kg^{-1}\ K^{-1}$ )	840
$(e_a - e_d)$	Vapor pressure deficit (kPa)	variable
ET	Crop Evapotranspiration ( $J\ m^{-2}\ hr^{-1}$ )	variable
$ET_o$	Reference Crop Evapotranspiration ( $mm\ hr^{-1}$ )	variable
G	Soil heat flux, ( $MJ\ m^{-2}\ h^{-1}$ )	variable
$h_{crop-air}$	Convective heat transfer coefficient between crop and greenhouse air ( $J\ m^{-2}\ hr^{-1}\ K^{-1}$ )	29000
$h_{glass-air}$	Convective heat transfer coefficient between glass and greenhouse air ( $J\ m^{-2}\ hr^{-1}\ K^{-1}$ )	29000
$h_{glass-ambient}$	Convective heat transfer coefficient between glass and ambient air ( $J\ m^{-2}\ hr^{-1}\ K^{-1}$ )	35000
$h_{glass-air2}$	Convective heat transfer coefficient between glass and greenhouse airspace 2 ( $J\ m^{-2}\ hr^{-1}\ K^{-1}$ )	29000
$h_{medium-air}$	Convective heat transfer coefficient between medium surface and greenhouse air ( $J\ m^{-2}\ hr^{-1}\ K^{-1}$ )	29000
$h_{shade-air1}$	Convective heat transfer coefficient between shade and greenhouse airspace 1 ( $J\ m^{-2}\ hr^{-1}\ K^{-1}$ )	29000
$h_{shade-air2}$	Convective heat transfer coefficient between shade and greenhouse airspace 2 ( $J\ m^{-2}\ hr^{-1}\ K^{-1}$ )	29000
$h_{water}$	Convective heat transfer coefficient between concrete and nutrient solution, and the nutrient solution and medium ( $J\ m^{-2}\ hr^{-1}\ K^{-1}$ )	42000
$H_{fg}$	Heat of vaporization ( $kJ\ kg^{-1}$ )	variable

**Table 4.1** (Continued)

K	Canopy extinction coefficient (unitless)	0.74
LAI	Leaf Area Index ( $\text{m}^2$ leaf area $\text{m}^{-2}$ ground surface area for plant canopy)	0.7
$k_{\text{soil}}$	Thermal conductivity of soil ( $\text{J m}^{-1} \text{hr}^{-1} \text{K}^{-1}$ )	6480
$k_{\text{concrete}}$	Thermal conductivity of concrete ( $\text{J m}^{-1} \text{hr}^{-1} \text{K}^{-1}$ )	4608
$k_{\text{medium}}$	Thermal conductivity of medium ( $\text{J m}^{-1} \text{hr}^{-1} \text{K}^{-1}$ )	700
$K_c$	Crop coefficient to convert reference ET to spinach ET (unitless)	1.84
$n_1$	Air exchanged with the outside due to infiltration ( $\text{m}^3 \text{m}^{-2} \text{hr}^{-1}$ )	4
$n_{v1-v2}$	Volume of air exchanged between airspace 1 and 2 ( $\text{m}^3 \text{m}^{-2} \text{hr}^{-1}$ )	2
$\dot{q}_{\text{heater}}$	Heat input to the greenhouse air due to heating system ( $\text{J m}^{-2} \text{hr}^{-1}$ )	1.33E6
$\dot{q}_{\text{lamps}}$	Heat input to the greenhouse air due to supplemental lights ( $\text{J m}^{-2} \text{hr}^{-1}$ )	5.96E5
$\dot{q}_{\text{pump}}$	Heat input to the nutrient solution by the pump ( $\text{J m}^{-2} \text{hr}^{-1}$ )	6770
$R_n$	Net radiation at the crop surface ( $\text{MJ m}^{-2} \text{h}^{-1}$ )	variable
$T_{\text{air}}$	Temperature of the greenhouse air (K)	variable
$T_{\text{air}, t-1}$	Temperature of the greenhouse air in the previous hour (K)	variable
$T_{\text{air}1}$	Temperature of the greenhouse air in airspace 1 (K)	variable
$T_{\text{air}1, t-1}$	Temperature of the greenhouse air in airspace 1 in the previous hour (K)	variable
$T_{\text{air}2}$	Temperature of the greenhouse air in airspace 2 (K)	variable
$T_{\text{air}2, t-1}$	Temperature of the greenhouse air in airspace 2 in the previous hour (K)	variable

**Table 4.1** (Continued)

$T_{\text{ambient}}$	Temperature of the air outside the greenhouse (K)	variable
$T_{\text{concrete}}$	Temperature of the concrete layer (K)	variable
$T_{\text{concrete, t-1}}$	Temperature of the concrete layer in the previous hour (K)	variable
$T_{\text{crop}}$	Temperature of the crop (K)	variable
$T_{\text{crop, t-1}}$	Temperature of the crop in the previous hour (K)	variable
$T_{\text{glass}}$	Temperature of the greenhouse glass (K)	variable
$T_{\text{glass, t-1}}$	Temperature of the greenhouse glass in the previous hour (K)	variable
$T_{\text{medium}}$	Temperature of the medium layer (K)	variable
$T_{\text{medium, t-1}}$	Temperature of the medium layer in the previous hour (K)	variable
$T_{\text{shade}}$	Temperature of the shade curtain (K)	variable
$T_{\text{shade, t-1}}$	Temperature of the shade curtain in the previous hour (K)	variable
$T_{\text{sky}}$	Temperature of the sky (K)	variable
$T_{\text{soil}}$	Temperature of the soil layer (K)	variable
$T_{\text{soil,t-1}}$	Temperature of soil in the previous hour (K)	variable
$T_{\text{soil}\infty}$	Temperature of soil at depth where annual oscillations are $\pm 10\%$ (K)	variable
$T_{\text{surface}}$	Temperature of the medium surface (K)	variable
$T_{\text{surface, t-1}}$	Temperature of the medium surface in the previous hour (K)	variable
$T_{\text{water}}$	Temperature of the nutrient solution layer (K)	variable
$T_{\text{water, t-1}}$	Temperature of the nutrient solution in the previous hour (K)	variable
$U_2$	Average windspeed at 2 m above the plant canopy ( $\text{m s}^{-1}$ )	0.12
$\dot{v}_1$	Volume of air vented during the hour ( $\text{m}^3 \text{m}^{-2}$ )	variable

**Table 4.1** (Continued)

V	Volume of air in greenhouse ( $\text{m}^3 \text{ m}^{-2}$ )	4
V <sub>1</sub>	Volume of air in greenhouse airspace 1 ( $\text{m}^3 \text{ m}^{-2}$ )	3
V <sub>2</sub>	Volume of air in greenhouse airspace 2 ( $\text{m}^3 \text{ m}^{-2}$ )	1
V <sub>saair</sub>	Specific volume of greenhouse air ( $\text{m}^3 \text{ kg}^{-1}$ )	variable
V <sub>saair1</sub>	Specific volume of greenhouse air in airspace 1 ( $\text{m}^3 \text{ kg}^{-1}$ )	variable
V <sub>saair2</sub>	Specific volume of greenhouse air in airspace 2 ( $\text{m}^3 \text{ kg}^{-1}$ )	variable
V <sub>saamb</sub>	Specific volume of the outside air ( $\text{m}^3 \text{ kg}^{-1}$ )	variable
V <sub>savent</sub>	Specific volume of the vent air ( $\text{m}^3 \text{ kg}^{-1}$ )	variable
W <sub>air</sub>	Humidity ratio of the greenhouse air ( $\text{kg water kg}^{-1}$ dry air)	variable
W <sub>air1</sub>	Humidity ratio of the greenhouse air in airspace 1 ( $\text{kg water kg}^{-1}$ dry air)	variable
W <sub>air2</sub>	Humidity ratio of the greenhouse air in airspace 2 ( $\text{kg water kg}^{-1}$ dry air)	variable
W <sub>amb</sub>	Humidity ratio of the outside air ( $\text{kg water kg}^{-1}$ dry air)	variable
W <sub>vent</sub>	Humidity ratio of the vent air ( $\text{kg water kg}^{-1}$ dry air)	variable
Z <sub>air</sub>	Thickness of air level (medium to outer glass) (m)	4.0
Z <sub>air1</sub>	Thickness of air level below shade (m)	3.0
Z <sub>air2</sub>	Thickness of air level above shade (m)	1.0
Z <sub>concrete</sub>	Thickness of concrete layer (m)	0.15
Z <sub>medium</sub>	Thickness of medium layer (m)	0.05
Z <sub>soil</sub>	Thickness of soil layer (m)	6.1
Z <sub>water</sub>	Thickness (depth) of nutrient solution layer (m)	0.3

## Greenhouse Model with shading

### Soil Layer:

The first layer is the soil beneath the greenhouse structure. It is bounded by the deep soil, and the concrete that forms the floor of the greenhouse. The thickness of this layer was determined by assuming the thermal properties of the soil, and determining the depth at which the yearly temperature swings would dampen to 10% of the surface value. To estimate this depth we can use the relation from Carslaw and Jaeger (1959):

$$\text{amplitude of temperature oscillation} = e^{-x\sqrt{\frac{\omega}{2k}}} \quad 4.7$$

where: x is the depth (m)

$\omega$  is the frequency of oscillation ( $s^{-1}$ )  $3.17E-08$  (1 year)

k is the thermal diffusivity of the soil ( $m^2 s^{-1}$ )  $1.14E-07$

It is assumed that energy flows into and out of this layer through conduction. The flux of energy into the soil is described in equation 4.8, the flux out, in equation 4.9, and the storage in equation 4.10.

$$\text{flux in} = A \left( \frac{1}{\frac{z_{soil}}{k_{soil}} + \frac{z_{concrete}}{k_{concrete}}} \right) (T_{concrete} - T_{soil}) \quad 4.8$$

$$\text{flux out} = A \left( \frac{k_{soil}}{\frac{z_{soil}}{2}} \right) (T_{soil} - T_{soil\infty}) \quad 4.9$$

$$\text{storage} = A(\rho_{soil} cp_{soil} z_{soil})(T_{soil} - T_{soil_{t-1}}) \quad 4.10$$

Putting these equations together and solving for the coefficients of each temperature term results in equations 4.11 and 4.12.

$$a_{00} = A \left( \frac{k_{soil}}{z_{soil}} \right) (T_{concrete}) \quad 4.11$$

$$a_{01} = (-1)A \left( \frac{k_{soil}}{z_{soil}} + \frac{k_{soil}}{\frac{z_{soil}}{2}} + \rho_{soil} cp_{soil} z_{soil} \right) (T_{soil}) \quad 4.12$$

The constant is given in equation 4.13.

$$b_0 = A \left( \frac{k_{soil}}{\frac{z_{soil}}{2}} \right) T_{soil_{\infty}} + A \rho_{soil} cp_{soil} z_{soil} T_{soil_{t-1}} \quad 4.13$$

### Concrete Layer:

The second layer is that of the concrete foundation or base of the pond. This layer is bounded below by the soil beneath the greenhouse, and above by the pond nutrient solution.

Energy is exchanged with the ground layer through conduction, whereas energy is exchanged with the pond layer through convection. The equation describing the flux of energy in is described in equation 4.14, the flux of energy out in equation 4.15, and the storage of energy in equation 4.16.

$$flux\ in = Ah_{water} (T_{water} - T_{concrete}) \quad 4.14$$

$$flux\ out = A \left( \frac{k_{soil}}{z_{soil}} + \frac{k_{concrete}}{z_{concrete}} \right) (T_{concrete} - T_{soil}) \quad 4.15$$

$$storage = A(\rho_{concrete} cp_{concrete} z_{concrete}) (T_{concrete} - T_{concrete_{t-1}}) \quad 4.16$$

Putting these equations together and solving for the coefficients of each temperature term results in equations 4.17, 4.18 and 4.19.

$$a_{10} = Ah_{water} (T_{water}) \quad 4.17$$

$$a_{11} = (-1)A \left( h_{water} + \frac{k_{soil}}{z_{soil}} + \frac{k_{concrete}}{z_{concrete}} + \rho_{concrete} cp_{concrete} z_{concrete} \right) (T_{concrete}) \quad 4.18$$

$$a_{12} = A \left( \frac{k_{soil}}{z_{soil}} + \frac{k_{concrete}}{z_{concrete}} \right) (T_{soil}) \quad 4.19$$

The constant is given in equation 4.20.

$$b_1 = A(\rho_{concrete} cp_{concrete} z_{concrete}) (T_{concrete_{t-1}}) \quad 4.20$$

### **Nutrient Solution Layer:**

The third layer of the model is the nutrient solution. It is bounded by the concrete layer beneath it, and the floating medium above it. Energy flows into the layer from the medium layer through convection, and out to the concrete layer also through convection. The assumption is also made that the pond is covered 100% of the time so that evaporation and radiation exchanges can be neglected. Heat is added to the nutrient solution through the operation of the circulation pump. The equation describing the flux of energy in is described in equation 4.21, the flux of energy out in equation 4.22, and the storage of energy in equation 4.23.

$$flux\ in = A(h_{water} (T_{media} - T_{water}) + \dot{q}_{pump}) \quad 4.21$$

$$flux\ out = Ah_{water} (T_{water} - T_{concrete}) \quad 4.22$$

$$storage = A(\rho_{water} cp_{water} z_{water}) (T_{water} - T_{water_{t-1}}) \quad 4.23$$

Putting these equations together and solving for the coefficients of each temperature term results in equations 4.24, 4.25 and 4.26.

$$a_{21} = Ah_{water} (T_{media}) \quad 4.24$$

$$a_{22} = (-1)A(h_{water} + h_{water} + \rho_{water} cp_{water} z_{water})(T_{water}) \quad 4.25$$

$$a_{23} = Ah_{water}(T_{concrete}) \quad 4.26$$

The constant is given in equation 4.27.

$$b_2 = A((\rho_{water} cp_{water} z_{water})(T_{water_{t-1}}) + \dot{q}_{pump}) \quad 4.27$$

### Medium Layer:

The fourth layer of the model is the growth medium, which also includes the styrofoam floats in which it is placed. It is bounded by the nutrient solution layer beneath it, and the surface of the growth medium above it. Following the assumptions of the lumped parameter approach, the surface temperature of the medium equals the medium temperature. However, the medium surface layer was inserted into the model to facilitate the future inclusion of a reflective surface covering to reduce solar heat gain of the medium. Setting the properties of the surface the same as the medium, results in the same effects as if the surface layer was not included. Energy flows into the layer from the medium surface through conduction, and out to the nutrient solution layer through convection. The equation describing the flux of energy in is described in equation 4.28, the flux of energy out in equation 4.29, and the storage of energy in equation 4.30.

$$flux\ in = A\left(\frac{k_{media}}{\frac{z_{media}}{2}}\right)(T_{surface} - T_{media}) \quad 4.28$$

$$flux\ out = Ah_{water}(T_{media} - T_{water}) \quad 4.29$$

$$storage = A(\rho_{media} cp_{media} z_{media})(T_{media} - T_{media_{t-1}}) \quad 4.30$$



Putting these equations together and solving for the coefficients of each temperature term results in equations 4.31, 4.32 and 4.33.

$$a_{32} = A \left( \frac{k_{media}}{\frac{z_{media}}{2}} \right) (T_{surface}) \quad 4.31$$

$$a_{33} = (-1)A \left( \frac{k_{media}}{\frac{z_{media}}{2}} + h_{water} + \rho_{media} c_{p_{media}} z_{media} \right) (T_{media}) \quad 4.32$$

$$a_{34} = Ah_{water} (T_{water}) \quad 4.33$$

The constant is given in equation 4.34.

$$b_3 = A(\rho_{media} c_{p_{media}} z_{media}) (T_{media_{t-1}}) \quad 4.34$$

### **Medium Surface Layer:**

The fifth layer of the model is the medium surface. It is bounded by the medium layer beneath it, and the air of the greenhouse above it. The surface also exchanges energy through radiation with the crop canopy and greenhouse shade (or glass, depending on whether the shade is open or closed). Energy flows into the layer from the medium surface through convective heat transfer from the air, and through radiative heat transfer from the crop canopy and greenhouse shade. Energy flows out to the medium layer through conduction. Since it is a surface there is no thickness associated with it (and so no storage of heat). The equation describing the flux of energy in is described in equation 4.35, the flux of energy out in equation 4.36, and the storage of energy in equation 4.37.

$$\begin{aligned}
flux\ in = A & \left( h_{media-air} (T_{air1} - T_{surface}) \right. & 4.35 \\
& + (1 - \rho_{crop}) (\theta_{solar,shade} + \theta_l) e^{-kLAI} \alpha_{media} \\
& + 4LAI \varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{crop}}{2} \right)^3 (T_{crop} \\
& - T_{surface}) \\
& + 4(1 \\
& - LAI) \varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{shade}}{2} \right)^3 (T_{shade} \\
& \left. - T_{surface}) \right)
\end{aligned}$$

$$flux\ out = A \left( \frac{k_{media}}{\frac{z_{media}}{2}} \right) (T_{surface} - T_{media}) \quad 4.36$$

$$storage = 0 \quad 4.37$$

Putting these equations together and solving for the coefficients of each temperature term results in equations 4.38, 4.39, 4.40, 4.41 and 4.42.

$$a_{43} = Ah_{media-air} (T_{air1}) \quad 4.38$$

$$\begin{aligned}
a_{44} = (-1)A & \left( h_{media-air} + 4LAI \varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{crop}}{2} \right)^3 \right. & 4.39 \\
& + 4(1 - LAI) \varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{shade}}{2} \right)^3 \\
& \left. + \frac{k_{media}}{\frac{z_{media}}{2}} \right) (T_{surface})
\end{aligned}$$

$$a_{45} = 4ALAI\varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{crop}}{2} \right)^3 (T_{crop}) \quad 4.40$$

$$a_{46} = 4A(1 - LAI)\varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{shade}}{2} \right)^3 (T_{shade}) \quad 4.41$$

$$a_{47} = A \left( \frac{k_{media}}{\frac{z_{media}}{2}} \right) (T_{media}) \quad 4.42$$

The constant is given in equation 4.43.

$$b_4 = -A(1 - \rho_{crop})(\theta_{solar,shade} + \theta_l)e^{-kLAI} \alpha_{media} \quad 4.43$$

### **Crop Canopy Layer:**

The sixth layer of the model is the crop canopy. To simplify the interactions with the crop canopy the assumption was made that the canopy is represented by the Big Leaf model which assumes a single large leaf described by the Leaf Area Index, rather than many smaller individual leaves. Energy flows into the layer from the greenhouse air through convective heat transfer, through the absorption of sunlight and supplemental lighting, and through radiative heat transfer from the greenhouse shade. Some energy is gained by reflection of radiation from the medium surface, however, this portion is assumed to be small and is omitted for simplicity. Energy flows out through radiative exchange with the surface of the medium, and through evapotranspiration. Since it is considered a surface there is no thickness associated with it. The equation describing the flux of energy in is described in equation 4.44, the flux of energy out in equation 4.45, and the storage of energy in equation 4.48.

$$flux\ in = A \left( h_{crop-air} (T_{air\ 1} - T_{crop}) \right. \quad 4.44$$

$$+ (1 - \rho_{crop}) (\theta_{solar,shade} + \theta_l) (1 - e^{-kLAI}) \alpha_{crop}$$

$$\left. + 4LAI \varepsilon_{crop} \sigma \left( \frac{T_{shade} + T_{crop}}{2} \right)^3 (T_{shade} - T_{crop}) \right)$$

$$flux\ out = A \left( 4LAI \varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{crop}}{2} \right)^3 (T_{crop} - T_{surface}) \right. \quad 4.45$$

$$\left. + ET \right)$$

Evapotranspiration (ET) is determined by using the Penman-Monteith method (Pereira et al., 1996) to calculate the reference evapotranspiration. For simplification it was assumed that soil heat flux (G) was equal to zero.

$$ET_o = \quad 4.46$$

$$H_{fg} \rho_{water} K_c \left( \frac{0.408 \Delta (R_n - G) + \gamma \left( \frac{900}{T_{air,t-1} + 273.15} \right) U_2 (e_a - e_d)}{\Delta + \gamma (1 + 0.34 U_2)} \right)$$

To convert the reference crop evapotranspiration to an evapotranspiration similar to spinach, it is necessary to multiply by a crop coefficient,  $K_c$ . It was assumed that spinach transpires in a similar fashion to lettuce, for which Ciolkosz and Albright (2000) determined a value of 1.84 for  $K_c$ . It is also necessary to convert the depth of water into an energy equivalent which is done by multiplying by the heat of vaporization of water at the canopy temperature ( $H_{fg}$ ).

$$ET = H_{fg} \rho_{water} K_c ET_o \quad 4.47$$

$$storage = 0 \quad 4.48$$

Putting these equations together and solving for the coefficients of each temperature term results in equations 4.49, 4.50, 4.51, and 4.52.

$$a_{54} = Ah_{crop-air} (T_{air1}) \quad 4.49$$

$$a_{55} \quad 4.50$$

$$= (-1)A \left( h_{crop-air} + 4LAI\varepsilon_{crop} \sigma \left( \frac{T_{shade} + T_{crop}}{2} \right)^3 + 4LAI\varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{crop}}{2} \right)^3 \right) (T_{crop})$$

$$a_{56} = 4ALAI\varepsilon_{crop} \sigma \left( \frac{T_{shade} + T_{crop}}{2} \right)^3 (T_{shade}) \quad 4.51$$

$$a_{57} = 4ALAI\varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{crop}}{2} \right)^3 (T_{surface}) \quad 4.52$$

The constant is given in equation 4.53.

$$b_5 = (-1)A \left( (1 - \rho_{crop}) (\theta_{solar,shade} + \theta_l) (1 - e^{-kLAI}) \alpha_{crop} + ET \right) \quad 4.53$$

### **Greenhouse Air Layer (below the shade):**

The seventh layer of the model is the air in the greenhouse below the shade canopy. Energy flows into the layer from the bulk flow of air from above the shade, outside the greenhouse through infiltration, and through the air vents, which, depending on whether the cooling pads are operating, may be either outdoor air, or cooled air. This greenhouse air layer also receives energy from convective heat transfer from the shade curtain, as well as from the greenhouse heater (if functioning), and from the supplemental lights, if they are on. Evapotranspiration is also considered

an energy input to this layer. Energy flows out through convective heat transfer with the crop canopy and the medium surface, as well as through the exchange of air from above the shade curtain, and to the outside through infiltration and venting.

The equation describing the flux of energy in is described in equation 4.54, the flux of energy out in equation 4.55, and the storage of energy in equation 4.56.

$$\begin{aligned}
 \text{flux in} & \qquad \qquad \qquad 4.54 \\
 & = A \left( \dot{q}_{heater} + \dot{q}_{lamps} \right. \\
 & \quad + ET + h_{shade-air1} (T_{shade} - T_{air1}) \\
 & \quad + \left( \frac{\dot{v}_1}{V_{sa\ vent}} \right) (1006(T_{vent} - 273.15) \\
 & \quad + W_{vent} (2501000 \\
 & \quad + 1805(T_{vent} - 273.15))) \\
 & \quad + \left( \frac{n_{V1-V2}}{V_{sa\ air\ 2}} \right) (1006(T_{air\ 2} - 273.15) \\
 & \quad + W_{air\ 2} (2501000 \\
 & \quad + 1805(T_{air\ 2} - 273.15))) \\
 & \quad + \left( \frac{n_1}{V_{sa\ amb}} \right) (1006(T_{amb} - 273.15) \\
 & \quad + W_{amb} (2501000 \\
 & \quad + 1805(T_{amb} - 273.15)))
 \end{aligned}$$

$$flux\ out = A \left( h_{crop-air} (T_{air1} - T_{crop}) \right. \quad 4.55$$

$$\left. + h_{media-air} (T_{air1} - T_{surface}) \right)$$

$$+ \left( \frac{\dot{v}_1}{V_{sa\ air1}} \right) (1006(T_{air1} - 273.15)$$

$$+ W_{air1} (2501000 + 1805(T_{air1} - 273.15)))$$

$$+ \left( \frac{n_{V1-V2}}{V_{sa\ air1}} \right) (1006(T_{air1} - 273.15)$$

$$+ W_{air1} (2501000 + 1805(T_{air1} - 273.15)))$$

$$+ \left( \frac{n_1}{V_{sa\ air1}} \right) (1006(T_{air1} - 273.15)$$

$$+ W_{air1} (2501000 + 1805(T_{air1} - 273.15)))$$

$$storage = \left( \frac{V_1}{V_{sa\ air1}} \right) (1006(T_{air1} - 273.15) \quad 4.56$$

$$+ W_{air1} (2501000 + 1805(-273.15)))$$

$$- \left( \frac{V_1}{V_{sa\ air1\ t-1}} \right) (1006(T_{air1\ t-1} - 273.15)$$

$$+ W_{air1\ t-1} (2501000$$

$$+ 1805(T_{air1\ t-1} - 273.15)))$$

Putting these equations together and solving for the coefficients of each temperature term results in equations 4.57, 4.58, 4.59, 4.60 and 4.61.

$$a_{64} = Ah_{media-air}(T_{surface}) \quad 4.57$$

$$a_{65} = Ah_{crop-air}(T_{crop}) \quad 4.58$$

$$a_{66} \quad 4.59$$

$$= (-1) \left( A(h_{shade-air1} + h_{crop-air} + h_{media-air}) + \frac{(1006 + W_{air1} * 1805)(n_1 + V_1 + v_1 + n_{V1-V2})}{V_{sa1t-1}} \right) (T_{air1})$$

$$a_{67} = Ah_{shade-air1}(T_{shade}) \quad 4.60$$

$$a_{68} = \left( \frac{(1006 + W_{air2} * 1805)(n_{V1-V2})}{V_{sa2t-1}} \right) (T_{air2}) \quad 4.61$$

The constant is given in equation 4.62.



$$\begin{aligned}
b_6 = (-1) & \left( A(\dot{q}_{heater} + \dot{q}_{lamps} + ET) \right. \\
& + \left( \frac{\dot{v}_1}{V_{sa_{vent}}} \right) (1006(T_{vent} - 273.15) \\
& + W_{vent} (2501000 + 1805(T_{vent} - 273.15))) \\
& + \left( \frac{n_1}{V_{sa_{amb}}} \right) (1006(T_{amb} - 273.15) \\
& + W_{amb} (2501000 + 1805(T_{amb} - 273.15))) \\
& - \left( \frac{n_1 + V_1 + \dot{v}_1 + n_{V1-V2}}{V_{sa_{air\ 1t-1}}} \right) (1006(-273.15) \\
& + W_{air\ 1} (2501000 + 1805(-273.15))) \\
& + \left( \frac{n_{V1-V2}}{V_{sa_{air\ 2t-1}}} \right) (1006(-273.15) \\
& + W_{air\ 2} (2501000 + 1805(-273.15))) \\
& + \left( \frac{V_1}{V_{sa_{air\ 1t-1}}} \right) (1006(T_{air\ 1t-1} - 273.15) \\
& \left. + W_{air\ 1t-1} (2501000 + 1805(T_{air\ 1t-1} - 273.15))) \right)
\end{aligned}$$

### Shade Layer:

The eighth layer of the model is the shade canopy. For the purposes of the model it is assumed to have no thickness (no heat storage). Energy flows into the shade through convection exchange with the greenhouse air layer above the shade (and below the glass), through radiative exchange with the greenhouse glass, and from the portion of sunlight passing through the glass that the shade intercepts. Energy flows out from the shade through convective exchange with the air layer beneath the

shade, and through radiative exchange with both the crop canopy and medium surface. The equation describing the flux of energy in is described in equation 4.63, the flux of energy out in equation 4.64, and the storage of energy in equation 4.65.

$$flux\ in = A \left( h_{shade-air\ 2} (T_{air\ 2} - T_{shade}) \right) \quad 4.63$$

$$+ 4\epsilon_{glass} \sigma \left( \frac{T_{glass} + T_{shade}}{2} \right)^3 (T_{glass} - T_{shade})$$

$$+ (1 - \rho_{glass}) (\theta_{solar, glass}) \alpha_{shade} \right)$$

$$flux\ out = A \left( h_{shade-air\ 1} (T_{shade} - T_{air\ 1}) \right) \quad 4.64$$

$$+ 4(1 - LAI) \epsilon_{media} \sigma \left( \frac{T_{surface} + T_{shade}}{2} \right)^3 (T_{shade} - T_{surface})$$

$$+ 4(LAI) \epsilon_{crop} \sigma \left( \frac{T_{shade} + T_{crop}}{2} \right)^3 (T_{shade} - T_{crop}) \right)$$

$$storage = 0 \quad 4.65$$

Putting these equations together and solving for the coefficients of each temperature term results in equations 4.66, 4.67, 4.68, 4.69, 4.70 and 4.71.

$$a_{74} = 4A(1 - LAI) \epsilon_{media} \sigma \left( \frac{T_{surface} + T_{shade}}{2} \right)^3 (T_{surface}) \quad 4.66$$

$$a_{75} = 4A(LAI) \epsilon_{crop} \sigma \left( \frac{T_{shade} + T_{crop}}{2} \right)^3 (T_{crop}) \quad 4.67$$

$$a_{76} = Ah_{shade-air1}(T_{air1}) \quad 4.68$$

$$a_{77} = (-1)A \left( h_{shade-air2} + 4\varepsilon_{glass} \sigma \left( \frac{T_{glass} + T_{shade}}{2} \right)^3 \right. \\ \left. + h_{shade-air1} \right. \\ \left. + 4(1 - LAI)\varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{shade}}{2} \right)^3 \right. \\ \left. + 4(LAI)\varepsilon_{crop} \sigma \left( \frac{T_{shade} + T_{crop}}{2} \right)^3 \right) (T_{shade}) \quad 4.69$$

$$a_{78} = Ah_{shade-air2}(T_{air2}) \quad 4.70$$

$$a_{79} = 4A\varepsilon_{glass} \sigma \left( \frac{T_{glass} + T_{shade}}{2} \right)^3 (T_{glass}) \quad 4.71$$

The constant is given in equation 4.72.

$$b_7 = (-1)A(1 - \rho_{glass})(\theta_{solar,glass})\alpha_{shade} \quad 4.72$$

### **Greenhouse Air Layer (above shade):**

The ninth layer of the model is the air above the shade curtain. Energy flows into the layer through convective exchange with the greenhouse glass. Energy also flows into this air layer through the bulk exchange of air with the outside through infiltration, and with the layer below the shade. Energy flows out of the layer through convective exchange with the shade curtain and through the exchange of air with outside air and the air layer below the shade through infiltration. The equation

describing the flux of energy in is described in equation 4.73, the flux of energy out in equation 4.74, and the storage of energy in equation 4.75.

$$flux\ in = Ah_{glass-air\ 2}(T_{glass} - T_{air\ 2}) \quad 4.73$$

$$+ \left( \frac{n_2}{V_{sa\ amb}} \right) (1006(T_{amb} - 273.15) \\ + W_{amb} (2501000 + 1805(T_{amb} - 273.15))) \\ + \left( \frac{n_{V1-V2}}{V_{sa\ air\ 1}} \right) (1006(T_{air\ 1} - 273.15) \\ + W_{air\ 1} (2501000 + 1805(T_{air\ 1} - 273.15)))$$

$$flux\ out \quad 4.74$$

$$= Ah_{shade-air\ 2}(T_{air\ 2} - T_{shade}) \\ + \left( \frac{n_2}{V_{sa\ air\ 2}} \right) (1006(T_{air\ 2} - 273.15) \\ + W_{air\ 2} (2501000 \\ + 1805(T_{air\ 2} - 273.15))) \\ + \left( \frac{n_{V1-V2}}{V_{sa\ air\ 2}} \right) (1006(T_{air\ 2} - 273.15) \\ + W_{air\ 2} (2501000 \\ + 1805(T_{air\ 2} - 273.15)))$$

$$\begin{aligned}
storage = & \left( \frac{V_2}{V_{sa\ air\ 2}} \right) (1006(T_{air\ 2} - 273.15) & 4.75 \\
& + W_{air\ 2}(2501000 + 1805(-273.15))) \\
& - \left( \frac{V_2}{V_{sa\ air\ 2_{t-1}}} \right) (1006(T_{air\ 2_{t-1}} - 273.15) \\
& + W_{air\ 2_{t-1}}(2501000 \\
& + 1805(T_{air\ 2_{t-1}} - 273.15)))
\end{aligned}$$

Putting these equations together and solving for the coefficients of each temperature term results in equations 4.76, 4.77, 4.78, and 4.79.

$$a_{86} = \left( \frac{(1006 + W_{air\ 1} * 1805)(n_{V1-V2})}{V_{sa1t-1}} \right) (T_{air\ 1}) \quad 4.76$$

$$a_{87} = Ah_{shade-air\ 2}(T_{shade}) \quad 4.77$$

$$\begin{aligned}
a_{88} & \quad 4.78 \\
= & (-1) \left( A(h_{glass-air\ 2} + h_{shade-air\ 2}) \right. \\
& \left. + \frac{(1006 + W_{air\ 2} * 1805)(n_2 + V_2 + n_{V1-V2})}{V_{sa2t-1}} \right) (T_{air\ 2})
\end{aligned}$$

$$a_{89} = Ah_{glass-air\ 2}(T_{glass}) \quad 4.79$$

The constant is given in equation 4.80.

$$\begin{aligned}
 b_8 = (-1) & \left( \left( \frac{n_2}{V_{sa\ amb}} \right) (1006(T_{amb} - 273.15)) \right. & 4.80 \\
 & + W_{amb} (2501000 + 1805(T_{amb} - 273.15)) \\
 & + \left( \frac{V_2}{V_{sa\ air\ 2_{t-1}}} \right) (1006(T_{air\ 2_{t-1}} - 273.15)) \\
 & + W_{air\ 2_{t-1}} (2501000 \\
 & + 1805(T_{air\ 2_{t-1}} - 273.15)) \\
 & + \left( \frac{n_{V1-V2}}{V_{sa\ air\ 1_{t-1}}} \right) (1006(-273.15)) \\
 & + W_{air\ 1} (2501000 + 1805(-273.15)) \\
 & - \left( \frac{n_2 + V_2 + n_{V1-V2}}{V_{sa\ air\ 2_{t-1}}} \right) (1006(-273.15)) \\
 & \left. + W_{air\ 2} (2501000 + 1805(-273.15)) \right)
 \end{aligned}$$

### **Greenhouse Glass Layer:**

The tenth and final layer of the model is the greenhouse glass. For the purposes of the model it is assumed to have no thickness, store no heat and have negligible thermal resistance. Energy flow into the glass is from convective exchange with the outside (ambient) air, from the portion of the sunlight the glass intercepts, and from radiative exchange with the sky. As the temperature of the sky is not provided in

the data file which this simulation uses, an estimate for the temperature of the sky was used, based on the relation (Swinbank 1963):

$$T_{sky} = 0.0552T_{amb}^{1.5} \quad 4.81$$

This relation is valid for a clear sky and does not really work for a cloudy sky situation which is significantly more complex. However, the clear sky approach was used as a quick approximation sufficient for the purpose of the overall greenhouse simulation model.

Energy flows out from the layer through convection exchange with the air layer below, and through radiative exchange with the shade curtain. The equation describing the flux of energy in, is described in equation 4.82, the flux of energy out in equation 4.83, and the storage of energy in equation 4.84.

$$\begin{aligned} flux\ in = A \left( h_{glass-ambient} (T_{ambient} - T_{glass}) \right. & \quad 4.82 \\ & + (1 - \rho_{glass})(\theta_{solar, outside})\alpha_{glass} \\ & \left. + 4\varepsilon_{glass} \sigma \left( \frac{T_{glass} + T_{sky}}{2} \right)^3 (T_{sky} - T_{glass}) \right) \end{aligned}$$

$$\begin{aligned} flux\ out = A \left( h_{glass-air2} (T_{glass} - T_{air2}) \right. & \quad 4.83 \\ & + 4\varepsilon_{glass} \sigma \left( \frac{T_{glass} + T_{shade}}{2} \right)^3 (T_{glass} \\ & \left. - T_{shade}) \right) \end{aligned}$$

$$storage = 0 \quad 4.84$$

Putting these equations together and solving for the coefficients of each temperature term results in equations 4.85, 4.86, and 4.87.

$$a_{97} = 4A\varepsilon_{glass} \sigma \left( \frac{T_{glass} + T_{shade}}{2} \right)^3 (T_{shade}) \quad 4.85$$

$$a_{98} = (-1)A \left( h_{glass-ambient} + 4\varepsilon_{glass} \sigma \left( \frac{T_{glass} + T_{sky}}{2} \right)^3 \right. \\ \left. + h_{glass-air2} \right. \\ \left. + 4\varepsilon_{glass} \sigma \left( \frac{T_{glass} + T_{shade}}{2} \right)^3 \right) (T_{glass}) \quad 4.86$$

$$a_{99} = Ah_{glass-air2} (T_{air2}) \quad 4.87$$

The constant is given in equation 4.88.

$$b_9 = (-1)A \left( h_{glass-ambient} (T_{ambient}) \right. \\ \left. + (1 - \rho_g)(\theta_{solar, outside}) \alpha_{glass} \right. \\ \left. + 4\varepsilon_{glass} \sigma \left( \frac{T_{glass} + T_{sky}}{2} \right)^3 (T_{sky}) \right) \quad 4.88$$



### Greenhouse Model Without Shade Curtain

The model implements light and shade control, and decisions about the status of the lights and shade are made every hour. Depending on whether the shade is opened or closed the number of layers of the model changes. If the shade is open, the shade layer drops out, and the two greenhouse airspaces are combined. If this is the case the model temperatures can be solved by putting the calculated coefficients into an 8 x 8 matrix (equation 4.89), inverting it, and then multiplying by the solution matrix (b<sub>0</sub>, b<sub>1</sub>, ..., b<sub>7</sub>) to get the temperatures of the 8 layers.

$$\begin{bmatrix} a_{00} & a_{01} & 0 & 0 & 0 & 0 & 0 & 0 \\ a_{10} & a_{11} & a_{12} & 0 & 0 & 0 & 0 & 0 \\ 0 & a_{21} & a_{22} & a_{23} & 0 & 0 & 0 & 0 \\ 0 & 0 & a_{32} & a_{33} & a_{34} & 0 & 0 & 0 \\ 0 & 0 & 0 & a_{43} & a_{44} & a_{45} & a_{46} & a_{47} \\ 0 & 0 & 0 & 0 & a_{54} & a_{55} & a_{56} & a_{57} \\ 0 & 0 & 0 & 0 & a_{64} & a_{65} & a_{66} & a_{67} \\ 0 & 0 & 0 & 0 & a_{74} & a_{75} & a_{76} & a_{77} \end{bmatrix} \times \begin{bmatrix} T_{\text{soil}} \\ T_{\text{concrete}} \\ T_{\text{water}} \\ T_{\text{medium}} \\ T_{\text{surface}} \\ T_{\text{canopy}} \\ T_{\text{air}} \\ T_{\text{glass}} \end{bmatrix} = \begin{bmatrix} b_0 \\ b_1 \\ b_2 \\ b_3 \\ b_4 \\ b_5 \\ b_6 \\ b_7 \end{bmatrix} \quad 4.89$$

The equations governing the soil to medium layers remain unchanged; however, the remaining layers have new equations as follow.

#### Medium Surface Layer:

The equation for the medium surface is nearly identical. The only difference is that rather than radiative exchange with the shade curtain, the radiative exchange is now directly with the greenhouse glass. All other energy exchanges remain the same. The equation describing the flux of energy in is described in equation 4.90, the flux of energy out in equation 4.91, and the storage of energy in equation 4.92.

$$\begin{aligned}
flux\ in = A & \left( h_{media-air} (T_{air} - T_{surface}) \right. & 4.90 \\
& + (1 - \rho_{crop})(\theta_{solar,shade} + \theta_l)e^{-kLAI} \alpha_{media} \\
& + 4LAI \varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{crop}}{2} \right)^3 (T_{crop} \\
& - T_{surface}) \\
& + 4(1 \\
& - LAI) \varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{glass}}{2} \right)^3 (T_{glass} \\
& \left. - T_{surface}) \right)
\end{aligned}$$

$$flux\ out = A \left( \frac{k_{media}}{\frac{z_{media}}{2}} \right) (T_{surface} - T_{media}) \quad 4.91$$

$$storage = 0 \quad 4.92$$

Putting these equations together and solving for the coefficients of each temperature term results in equations 4.93, 4.94, 4.95, 4.96 and 4.97.

$$a_{43} = Ah_{media-air} (T_{air}) \quad 4.93$$

$$\begin{aligned}
a_{44} = (-1)A & \left( h_{media-air} + 4LAI \varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{crop}}{2} \right)^3 \right. & 4.94 \\
& + 4(1 - LAI) \varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{glass}}{2} \right)^3 \\
& \left. + \frac{k_{media}}{\frac{z_{media}}{2}} \right) (T_{surface})
\end{aligned}$$

$$a_{45} = 4ALAI\epsilon_{media} \sigma \left( \frac{T_{surface} + T_{crop}}{2} \right)^3 (T_{crop}) \quad 4.95$$

$$a_{45} = 4A(1 - LAI)\epsilon_{media} \sigma \left( \frac{T_{surface} + T_{glass}}{2} \right)^3 (T_{glass}) \quad 4.96$$

$$a_{46} = A \left( \frac{k_{media}}{\frac{z_{media}}{2}} \right) (T_{media}) \quad 4.97$$

The constant is given in equation 4.98.

$$b_4 = -A(1 - \rho_{crop})(\theta_{solar,shade} + \theta_l)e^{-kLAI} \alpha_{media} \quad 4.98$$

### Crop Canopy Layer:

The unshaded crop canopy is identical to the shaded canopy, the only difference being that radiative exchange is with the greenhouse glass, and not the shade curtain. The equation describing the flux of energy in is described in equation 4.99, the flux of energy out in equation 4.100, and the storage of energy in equation 4.101.

$$\begin{aligned} flux\ in = A \left( h_{crop-air} (T_{air} - T_{crop}) \right. & \quad 4.99 \\ & + (1 - \rho_c)(\theta_s + \theta_l)(1 - e^{-kLAI})\alpha_{crop} \\ & + 4LAI\epsilon_{crop} \sigma \left( \frac{T_{glass} + T_{crop}}{2} \right)^3 (T_{glass} \\ & \left. - T_{crop}) \right) \end{aligned}$$

$$flux\ out = A \left( 4LAI\varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{crop}}{2} \right)^3 (T_{crop} - T_{surface}) + ET \right) \quad 4.100$$

$$storage = 0 \quad 4.101$$

Putting these equations together and solving for the coefficients of each temperature term results in equations 4.102, 4.103, 4.104, and 4.105.

$$a_{54} = Ah_{crop-air} (T_{air}) \quad 4.102$$

$$a_{55} = (-1)A \left( h_{crop-air} + 4LAI\varepsilon_{crop} \sigma \left( \frac{T_{air} + T_{crop}}{2} \right)^3 + 4LAI\varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{crop}}{2} \right)^3 \right) (T_{crop}) \quad 4.103$$

$$a_{56} = 4ALAI\varepsilon_{crop} \sigma \left( \frac{T_{glass} + T_{crop}}{2} \right)^3 (T_{glass}) \quad 4.104$$

$$a_{57} = 4ALAI\varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{crop}}{2} \right)^3 (T_{surface}) \quad 4.105$$

The constant is given in equation 4.106.

$$b_5 = (-1)A \left( (1 - \rho_c)(\theta_s + \theta_l)(1 - e^{-kLAI})\alpha_{crop} + ET \right) \quad 4.106$$

### **Greenhouse Air Layer:**

The seventh layer of the unshaded model is the air in the greenhouse, which now comprises the previous volumes of air above and below the shade canopy. In the hour that the shades open it is assumed that the two previous air layers mix

adiabatically, as described in equations 4.5 and 4.6. Differences between the shaded and unshaded equations are:

- 1) there are no longer any exchanges between different air layers (since there is only one air layer),
- 2) no convective exchanges with the shade curtain,
- 3) convective exchange with the greenhouse glass.

In the new unshaded air layer energy flows into the layer from the bulk flow of air from outside the greenhouse through infiltration, and through the air vents. This layer also receives energy from convective heat transfer from the greenhouse glass, as well as from the greenhouse heater (if functioning), and from the supplemental lights, (if they are on). Evapotranspiration is also considered an energy input to this layer as well. Energy flows out through convective heat transfer with the crop canopy and the medium surface, as well as through the exchange of air to the outside through infiltration and venting.

The equation describing the flux of energy in is described in equation 4.107, the flux of energy out in equation 4.108, and the storage of energy in equation 4.109.

$$\begin{aligned}
 \text{flux in} = A & \left( \dot{q}_{heater} + \dot{q}_{lamps} + ET \right. & 4.107 \\
 & \left. + h_{glass-air} (T_{glass} - T_{air}) \right) \\
 & + \left( \frac{\dot{v}_1}{V_{sa\ vent}} \right) (1006(T_{vent} - 273.15) \\
 & + W_{vent} (2501000 + 1805(T_{vent} - 273.15))) \\
 & + \left( \frac{n_1}{V_{sa\ amb}} \right) (1006(T_{amb} - 273.15) \\
 & + W_{amb} (2501000 + 1805(T_{amb} - 273.15)))
 \end{aligned}$$

$$flux\ out = A \left( h_{crop-air} (T_{air} - T_{crop}) \right) \quad 4.108$$

$$+ h_{air-media} (T_{air} - T_{surface} )$$

$$+ \left( \frac{v_1}{V_{sa\ air}} \right) (1006(T_{air} - 273.15))$$

$$+ W_{air} (2501000 + 1805(T_{air} - 273.15))$$

$$+ \left( \frac{n_1}{V_{sa\ air}} \right) (1006(T_{air} - 273.15))$$

$$+ W_{air} (2501000 + 1805(T_{air} - 273.15))$$

$$storage = \left( \frac{V_1}{V_{sa\ air}} \right) (1006(T_{air} - 273.15)) \quad 4.109$$

$$+ W_{air} (2501000 + 1805(-273.15))$$

$$- \left( \frac{V_1}{V_{sa\ air\ t-1}} \right) (1006(T_{air\ t-1} - 273.15))$$

$$+ W_{air\ t-1} (2501000$$

$$+ 1805(T_{air\ t-1} - 273.15))$$

Putting these equations together and solving for the coefficients of each temperature term results in equations 4.110, 4.111, 4.112, and 4.113.

$$a_{64} = Ah_{media-air} (T_{surface} ) \quad 4.110$$

$$a_{65} = Ah_{crop-air} (T_{crop} ) \quad 4.111$$

$$a_{66} = (-1) \left( A(h_{glass-air} + h_{crop-air} + h_{media-air}) \right. \quad 4.112$$

$$\left. + \frac{(1006 + W_{air} * 1805)(n_1 + V_1 + v_1)}{V_{sa_{t-1}}} \right) (T_{air})$$

$$a_{67} = Ah_{glass-air}(T_{glass}) \quad 4.113$$

The constant is given in equation 4.114.

$$b_6 = (-1) \left( A(\dot{q}_{heater} + \dot{q}_{lamps} + ET) \quad 4.114$$

$$+ \left( \frac{v_1}{V_{sa_{vent}}} \right) (1006(T_{vent} - 273.15)$$

$$+ W_{vent} (2501000 + 1805(T_{vent} - 273.15)))$$

$$+ \left( \frac{n_1}{V_{sa_{amb}}} \right) (1006(T_{amb} - 273.15)$$

$$+ W_{amb} (2501000 + 1805(T_{amb} - 273.15)))$$

$$- \left( \frac{n_1 + V_1 + v_1}{V_{sa_{air_{t-1}}}} \right) (1006(-273.15)$$

$$+ W_{air} (2501000 + 1805(-273.15)))$$

$$+ \left( \frac{V_1}{V_{sa_{air_{t-1}}}} \right) (1006(T_{air_{t-1}} - 273.15)$$

$$+ W_{air_{t-1}} (2501000$$

$$+ 1805(T_{air_{t-1}} - 273.15))) \right)$$

### Greenhouse Glass Layer:

The new eighth and final layer of the unshaded model is the greenhouse glass. It is different from the shaded version in that it now has radiative exchange with the crop canopy and the medium surface. The equation describing the flux of energy in, is described in equation 4.115, the flux of energy out in equation 4.116, and the storage of energy in equation 4.117.

$$\begin{aligned}
 flux\ in = A & \left( h_{glass-ambient} (T_{ambient} - T_{glass}) \right. & 4.115 \\
 & + (1 - \rho_c)(\theta_s)\tau\alpha_{glass} \\
 & \left. + 4\varepsilon_{glass}\sigma\left(\frac{T_{glass} + T_{sky}}{2}\right)^3 (T_{sky} - T_{glass}) \right)
 \end{aligned}$$

$$\begin{aligned}
 flux\ out = A & \left( h_{glass-air} (T_{glass} - T_{air}) \right. & 4.116 \\
 & + 4LAI\varepsilon_{crop}\sigma\left(\frac{T_{glass} + T_{crop}}{2}\right)^3 (T_{glass} \\
 & - T_{crop}) \\
 & + 4(1 \\
 & - LAI)\varepsilon_{media}\sigma\left(\frac{T_{surface} + T_{glass}}{2}\right)^3 (T_{glass} \\
 & \left. - T_{surface}) \right)
 \end{aligned}$$

$$storage = 0 \quad 4.117$$



Putting these equations together and solving for the coefficients of each temperature term results in equations 4.118, 4.119, 4.120 and 4.121.

$$a_{74} = 4A(1 - LAI)\varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{glass}}{2} \right)^3 (T_{surface}) \quad 4.118$$

$$a_{75} = 4ALAI\varepsilon_{crop} \sigma \left( \frac{T_{glass} + T_{crop}}{2} \right)^3 (T_{crop}) \quad 4.119$$

$$a_{76} = Ah_{glass-air} (T_{air}) \quad 4.120$$

$$\begin{aligned} a_{77} = & (-1)A \left( h_{glass-ambient} + 4\varepsilon_{glass} \sigma \left( \frac{T_{glass} + T_{sky}}{2} \right)^3 \right. & 4.121 \\ & + h_{glass-air} + 4\varepsilon_{glass} \sigma \left( \frac{T_{glass} + T_{shade}}{2} \right)^3 \\ & + 4LAI\varepsilon_{crop} \sigma \left( \frac{T_{glass} + T_{crop}}{2} \right)^3 \\ & + 4(1 \\ & \left. - LAI)\varepsilon_{media} \sigma \left( \frac{T_{surface} + T_{glass}}{2} \right)^3 \right) (T_{glass}) \end{aligned}$$

The constant is given in equation 4.122.

$$\begin{aligned} b_7 = & (-1)A \left( h_{glass-ambient} (T_{ambient}) + (1 - \rho_c)(\theta_s)\tau\alpha_{glass} \right. & 4.122 \\ & \left. + 4\varepsilon_{glass} \sigma \left( \frac{T_{glass} + T_{sky}}{2} \right)^3 (T_{sky}) \right) \end{aligned}$$

### 4.3.3 Greenhouse Controls:

#### *Heating/venting*

To generalize the model, the heating and venting capacities of the greenhouse model are based on the area of the greenhouse. The heating capacity of the model greenhouse was determined through the recommended standards of the ASABE (2006). Heaters are sized such that when the outdoor temperature is at the 99% winter design dry bulb temperature, the setpoint of the greenhouse can be maintained, with no solar input. The equation to determine the heater capacity is made up of losses due to radiation, conduction and convection, and through infiltration, neglecting losses through the floor and around the perimeter:

$$q_{heater} = UA_c(t_i - t_o) + 1800VN(t_i - t_o) \quad 4.123$$

where: U is the approximate overall, heat transfer coefficient ( $\text{W m}^{-2} \text{C}^{-1}$ )  
 $A_c$  is the area of the greenhouse cover ( $\text{m}^2$ )  
 $t_i$  is the greenhouse air temperature (C)  
 $t_o$  is the outside air temperature (C)  
1800 is the assumed specific heat of air ( $\text{J m}^{-3} \text{C}^{-1}$ )  
V is the volume of the greenhouse ( $\text{m}^3$ )  
N is the infiltration rate (air exchanges per second) ( $\text{s}^{-1}$ )

Typical values for these parameters are available in the ASABE standards. Assuming single sealed glass, U is  $6.2 \text{ W m}^{-2} \text{K}^{-1}$ , 1 hectare floor area greenhouse,  $A_c$  is  $12500 \text{ m}^2$ ,  $t_i$  is  $24\text{C}$ ,  $t_o$  is  $-15.8 \text{ C}$  (ASHRAE 2009, Binghampton, NY value), V is  $3.0\text{E}4 \text{ m}^3$ , N is  $2.8\text{E}-4 \text{ s}^{-1}$  due to well maintained glass, the total heat input required to match the load is  $3.68\text{E}6 \text{ W}$ . If we divide this number by the area this corresponds to a heating capacity of  $1327 \text{ kJ m}^{-2} \text{hr}^{-1}$  ( $370 \text{ W m}^{-2}$ ).

Venting capacity can be estimated from the equation:

$$(1 - E)\tau IA_f = UA_c(t_i - t_o) + \left(\frac{Q_v A_f c p_{ex}}{V_{ex}}\right)(t_i - t_{inlet}) \quad 4.124$$

where: E is the evapotranspiration coefficient  
 $\tau$  is the solar transmissivity of the greenhouse  
I is the solar radiation, ( $\text{W m}^{-2}$ )  
 $A_f$  is the floor area ( $\text{m}^2$ )  
 $V_{ex}$  is the specific volume of the air leaving the greenhouse ( $\text{m}^3 \text{kg}^{-1}$ )  
 $Q_v$  is the ventilation rate ( $\text{m}^3 \text{s}^{-1} \text{m}^{-2}$ )  
 $c p_{ex}$  is the specific heat of the air leaving the greenhouse ( $\text{J kg}^{-1} \text{C}^{-1}$ )  
 $t_{inlet}$  is the temperature of air entering the greenhouse (C)

Following the guidelines of the ASABE standards, values for these parameters can be estimated. Assuming a value of 0.5 for E, 0.7 for  $\tau$  (standards suggest 0.88, but that is for glass alone, not the entire greenhouse structure (Albright, personal communication)), for  $\tau$ ,  $900 \text{ W m}^{-2}$  for I for the July 21<sup>st</sup> solar noon,  $1.0\text{E}4$  for  $A_f$ ,  $30\text{C}$  for  $t_i$ ,  $0.8695 \text{ m}^3 \text{kg}^{-1}$  for  $V_{ex}$ ,  $1006 \text{ J kg}^{-1} \text{C}^{-1}$  for  $c p_{ex}$ ,  $28.0 \text{ C}$  for  $t_o$  (from the 1% cooling DB temperature for Binghamton, NY), and  $23.1 \text{ C}$  for  $t_{inlet}$  (assuming the temperature immediately inside the cooling pads is  $2.0\text{C}$  higher than the Mean Coincident Wetbulb Temperature from the 1% MCWB for Binghamton, NY of  $21.1 \text{ C}$ ) results in a ventilation rate ( $Q_v$ ) of  $0.0375 \text{ m}^3 \text{s}^{-1} \text{m}^{-2}$  ( $135 \text{ m}^3 \text{h}^{-1} \text{m}^{-2}$ ).

The calculation to determine the amount of heating and/or venting occurring during the simulated hour was based on a user defined air temperature setpoint, with a tolerance of  $0.1 \text{ K}$ . The simulation was then run with the maximum heating rate. If the resulting predicted air temperature was below the setpoint minus the tolerance, the maximum heating value was kept, as the heating/cooling setting. If the predicted air temperature was above the setpoint minus the tolerance, the simulation was run with

maximum venting. If the resulting predicted air temperature was above the setpoint plus the tolerance, the maximum venting rate was kept as the heating/cooling setting. If the predicted air temperature was below the setpoint plus the tolerance, it was determined that the setpoint temperature was within the ability of the system to achieve. To determine whether heating or cooling is required, the simulation was run with no heating or venting. If the predicted temperature was above the setpoint, then venting was required; below the setpoint, heating. To determine the correct heating (or venting) setting, a bisection search was used. The bisection algorithm was run until the achieved predicted temperature was within the setpoint +/- the tolerance.

#### *Pad cooling*

In the summer time, when keeping the greenhouse cool enough is not possible through venting with ambient air alone, evaporative cooling is enabled. The air drawn into the greenhouse for venting purposes is first passed through a pad saturated with water. The air causes some of the water to evaporate, increasing the humidity ratio of the air. However, the enthalpy of the air remains almost the same, which means the air temperature is decreased. A process efficiency of 0.75 was assumed. The new dry bulb temperature resulting is given by equation 4.125.

$$T_{dbnew} = T_{db} - (T_{db} - T_{wb})\epsilon_{pad} \quad 4.125$$

where:  $T_{db}$  is the dry bulb temperature (K)

$T_{wb}$  is the wet bulb temperature (K)

$\epsilon_{pad}$  is the process efficiency

For a well functioning cooling pad the process efficiency  $\epsilon_{pad}$  is assumed to be 0.75.

The new humidity ratio for the air is

$$W_{new} = \left( \frac{(2501.0 - 2.381(T_{wb} - 273.15))W_{sat} - (T_{dbnew} - 273.15) + (T_{wb} - 273.15)}{2501.0 + 1.805(T_{dbnew} - 273.15) - 4.186(T_{wb} - 273.15)} \right) \quad 4.126$$

where:  $W_{sat}$  is the saturation humidity ratio

The remainder of the properties of the air can be calculated from these two values.

The cooling pads are only used for certain months of the year, and the decision to employ them is made by the grower. To simplify this decision the simulation model assumes cooling pads are in use when the outside drybulb air temperature is greater than 20C. This decision is made on an hourly basis, though in reality a grower usually only decides whether or not to use the cooling pads on a daily or longer basis.

### *Lighting and shading*

The control of supplemental lighting and shading was simulated according to the LASSI (Light And Shade System Implementation) algorithm described in Albright (1996). It is a control algorithm to optimize the cost of providing a constant daily light integral. The decision timeframe for the algorithm is on an hourly basis which matches the simulation time-step.

Outside of the LASSI implementation, the shades were closed during the night to conserve energy. Shades were automatically closed in the hour after sunset occurs, and opened in the hour before sunrise is predicted.

To simulate the heat load, and light output of supplemental lighting, the number of luminaires  $m^{-2}$  was taken from a commercial greenhouse located in Dryden, NY (Albright personal communication). In this greenhouse, 146 high pressure sodium luminaires at 675 W each (including ballast) light  $595 m^2$  ( $6400 ft^2$ ). This provides a

light level of  $180 \mu\text{mols PPF m}^{-2} \text{ s}^{-1}$ . Assuming the lights are 20% efficient  $4.77\text{E}5 \text{ J m}^{-2} \text{ hr}^{-1}$  of heat is transferred to the greenhouse air, and  $1.19\text{E}5 \text{ J m}^{-2} \text{ hr}^{-1}$ , is transferred as radiation to the crop and medium.

#### *Nutrient solution temperature control*

If the nutrient solution temperature is to be set at a fixed temperature, as in the case of modeling nutrient solution temperature control, the assumption is made that nutrient solution temperature has a negligible effect on the air and crop temperatures. This assumption allows use of the same greenhouse climate data set for nutrient solution temperature control, as in the uncontrolled condition (though the values of the nutrient solution are set at the specified target). The assumption implies that nutrient solution cooling equipment is sized to be able to handle the cooling load under any conditions.

#### 4.3.4 Input to the model and initial conditions:

The ambient environmental conditions which constitute the input to the model, are the outdoor dry bulb temperature, the relative humidity, and the solar irradiation. The data set for the model was collected at the Game Farm Road station, at Cornell University in Ithaca, NY over the course of twelve years from 1985 to 1996. The original data files consisted of the year, month, day and hour, and the corresponding temperature in F, relative humidity in %, and solar irradiation in  $\text{BTU ft}^{-2}$ . The program automatically converts the units of temperature to C, and solar irradiation to  $\text{W m}^{-2}$ . The original 12 data files were combined into a single text file containing all the information, to facilitate the running of the program.

Initial conditions for the greenhouse model were generated by running the program using 1988 data with a deep soil temperature of  $9.88 \text{ C}$  ( $49.8 \text{ F}$ ) which is the average air temperature for the years 1985 to 1997. The values on December 31<sup>st</sup>, 11

pm, were then set as the initial temperatures. It was felt that any major divergence between the crop and air temperatures, which change quickly in response to climate conditions, is outweighed by the subsequent more typical values of soil, concrete, pond and medium temperatures which are slower to respond to changes in outdoor climate.

#### 4.3.5 Implementation of Greenhouse model

The greenhouse simulation model was coded in Java. The JAMA package was imported to facilitate solving of the matrices. Input and output files were encoded as tab delimited text files. Computer code is presented in Appendix A.

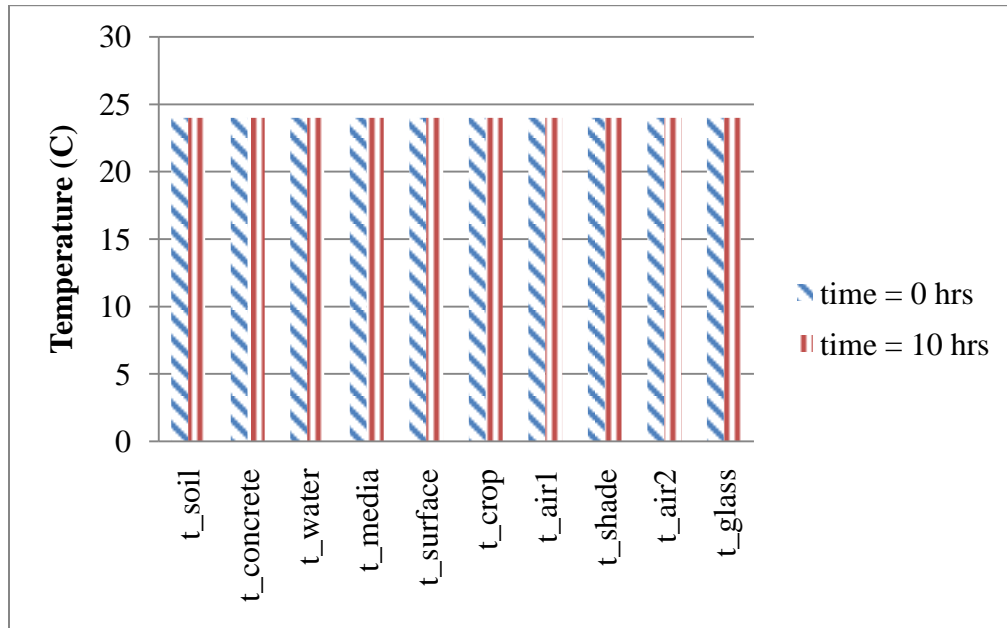
### **4.4 RESULTS:**

#### 4.4.1 Model testing

##### *Steady State*

To determine if the equations of the model have been input correctly and the system behaves as expected, steady state testing was used. Setting the initial temperature of all layers and the boundary conditions to the same temperature of 24C, and disabling all heat inputs such as the heating/cooling system, the pump and natural and supplemental light, should result in all layers maintaining their initial temperatures if the equations and signs of all terms are input correctly. The results of this test are plotted in Figure 4.2.

If the sign of a term was reversed it would result in a net gain (or loss) of energy in a particular layer which would then translate into a change in temperature within that and surrounding layers. Because the temperatures remain the same as their initial value, no such reversal is present in the equations.



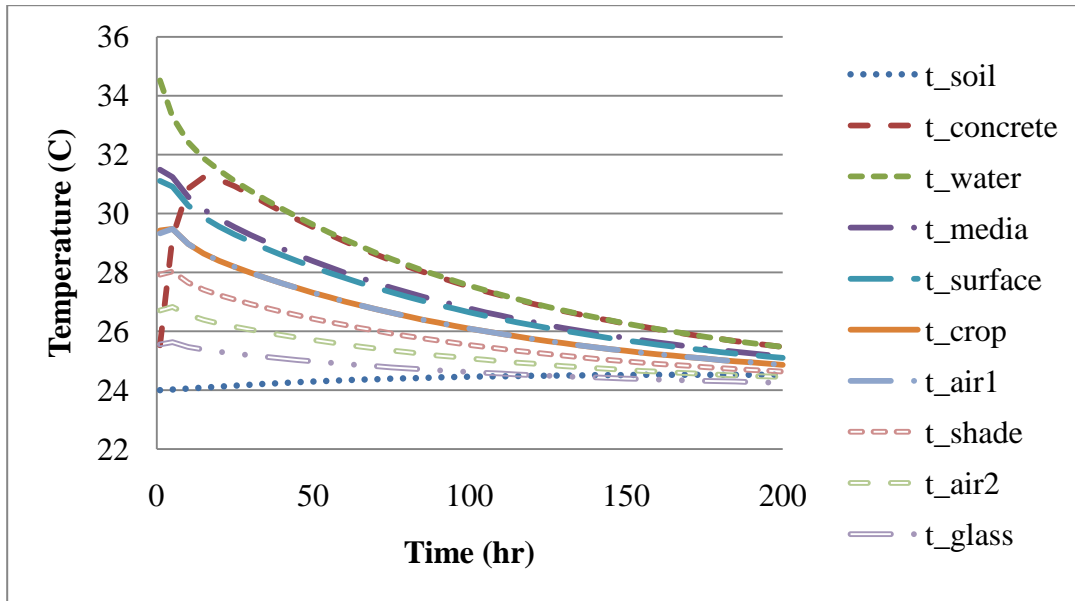
**Figure 4.2** Model temperatures with a common initial temperature and no heat inputs/outputs.

### *Step Input*

To determine if the equations correctly predict the direction of heat flow, and that parameter values for the convective, radiative and conductive heat transfer coefficients are reasonable, a step input test was used. By setting the initial temperature of a particular layer higher (or lower) than the rest of the layers, an energy imbalance is introduced. Differences in temperature (energy) drive heat flow, and so the model should seek to balance the level of energy within all the layers through heat transfer.

Figure 4.3 is a plot of the temperatures of the layers over the course of 200 hours, when the initial temperature of the pond was 35C, and the remaining layers, 24C.





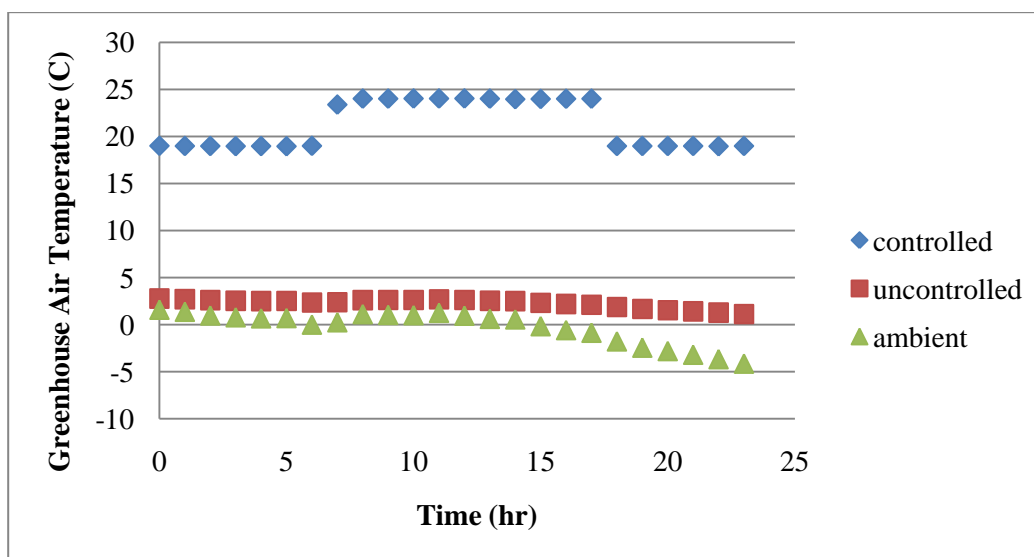
**Figure 4.3** Temperatures of the greenhouse simulation model layers as a function of time when the initial temperature of the pond is set to 35C and the remaining layers to 24C.

As expected the temperature in the pond only drops from 35C as there are no further energy inputs to the model. The concrete (because of its relatively large thermal mass) is slower to respond to the temperature change than the other layers, but eventually does, and then closely follows the temperature of the pond. The soil with an even larger thermal mass, barely responds at all. The medium and the medium surface are quick to respond to the temperature difference and also closely follow one another. The greenhouse air in greenhouse airspace 1 and 2, the shade and the greenhouse glass are also quick to respond to the energy input and follow the same trend as one another, however, the farther removed from the pond water layer, the less the response. All layers tend to approach the initial temperature of 24C, as the extra energy within the pond water layer is dispersed to the other layers.

### Heating and Cooling

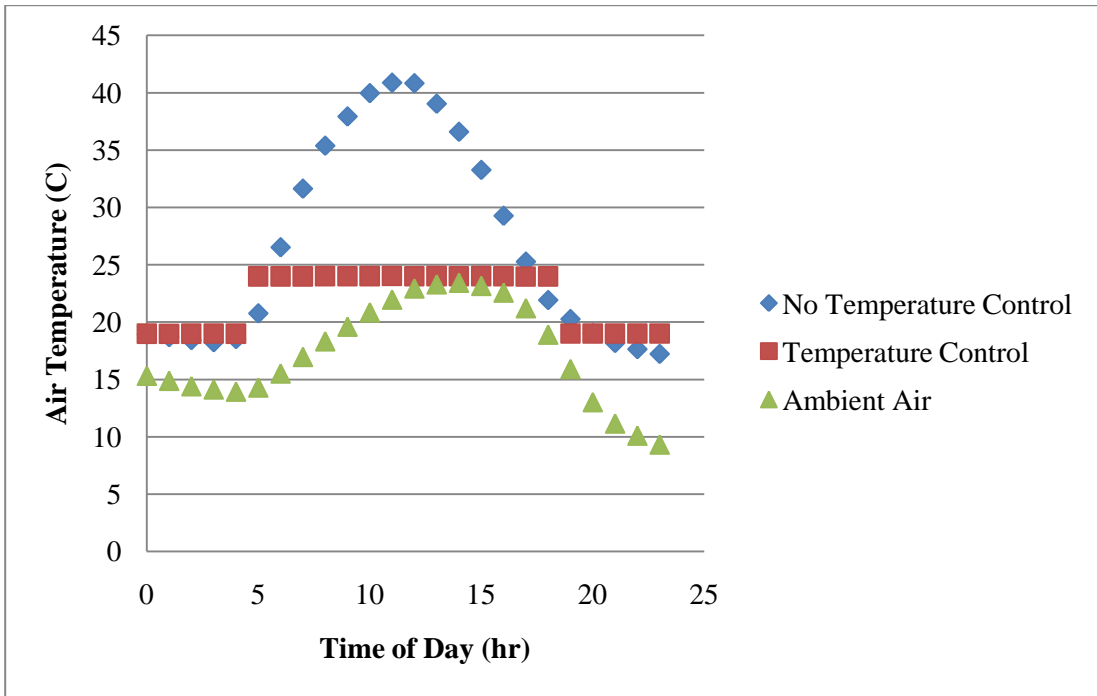
To test the functionality of the heating and cooling simulation systems it was necessary to use real environment data for the ambient conditions outside the greenhouse. The data used was collected in 1988 from the Northeast Climate Center weather station located on Game Farm Road near Ithaca, NY. 1988 data was used as that was the dataset with the fewest incidences of missing or incomplete data. Figure 4.4 illustrates the effect of the greenhouse heating system on the greenhouse air during a typical cold dark winter day. The controlled air temperatures closely follow the setpoints of 24C during the day and 19C during the night. It is apparent at 7:00 am the heating system is not quite capable of increasing the temperature within the greenhouse from the night temperature to the day temperature, though it is close, and matches real world experience.

The uncontrolled temperatures closely follow the ambient conditions and are unsuitable for growing spinach. This particular day was quite dark, as there was little change in temperature during the sunlit hours.



**Figure 4.4** Effect of heating (24/19 day/night setpoint) on simulated greenhouse air temperature during a typical cold, dark winter day.

Figure 4.5 is a plot of the greenhouse temperatures both with and without the cooling system functioning on a warm and sunny day.



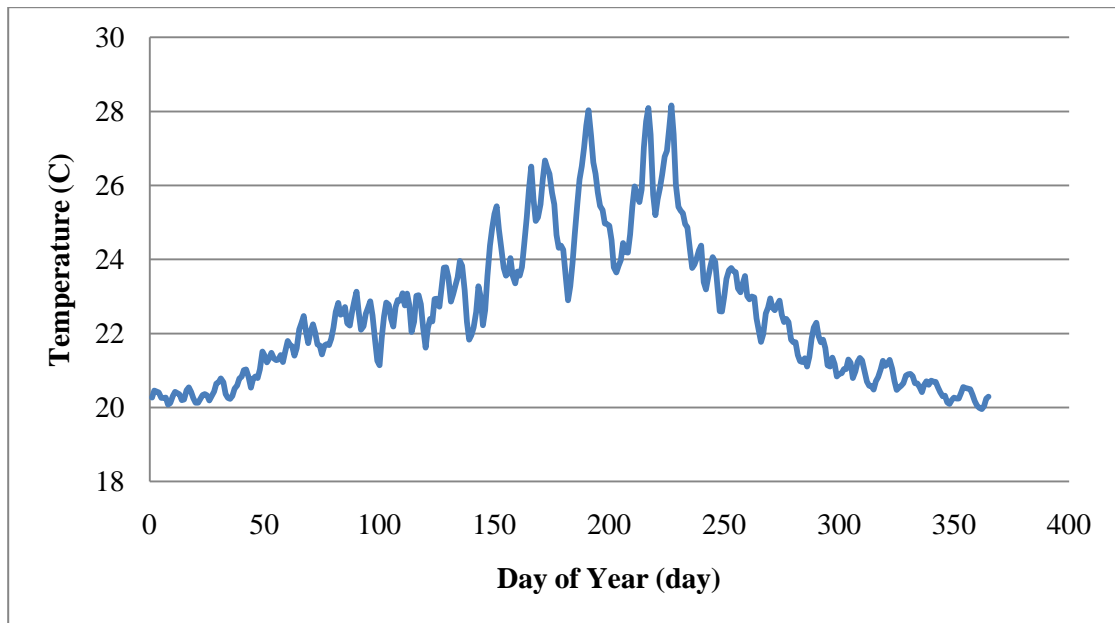
**Figure 4.5** Effect of pad cooling and venting on greenhouse air temperatures (24/19 day/night setpoint) during a typical warm sunny day.

On this particular day the heat gain from solar radiation is quite high and so the temperature within the unvented greenhouse reaches an unacceptably high temperature of 41C which also matches real world experience. Venting with evaporative cooling is quite capable of handling this extra heat load as the day temperature setpoint of 24C is not exceeded.

4.4.2 Validation of model

Unfortunately there is no readily comparable data with which to compare the predicted temperatures of the simulation model. The closest commercial facility, Fingerlakes Fresh (Challenge Industries, Ithaca, NY), collects pond temperatures on a daily basis, however, this data is of limited use because the ponds are manually heated

in the winter, and cooled in the summer with no associated record of the energy inputs. According to the temperature logs the ponds are kept at approximately 24C year round. Figure 4.6 is a plot of the predicted pond temperatures using the 1988 Ithaca data set.



**Figure 4.6** Simulated Pond temperatures over the course of the year using 1988 weather data from Ithaca, NY.

It is possible to examine this plot of temperatures for reasonability. Over the course of the year there are periods where the pond temperature rises considerably above the 24 C setpoint of the air temperature within the greenhouse. These periods of higher temperature occur in the summer when solar heat loads are high. In addition the pond temperature never drops below the night air temperature setpoint of 19 C. For the bulk of the year the pond temperature stays within the range of 19 to 24C. Experience has shown temperatures as high as 27C or so when the pond is not cooled for awhile. Also of significance is the fact that temperature changes are relatively

slow; certainly slower than the temperature changes of the greenhouse air. The largest changes are only on the order of a few degrees over the course of several days.

The temperatures are also similar to the published range of 27 (summer) and 17 (winter) that Bates and Stanghellini (1984) encountered in Arizona in their commercial spinach pond study.

#### **4.5 DISCUSSION:**

The temperature of the nutrient solution has been identified as a key aspect of the development of *Pythium* infections in hydroponic baby spinach production. To simulate the growth and development of both a spinach crop and *Pythium* infection it is essential a greenhouse model includes a prediction of the nutrient solution temperature. Another key aspect of *Pythium* infections is the time that spinach crops spend in the nutrient solution before they are harvested, and infected roots removed. It is also necessary that a greenhouse simulation model includes modern environmental controls, particularly lighting and shading, as the amount of light a crop receives dictates its size at harvest time.

The developed lumped parameter, step wise steady state model incorporates lighting and shading control in addition to the necessary heating and cooling controls present in commercial greenhouses located in the Northeastern United States. Using the solar radiation, drybulb temperature and relative humidity as input, the model predicts on an hourly basis, the temperature of the nutrient solution, and other temperatures within the greenhouse. Though no data is available to validate the predictions of the model directly, functionality and reasonability checks on the output of the model have shown the results are consistent with expectations.

The predicted nutrient solution temperatures respond to the change of season and roughly mimic temperatures encountered in other commercial greenhouse

settings. An important feature of the response is that temperature changes of the nutrient solution are slow due to the relatively large thermal mass of the pond.

The absolute accuracy of the model is also of lesser importance because of the eventual use of the model. The results of the model are to be used in a Monte-Carlo style simulation of various spinach production systems in the presence of *Pythium aphanidermatum*. To limit the effects of the greenhouse simulation model on the final results of the Monte-Carlo simulation, the greenhouse simulation model will be used to generate a common output that will be used for all of the Monte-Carlo runs. Any error due to the greenhouse simulation will be common to the results of the Monte-Carlo simulation.

Careful recording of the energy inputs used to heat and cool the nutrient solution in a commercial setting would allow a more rigorous validation of the model in the future. Such a model could then be modified to calculate the energy required to maintain pond temperatures under a variety of environmental conditions. These estimations would prove a valuable tool in sizing pond heating and cooling systems.

## CHAPTER 5: SPINACH GROWTH MODEL

### 5.1 INTRODUCTION:

To simulate the production of hydroponic spinach, a commonly used growth model SUCROS (Simple Universal CROp growth Simulator) (Goudriaan and van Laar, 1994) was adapted for controlled environment conditions.

SUCROS was originally developed at Wageningen University in The Netherlands (Spitters et al. 1989) as a means of describing crop growth as a function of the main processes of CO<sub>2</sub> assimilation and respiration as they are influenced by environmental conditions. Alternative approaches to crop modeling using regression modeling, rely on large sets of data with many different conditions, to accurately predict crop growth (Spitters 1990). SUCROS, or another similar mechanistic based model, is useful for modification and use in crop disease work since real-world data incorporating disease, environmental conditions and crop stage for use in a regression model would be impractical to obtain, and limited in use.

In his dissertation, Both (1995) modified SUCROS for use in predicting yield of a lettuce variety grown in controlled environment conditions. The original SUCROS model was originally developed from crops grown outdoors, not in the controlled conditions of a commercial greenhouse. Both also looked at supplemental light and natural light within the greenhouse environment and CO<sub>2</sub> as a means of achieving growth targets, which required further modification of the model as the original assumed ambient CO<sub>2</sub> concentrations, and natural lighting only.

SUCROS relies on a number of parameters to describe the growth and partition of assimilates into the roots and shoot (leaf and stem). Some of these parameters can be generalized from other crops, while others are more specific to spinach. The values of these parameters can be found by applying the model to real world data already collected to study the growth of hydroponic baby leaf spinach. Such data was

collected both as a part of the original NYSERDA spinach project, and also during the subsequent NYSERDA spinach disease study. From this data and the developed parameters, the SUCROS framework can be used to describe the growth and development of a spinach crop. Because the crop to be harvested is baby leaf spinach only vegetative growth needs to be considered. The crop is harvested before energy is put into reproductive structures, which would change the growth of the organs of commercial interest, namely the leaves.

The objectives of this chapter are to:

1. adapt the modified SUCROS growth model developed by Both, to an hourly basis,
2. develop the parameters necessary to describe hourly baby leaf spinach growth, from literature values,
3. use actual growth data/measured values to provide the parameters unavailable from the literature,
4. use actual growth data to validate the model.

## **5.2 LITERATURE REVIEW:**

There are many options available for modeling the growth of a particular crop. Simple regression analyses can allow the estimation of crop yields based on environmental inputs such as light and temperature. However, a major drawback associated with these types of models is that they require the data used to develop the regression parameters vary over the range for which the model is expected to be valid. To acquire such data would require a prohibitive number of experiments under varying conditions.

Another form of crop modeling is the mechanistic approach. This form of model seeks to predict yields based on the underlying physical processes that drive it.



These types of models are more complicated in nature and, overall, predictive accuracy may suffer due to the many parameters used, each of which may have its own level of inaccuracy. However many of the parameter values can be found in the literature either for a specific crop, or as a general value for C3 or C4 photosynthesis. In addition, the conditions within a greenhouse are usually held within a tightly controlled range, and are not subject to the large seasonal variations that an outdoor crop faces. This is an important consideration as the resulting model should not be used to predict results for situations that are far outside the range of conditions used to develop the model.

For the spinach growth model it was decided a mechanistic model would be best suited for the purposes of modeling the growth of the crop under the conditions of damage done by *Pythium aphanidermatum*. A mechanistic model offers the opportunity to directly modify the parameters affecting growth due to *Pythium* damage; this modification would not be as meaningful/applicable in a regression type model. The mechanistic model is also more suitable for making calculations on an hourly basis.

Many mechanistic models have been developed, but one of the most popular is the Simple and Universal CROp growth Simulator (SUCROS) (Spitters et al. 1989). SUCROS functions by determining crop growth based on processes such as CO<sub>2</sub> assimilation, light interception, respiration and assimilate partitioning. The model itself is made up of several submodels, each used to describe some process of growth. Many of the parameters necessary for the model can be found or adapted from the literature, or determined experimentally.

SUCROS simulates the dry matter accumulation (growth) of the crop by partitioning the carbohydrates produced through photosynthesis into what is required for maintenance (respiration), and what is available for plant growth. The

carbohydrates available for crop growth are then partitioned into the roots/shoot, of which the stem portion is further divided into that going to the stem and that to the leaves. Baby spinach is harvested very early in its growth cycle when growth is strictly vegetative; it is not necessary to allocate carbohydrates to reproductive structures such as flowers and seeds. To determine the amount of carbohydrate available for distribution throughout the plant, SUCROS calculates the CO<sub>2</sub> assimilation rate, which is dependent on the PAR (Photosynthetically Active Radiation) intercepted by the canopy. This assimilation rate in turn is a function of the light intensity/characteristics (direct and diffuse) and the leaf area of the crop.

The original SUCROS model was developed to describe growth in field conditions which are obviously quite different from those encountered in controlled environment agriculture. In his dissertation, Both (1995) modified the SUCROS model to account for supplemental lighting and CO<sub>2</sub> enrichment. Another factor affecting the interception of light by the crop is the greenhouse structure itself. The transmittance of the greenhouse glass (which varies with the angle of the sun) and structure is also accounted for in Both's modifications.

### **5.3 METHODS AND MATERIALS:**

During the original NYSERDA funded spinach production project (Albright et al. 2005) an experiment was conducted to collect growth data for the purposes of developing a model to help predict optimal harvest times under specific environmental conditions previously selected as optimal. This experiment consisted of growing a spinach crop and harvesting a number of rows of plants every two days. The harvested material was then weighed, and the leaf area measured. A subsample of the harvest was used to obtain the dry matter content. Root to shoot ratio was measured for new seedlings and plants at final harvest by removing the plug of medium the

individual plant was growing in, along with the roots. The shoot was removed and placed in a bag for dry mass measurements while the medium was rinsed from the roots which were also bagged for dry mass measure.

### 5.3.1 Spinach Growth Experiments:

#### *Facilities:*

The experiment was conducted in a single section of greenhouse (section 15A of the Kenneth Post Greenhouse complex on the Cornell University Ithaca campus) from December 4<sup>th</sup>, 2004 to December 20<sup>th</sup>, 2004. This section of greenhouse measures 8.5 m by 10.7 m with an eave height of 2.2 m and a roof slope of 25 degrees. It is covered with glass with an estimated structural light transmittance of 0.7 at midday. Supplemental lighting was provided by an array of 21 HPS (high pressure sodium) luminaires arranged to provide a uniform  $180 \mu\text{mol m}^{-2} \text{ s}^{-1}$  of PAR at the crop height of 1.1 m above the floor.

The crop was grown in two blue plastic ponds (1.2 m \* 2 m) with a depth of 0.3 m, containing standard half-strength Hoaglands nutrient solution. The sides and bottom of the ponds were insulated with 0.07m of polyurethane foam insulation. Circulation of the nutrient solution was achieved with a 1/20 hp centrifugal pump. The crop was grown in Styrofoam “Speedling” style floats, which were divided into 26 rows with 13 cells per row. Spinach (cv. Alrite) was seeded into Rediearth® medium at a moisture content of 3:1 (water to medium by weight) and allowed to germinate 48 hours at 25 C in a darkened growth chamber. At the time of floating, the seedling emergence percentage was measured and corrected to provide the same number of plants per float. Approximately 75% of the cells contained viable plants after the thinning, resulting in a plant density of 1200 plants  $\text{m}^{-2}$ .

The air temperature of the greenhouse compartment averaged 23.0 C (sd 0.44) day/night over the course of the experiment. Solution temperature was not continuously measured or recorded, but typically follows air temperature closely, particularly at the time of year the experiment was conducted. Electroconductivity was maintained at 1200  $\mu\text{S cm}^{-1}$  and pH 5.8. Measurements and corrections of the solution were made every two days. Daily light integral over the course of the experiment averaged 19.3  $\text{Mol m}^{-2} \text{day}^{-1}$  (sd 1.47).

#### *Harvests:*

Harvesting of two rows of plants per pond started two days after floatation when rows 1 and 2 were harvested. At this stage of growth the plants were small and edge effects due to plant interactions were felt to be negligible. Four days after floatation, rows 3 and 4 were harvested. At six days after floatation the plants were beginning to interact with each other, so three rows were harvested. The exposed row acted as a guard row, which was not used for measurements. The use of a guard row continued until the final harvest 16 days after floatation. Immediately upon cutting, the fresh weight for each plant was measured, recorded and placed in a labeled brown paper bag. Following harvest, the leaf areas of the cotyledons and true leaves were measured and recorded. After leaf area measurements the plants were placed back in their bags and placed in a drying oven at 70 C. After 72 hours, they were removed from their bags and weighed to determine dry mass. This procedure was done in both ponds at the same time to provide a comparison between ponds.

The ratio of root to shoot was determined by selecting 10 plants from a float and removing entire individual plants (shoots and roots) along with their medium. The shoots were removed and bulked into paper bags for dry weight measurement. The roots were rinsed to remove the medium and also bulked into a different paper

bags for dry weight measurement. This procedure was carried out on three different flats at both the time the crop was originally floated, and at final harvest.

### 5.3.2 Adaptation of SUCROS to baby spinach growth:

The original SUCROS model by Spitters (Spitters et al. 1989) was developed for wheat and was adapted by Both (1995) for greenhouse production of lettuce. A major modification from the original SUCROS and Both's version is the time step used for the model. When modeling crops such as wheat in outdoor conditions the growth cycle is longer than the relatively quick sixteen day crop cycle used to produce baby leaf spinach. To account for the addition of supplemental lighting, the controlled conditions in greenhouses and the short crop cycle of baby leaf spinach, it was decided to shorten the time step from one day to one hour. This time step also corresponds to the time step used in the greenhouse climate simulation model, the time step used in supplemental light and shading decisions, as well as to the frequency of most climatic data collection.

Many of the parameters to describe growth can be extended from the wheat (original SUCROS) and lettuce (Both version of SUCROS) to spinach; however some adaptation of their values was necessary to more closely fit the growth curves observed in the baby spinach growth experiment detailed previously. Data from pond one were used to develop the parameters used in the model, and the results of the model compared to the data from pond two.

Equations for the SUCROS growth model are not presented in this text unless they were modified. The equations can be found in Appendix A, the computer coding of the model; however, a more detailed explanation and derivation of these equations is presented in Goudriaan and van Laar (1994). The values of parameters required for

the model are presented in the following sections, along with a justification for their selection. The parameters are summarized in Table 5.1.

**Table 5.1** Symbols and definitions of parameters and variables used in the modified SUCROS hourly spinach growth model.

$\tau$	Greenhouse transmittance
A	Coefficient relating leaf mass to Leaf Area Index
AMX	Maximum CO <sub>2</sub> assimilation rate (kg CO <sub>2</sub> ha <sup>-1</sup> hr <sup>-1</sup> )
DAYL	Length of day (sunrise to sunset) (hr)
DEC	Declination angle of the sun (radians)
DOY	Day of the year
DSINB	daily integral of the sine of solar inclination (s day <sup>-1</sup> )
DSINBE	Daily integral of SINB with a correction for lower atmospheric transmission at lower solar elevations (s day <sup>-1</sup> )
DTGA	Daily gross crop CO <sub>2</sub> assimilation (kg CO <sub>2</sub> ha <sup>-1</sup> d <sup>-1</sup> )
EFF	Initial light conversion factor for individual leaves (kg CO <sub>2</sub> ha <sup>-1</sup> leaf h <sup>-1</sup> )/(J m <sup>-2</sup> s <sup>-1</sup> )
FRDF	Fraction of radiation that is diffuse
FLV	Fraction of photosynthate diverted to leaves
FRT	Fraction of photosynthate diverted to roots
FSH	Fraction of photosynthate diverted to shoot
FST	Fraction of photosynthate diverted to stem
H <sub>d</sub>	Average diffuse radiation (W m <sup>-2</sup> )
H <sub>day</sub>	Daily light integral outside the atmosphere (W m <sup>-2</sup> )

**Table 5.1 continued**

H <sub>hour</sub>	Hourly integral outside the atmosphere ( $\text{W m}^{-2}$ )
HTGA	Hourly gross crop $\text{CO}_2$ assimilation ( $\text{kg CO}_2 \text{ ha}^{-1} \text{ h}^{-1}$ )
HTR	Hourly total radiation ( $\text{W m}^{-2}$ )
K <sub>t</sub>	Ratio of actual daily radiation on a horizontal surface to the daily extraterrestrial radiation on a horizontal surface
KDF	Extinction coefficient for the canopy ( $\text{ha ground ha}^{-1} \text{ leaf}$ )
LAI	Leaf Area Index. Area of leaves per unit area of ground
LAT	Latitude of the greenhouse location (radians), + if north
PAR	Photosynthetically active radiation ( $\text{W m}^{-2}$ )
pcPAR	Percentage of radiation that is in the PAR wavelengths
SCP	Scattering coefficient of leaves for PAR (unitless)
SC	Solar constant ( $\text{W m}^{-2}$ )
SHA	Sunset Hour angle (radians)
WLV	Mass of leaves ( $\text{kg ha}^{-1}$ )
WRT	Mass of roots ( $\text{kg ha}^{-1}$ )

*Initial conditions:*

The SUCROS model starts when the seedlings first emerge from the growth medium. This emergence also corresponds approximately to the time that baby spinach seedlings are placed in the pond. The initial conditions required for the model are:

1. leaf area index
2. initial amount of dry matter in the leaves and roots.

These values were determined through experimental measurement of a newly emerged crop. The typical planting density used in experiments was approximately 1200 plants  $\text{m}^{-2}$ . The leaf area index at time of floating is approximately 0.1 ( $\text{ha leaf ha}^{-1}$  ground); the initial dry matter in the leaves is approximately 33  $\text{kg leaf ha}^{-1}$ , and roots 5  $\text{kg roots ha}^{-1}$ .

#### *Crop Development:*

Because we are concerned with a babyleaf product, determination of the developmental stage of the crop is unnecessary. The crop always stays in a vegetative growth state, which makes the development stage and development rate unneeded in this model.

#### *Leaf CO<sub>2</sub> assimilation:*

Leaf CO<sub>2</sub> assimilation rate is another parameter that is affected by temperature. The maximum value (AMX) at light saturation is 40  $\text{kg CO}_2 \text{ ha}^{-1} \text{ hr}^{-1}$  at ambient CO<sub>2</sub> concentrations. Following Goudrian and van Laar (1994) the CO<sub>2</sub> assimilation rate (AMAX) linearly increases from 0 to 100% of the maximum value for temperatures from 0 to 10 C, is 100% of the maximum value from 10 to 25 C, and then linearly decreases from 100% to 0% for temperatures from 25 to 35 C.

#### *Daily Gross CO<sub>2</sub> assimilation*

The original SUCROS model determined the daily gross crop CO<sub>2</sub> assimilation (DTGA), but because the spinach model is hourly, an hourly gross crop CO<sub>2</sub> assimilation (HTGA,  $\text{kg CO}_2 \text{ ha}^{-1} \text{ h}^{-1}$ ) was used. The gross CO<sub>2</sub> assimilation is determined from the amount of PAR intercepted by the crop ( $\text{J m}^{-2} \text{ s}^{-1}$ ) canopy and the



CO<sub>2</sub> assimilation/light response of the individual leaves. Three parameters are used in the assimilation/light response of the leaves, EFF, KDF and SCP.

1. EFF is the initial light conversion factor for individual leaves ( $\text{kg CO}_2 \text{ ha}^{-1} \text{ leaf h}^{-1} / (\text{J m}^{-2} \text{ s}^{-1})$ ).
2. KDF is the extinction coefficient for the canopy ( $\text{ha ground ha}^{-1} \text{ leaf}$ ).
3. SCP is the scattering coefficient of leaves for PAR (unitless).

For ambient CO<sub>2</sub> concentrations (which is what is assumed in this model) a typical value of EFF is  $0.45 \text{ kg CO}_2 \text{ ha}^{-1} \text{ h}^{-1} \text{ per J m}^{-2} \text{ s}^{-1}$ . Both (1995) used this value in his model for lettuce growth. 0.45 is also the value in the original SUCROS model for wheat, and the value selected for this baby spinach model.

The extinction coefficient for leaves (KDF) suggested in the original SUCROS model for wheat is  $0.6 \text{ ha ground ha}^{-1} \text{ leaf}$ . Both (1995) used a value of 0.72 for lettuce. Monteith (1969) tabulated several values of KDF for several different species, and categorized crop foliage as either planophile or erectophile; planophile canopies are more evenly spread out, while erectophile canopies tend to be more upright and potentially clustered closely around the plant stem. The value of KDF is primarily a function of the angle of the leaves to the incident radiation (which is why there is the distinction between planophile and erectophile canopies). Values of KDF ranged from 1.13 for cotton, down to 0.29 for Wimmera ryegrass. Both's value of 0.72 for lettuce is the same value as measured for orchardgrass. Both's value for lettuce is for the entire crop period, at the end of which the lettuce plant has a tightly clustered canopy. Because the baby spinach plants are grown at a high density, the leaves point upwards as they do not have the room to spread out. This condition makes the spinach more like the erectophiles with a low value for KDF. For this reason a value of 0.50, which is more similar to alfalfa, was selected for spinach.

The original SUCROS model, and Both (1995), use a value of 0.2 for the scattering coefficient of the leaves for PAR (SCP). Monteith (1969) states the proportions of radiation transmitted and reflected by crop leaves are usually similar. For this reason a value of 0.2 for SCP is also used for baby spinach.

To determine the amount of PAR available within the greenhouse for the leaves to use, a series of calculations was conducted. The weather data used by the model includes outdoor solar radiation. This data must be modified to take into account losses through the greenhouse glazing, and losses through shading. In addition the angle of the incident radiation is considered when calculating the amount and character of PAR (direct or diffuse) at different layers of the canopy. These calculations followed Both (1995), but were modified for hourly calculation.

The first step is to calculate the solar parameters for the entire day:

$$DEC = -\arcsin\left(\sin(0.40928) * \cos\left(2\pi \frac{DOY + 10}{365}\right)\right) \quad 5.1$$

where: DEC is the declination of the sun (radians)

DOY is the day of the year

And intermediate variables:

$$SINLD = \sin(LAT) * \sin(DEC) \quad 5.2$$

$$COSLD = \cos(LAT) * \cos(DEC) \quad 5.3$$

where: LAT is the latitude (radians), + if north.

The daylength (DAYL) in hours is given by equation 5.4:

$$DAYL = 12 + \frac{24}{\pi * \arcsin\left(\frac{SINLD}{COSLD}\right)} \quad 5.4$$

The daily integral of the sine of solar inclination (DSINB) in s day<sup>-1</sup> is:

$$DSINB = 3600 * \left( DAYL * SINLD + \frac{24 * COSLD * \sqrt{1 - \left(\frac{SINLD}{COSLD}\right)^2}}{\pi} \right) \quad 5.5$$

The daily integral of SINB with a correction for lower atmospheric transmission at lower solar elevations (DSINBE) in s day<sup>-1</sup> is:

$$\begin{aligned} DSINB \\ = 3600 \\ * \left( DAYL * (SINLD + 0.4 * (SINLD^2 + 0.5 * COSLD^2)) \right. \\ \left. + \frac{12 * COSLD * (2 + 3 * 0.4 * SINLD) * \sqrt{1 - \left(\frac{SINLD}{COSLD}\right)^2}}{\pi} \right) \end{aligned} \quad 5.6$$

The solar constant (SC) (W/m<sup>2</sup>) corrected for a varying earth-sun distance is:

$$SC = 1370 * \left( 1 + 0.033 * \cos \left( 2\pi * \frac{DOY}{365} \right) \right) \quad 5.7$$

These parameters need only be calculated daily. To determine the hourly PAR and the proportions of PAR which are diffuse and direct, a series of calculations is necessary:

The first step is to determine the proportions of PAR that are diffuse and direct. In the original SUCROS model and in Both, an average daily value for the fraction diffuse (FRDF) is used. However, because this model has an hourly basis, it is necessary to modify the procedure to estimate the fraction diffuse. By comparing the measured value of total radiation (contained in the weather data file), with the expected amount of solar radiation outside the atmosphere, it is possible to calculate this fraction. The first step is to calculate the daily integral outside the atmosphere (W m<sup>-2</sup>):

$$H_{day} = \left(\frac{24}{\pi}\right) SC \left[1 + \cos\left(\frac{2\pi DOY}{365}\right)\right] \cos(LAT) \cos(DEC) \sin(SHA) \quad 5.8$$

where: SHA is the Sunset Hour angle (radians)

The Sunset Hour angle is the same as the sunrise hour angle (only it is defined as being positive rather than negative) and can be determined from the formula:

$$SHA = -1 * \cos^{-1}(-\tan(LAT)\tan(DEC)) \quad 5.9$$

$H_{day}$  is the value for the entire day and must be scaled for the amount occurring during each hour. By assuming the solar radiation follows a sine curve between 0 and pi for the hours between sunrise and sunset it is possible to determine the hourly contribution by integrating over the specific hour. The contribution of each hour is then:

$$H_{hour} = H_{day} \frac{\left\{ \cos\left[\left(\frac{hour - sunrise}{sunset - sunrise}\right)\pi\right] - \cos\left[\left(\frac{hour + 1 + sunrise}{sunset - sunrise}\right)\pi\right] \right\}}{2} \quad 5.10$$

This calculation is only performed for the hours between sunrise and sunset when solar PAR is present.

The term  $K_t$  is defined as the ratio of actual daily radiation on a horizontal surface to the daily extraterrestrial radiation on a horizontal surface. For the purposes of determining the fraction of PAR that is diffuse on an hourly basis, it is assumed  $K_t$  can be calculated on an hourly basis rather than a daily one. By dividing the measured value of hourly total radiation HTR (from the weather file) by the expected hourly extraterrestrial radiation as determined above, it is possible to determine  $K_t$ .

$$K_t = \frac{HTR}{H_{hour}} \quad 5.11$$

With  $K_t$  it is then possible to compute the ratio of the average diffuse radiation to the average total radiation  $H_d/HTR$ :

$$\frac{H_d}{HTR} = 1.39 - 4.03K_t + 5.53(K_t)^2 - 3.11(K_t)^3 \quad 5.12$$

The remainder is the direct portion of PAR.

$$1 - H_d/HTR \quad 5.13$$

The amount of solar PAR (direct and diffuse) reaching the crop within the greenhouse (in  $\text{J m}^{-2} \text{ s}^{-1}$ ) is:

$$PAR = HTR * pcPAR * \tau \quad 5.14$$

where: pcPAR is the percentage of radiation that is in the PAR wavelengths (assumed to be a constant 0.55).

$\tau$  is the greenhouse transmittance assumed to be a constant 0.7.

HTR is hourly total radiation ( $\text{J m}^{-2} \text{ s}^{-1}$ )

The remaining procedures and equations used to calculate HTGA are identical to those used by Both, and the original SUCROS model, with the exception that since gross assimilation is calculated hourly, there is no need to use a five point Gaussian approximation to determine daily gross assimilation by calculating hourly gross assimilation at the five times of day specified by the Gaussian approximation method. The five point Gaussian approximation is, however, used to calculate the effects of PAR (diffuse and direct) on the canopy as used in the original models.

*Carbohydrate Production:*

CO<sub>2</sub> is converted to carbohydrate (CH<sub>2</sub>O) with a ratio of 30/44 (30 being the molecular weight of carbohydrate, and 44 the molecular weight of CO<sub>2</sub>). The hourly production of carbohydrate is then just the hourly gross crop CO<sub>2</sub> assimilation rate (HTGA) multiplied by the ratio 30/44.

*Maintenance:*

Part of the carbohydrate produced is required for maintaining the plant. The cells within the various plant structures respire and this respiration rate is a function of temperature. The original SUCROS model uses values of 0.03 (% of mass) for the leaves, and 0.015 (% of mass) for the roots and stems. Both (1995) also used these values for lettuce. These values are for daily maintenance and were scaled to an hourly percentage by dividing by 24.

*Dry Matter partitioning:*

Carbohydrate production not used for maintenance of the plant is available for plant growth. The SUCROS model assumes the fractions that describe the distribution are constant, or else vary as a function of the developmental stage. Because the spinach growth model is only considering early growth, the distribution factors are not varied with the crop age.

The excess carbohydrate is first partitioned between the roots and the shoots (FRT, and FSH). Further partitioning of the shoot portion occurs between the leaves and the stems (FLV, and FST). To simplify the model, this secondary partitioning was not used because the entire shoot is harvested, not just the leaf.

To determine the fraction of roots, data from the growth experiments was used. Observed values at initial floating and at final harvest remained relatively constant 0.162 (stdev 0.03) at floating and 0.163 (stdev 0.4) at harvest. A constant value of 0.16 was assumed for all stages of the crop.

*Growth of Plant organs and Translocation:*

Once the produced carbohydrates have been partitioned into the various plant structures, it is possible to estimate the change in dry weight. Different structures

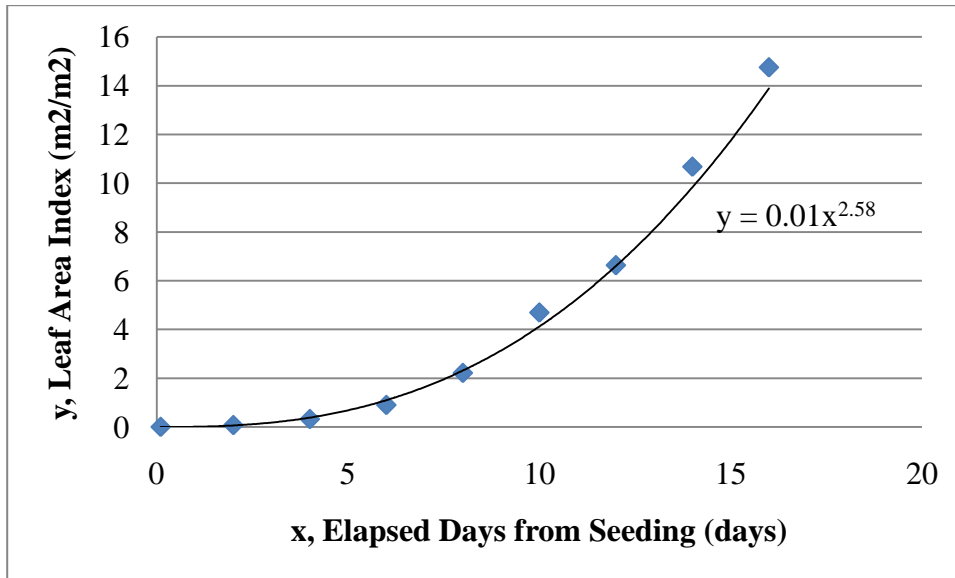
have different conversion factors since leaves, stems and storage organs have different requirements. As baby leaf spinach is an immature plant with negligible stems, we can consider values for just the leaves and roots. Typical values for leaves are 1.463 kg CH<sub>2</sub>O required to produce 1 kg dry matter, and 1.444 kg CH<sub>2</sub>O for the roots (Goudriaan and van Laar, 1994).

Another simplification that arises due to using a very young crop is we can ignore translocation from organ to organ which occurs as a plant matures and moves resources from older, shaded leaves and stems into fruits/other leaves.

#### *Leaf Development:*

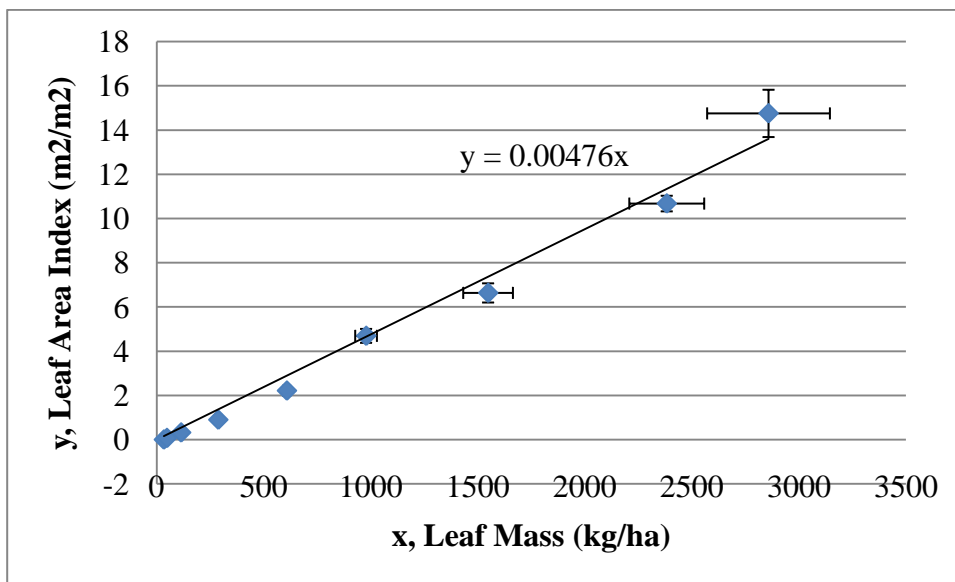
In the original SUCROS model, Goudriaan and van Laar considered the initial phase of leaf growth to be exponential and temperature limited. Assimilates are not considered limiting as stores from the seed supplement photosynthesis in the cotyledons and leaves. According to Goudriaan and van Laar this phase lasts until a leaf area index (LAI) of 0.75 is reached. From the spinach growth experiment a rapid and non-linear increase in the leaf area of the plants was also observed (Figure 5.1).

The peak LAI reached during the 16 day growing cycle was approximately 15 which is more than the cutoff of 0.75 suggested by the SUCROS model (and in fact at a level where senescence of lower leaves would normally occur). But the baby spinach production system is not typical of normal plant growth since the plants are placed at a density of approximately 1200 per m<sup>2</sup> and are not respaced. Because the crop is to be harvested as a baby product the high leaf area indexes are tolerated.



**Figure 5.1** Leaf Area Index during spinach crop cycle (1200 plants m<sup>-2</sup>)

Rather than use a power type growth equation for leaf development, it was found that a simple linear relationship between the mass of the leaves and the Leaf Area Index provided a good fit to the data (Figure 5.2).



**Figure 5.2** Leaf Area Index as a function of leaf mass.



Forcing the intercept of the Leaf Area Index through the origin, the formula relating Leaf Area Index (LAI, m<sup>2</sup> m<sup>-2</sup>) to leaf mass (WLV, kg ha<sup>-1</sup>) is:

$$LAI = A * WLV \quad 5.15$$

where A = 0.00476.

#### *Dry matter production:*

To determine the total dry weight of each plant organ (and the entire plant as well), the individual growth rates are multiplied by the integration period (timestep, 1 hour) and then added to their previous dry weights of plant organs. As done previously, the methods of the original SUCROS, and Both, models were followed with the exception that calculations were hourly rather than daily. During times of day when no photosynthesis is occurring, a loss of biomass occurs as maintenance costs are a constant and there is no production of photosynthate to overcome these losses.

#### 5.3.3 Implementation of the modified SUCROS model:

The SUCROS model was implemented as a series of functions programmed in Java. For each hour of the simulation, the program would follow the same algorithmic steps as the original SUCROS model.

The controlled environment setpoints of the experiment (air temperature and target light integrals) were put into the greenhouse simulation model and the simulation run. Data from the greenhouse simulation model, such as estimated temperatures and light levels (both solar and supplemental) within the greenhouse, were saved in data files which provided the input to the crop growth model. Hourly values of leaf biomass and Leaf Area Index were saved in text files for comparison with measured data.

A listing of the code used is in Appendix A.

#### 5.3.4 Validation of the spinach growth model:

A number of tests were performed to determine the validity of the spinach growth model. The first test was a direct comparison of the calculated biomass accumulation with the measured values. Leaf area was not directly compared as the simulated leaf area is a function of the biomass. This test was performed for the conditions of the experiment and was also run for daily target integrals of 5 to 25 mols  $\text{m}^{-2}$  (in increments of 5 mols). The second comparison was performed to determine if the growth of the crop was directly proportional to the amount of light it received, as suggested by Both et al. (1996). The range of 5 to 25 mols  $\text{m}^{-2}$  was selected as what could be expected within a greenhouse with no supplemental lighting or shade control.

To test the sensitivity of the model to the values of particular parameters, each of the parameters was varied plus and minus 10% of the original value, and the model run (with the original values for the remaining parameters). The final biomass accumulation at harvest as predicted by the model was recorded. The variability in the final biomass accumulation for each parameter was sorted in a descending manner (from greatest to least variability) and results plotted in a tornado style plot, to illustrate which parameters have the greatest effect on the model.

### **5.4 RESULTS:**

#### 5.4.1 Spinach growth experiments:

The crop was successfully grown for the duration of the experiment with no evidence of root disease or other factors that could potentially negatively affect the results. Harvest data including leaf area and fresh and dry weights, in addition to photosynthetically active radiation (PAR) totals are summarized in Table 5.2.

**Table 5.2** Individual fresh weight, dry weight, leaf area and cumulated PAR totals at each harvest day during spinach growth experiment.

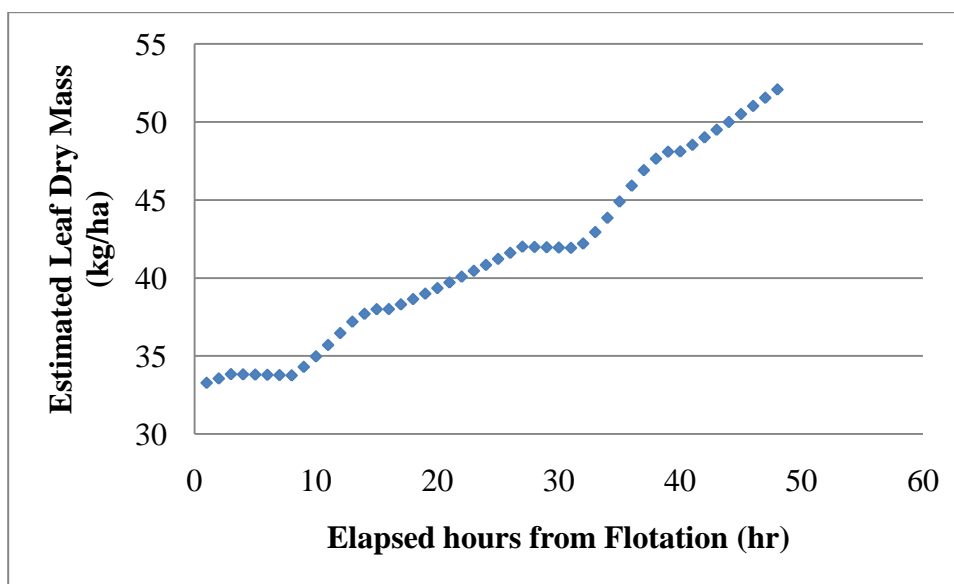
Days elapsed from flotation (d)	Individual Plant	n	Average	Standard Deviation	PAR Accumulation (mol m <sup>-2</sup> )
2	Fresh Weight (g)	24	0.027	0.007	38.5
	Dry Weight (g)	24	0.003	0.002	
	Leaf Area (cm <sup>2</sup> )	24	0.6	0.2	
4	Fresh Weight (g)	24	0.104	0.023	78.4
	Dry Weight (g)	24	0.008	0.003	
	Leaf Area (cm <sup>2</sup> )	24	2.7	0.9	
6	Fresh Weight (g)	25	0.296	0.089	116
	Dry Weight (g)	25	0.019	0.005	
	Leaf Area (cm <sup>2</sup> )	25	7.5	2.5	
8	Fresh Weight (g)	30	0.713	0.099	154
	Dry Weight (g)	30	0.041	0.006	
	Leaf Area (cm <sup>2</sup> )	9	18.4	1.6	
10	Fresh Weight (g)	27	1.607	0.270	192.7
	Dry Weight (g)	21	0.065	0.015	
	Leaf Area (cm <sup>2</sup> )	8	39.1	7.4	
12	Fresh Weight (g)	26	2.352	0.578	230.4
	Dry Weight (g)	17	0.103	0.032	
	Leaf Area (cm <sup>2</sup> )	17	55.3	14.9	
14	Fresh Weight (g)	24	3.712	0.985	269.3
	Dry Weight (g)	16	0.159	0.047	
	Leaf Area (cm <sup>2</sup> )	13	89.0	10.6	
16	Fresh Weight (g)	49	4.336	1.745	308.7
	Dry Weight (g)	28	0.191	0.101	
	Leaf Area (cm <sup>2</sup> )	10	122.9	28.2	

#### 5.4.2 Validation of the SUCROS baby spinach growth model

Measurements of leaf area and plant biomass from the growth experiments were collected based on individual plant measurements which were then extrapolated based on plant density to a crop sized hectare basis to be compared to the results from the adapted SUCROS model. Because SUCROS was originally developed for use in

predicting crop growth in field production, many of its parameters have units based on the hectare. These parameter values can be scaled to a greenhouse typical unit of measure such as square meter; however, it was decided to leave them in the original form to facilitate comparison to values in the literature. In addition, it is an easier task to convert the final biomass production from a hectare basis to a square meter or per plant basis, than to modify each of the individual parameters to achieve the same result.

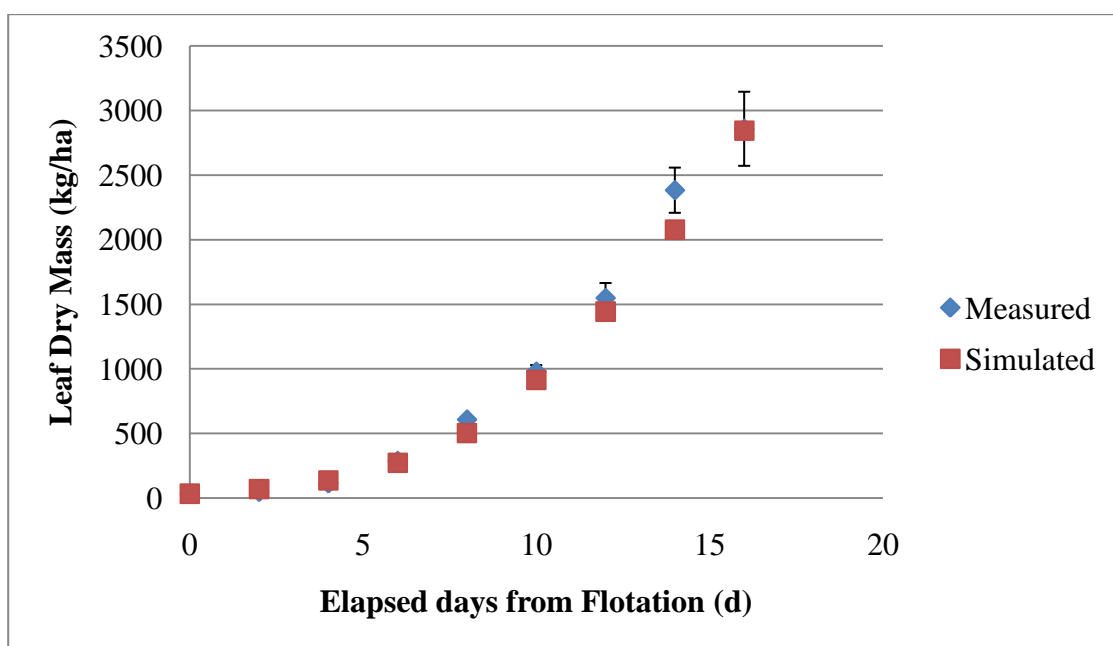
Figure 5.3 shows the simulated growth of the crop over the first 48 hours of the experiment (assuming light control was provided through LASSI rules to a daily target of 19.2 mols). In this plot it is clear that growth of the leaves occurs only during periods of light (which provides the necessary driving force for photosynthate production). The initial steep growth from hour 9 to 15 represents growth during the day, under natural light.



**Figure 5.3** Simulated Leaf Dry Mass Growth over first 48 hours with LASSI controlled lighting to a daily integral of 19.2 mols m<sup>-2</sup>.

After sunset supplemental lighting is applied to meet the target light requirement; however, this light is applied at a lower rate than natural lighting (approximately  $180 \mu\text{mol m}^{-2} \text{s}^{-1}$ ). This period is followed by a period of decrease in the mass of leaves which represents a time when respiration is consuming photosynthate and none is being produced.

Figure 5.4 is a plot of both the simulated growth and the actual measured growth from Pond two (Pond one data was used to generate the parameters used in the model that were not determined from the literature such as relating plant biomass to the leaf area).



**Figure 5.4** Measured and simulated leaf dry mass over the course of the crop cycle. Grown with  $19.2 \text{ mol m}^{-2} \text{ day}^{-1}$  PAR.

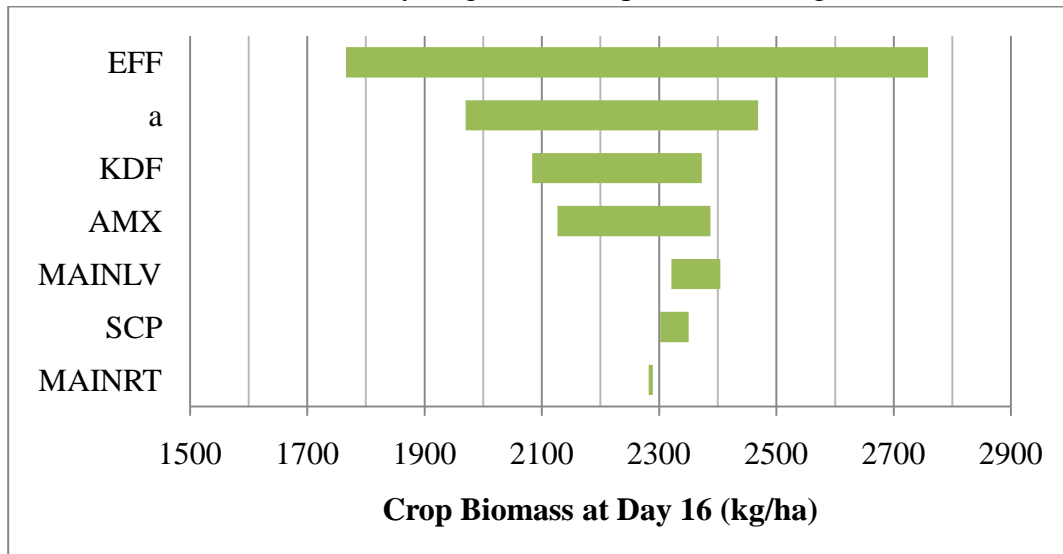
The model was developed based on growth experiments conducted under a single daily light integral of 19.2 mols. To determine whether the model scales production to fit the amount of light received, the model was run with daily light targets from 5 to 25 Mols. The results are displayed in Table 5.3.

**Table 5.3** Predicted biomass as a function of daily light integral compared to the baseline 19.2 mols with a production of 2565 kg/Ha at 16 days

DLI (mol m <sup>-2</sup> )	Biomass (kg/Ha)	Expected (DLI/19.2)	Actual (Biomass/2565)
5	592	0.26	0.23
10	1302	0.52	0.51
15	1987	0.78	0.77
20	2682	1.04	1.05
25	3416	1.30	1.33

The model appears to scale very well to the amount of light received by the crop with a very slight underestimation at low daily integrals and a slight overestimation at high daily integrals. This verifies the expected linear relationship between crop growth and light intercepted by the crop, with a range typical of what might be found in a greenhouse.

To test the sensitivity of the model to the values selected for each parameter the following parameters were varied  $\pm 10\%$  and the simulated biomass accumulation on harvest day 16 recorded: AMX, EFF, KDF, SCP, A, MAINLV, and MAINRT. The results were tabulated, sorted by magnitude and presented in Figure 5.5.



**Figure 5.5** Tornado plot of the effect of varying model parameters  $\pm 10\%$  on the simulated crop biomass (kg/ha) at harvest day 16.

Variables relating to the amount of PAR and the character of the PAR were not selected for sensitivity testing as they relate to light, to which the model has been shown to behave in a linear fashion. Though the values of the parameters such as the transmittance of the greenhouse and the shade curtain, and the percentage of PAR in solar radiation are estimates, their variability affects the model in a predictable and linear fashion.

## **5.5 DISCUSSION:**

The SUCROS growth model is a framework that has been used by researchers to model many different crops. Typically the crops modeled are field crops grown under natural outdoor conditions. Both (1995) modified this model for greenhouse production, by adding in the effects of supplemental CO<sub>2</sub> and lighting, as well as the effect of the greenhouse structure on the quantity and quality (diffuse/direct ratio) of light. However, as in the original SUCROS model, Both was interested in a mature crop and so the time step he used was one day. To model a fast growing short cycle crop like hydroponic baby spinach, and to be responsive to the hourly time step of light control, it was felt a time step of one hour would perform better. The primary adaptation of the original SUCROS model, and Both's version, was to develop the equations necessary to determine the ratio of diffuse and direct PAR in the greenhouse. To calculate this ratio a technique was developed (Albright, personal communication) by which the extrasolar radiation for the hour could be determined by taking the daily value and scaling it using a sine curve for the daylight hours. Using hourly measurements it was then possible to use the same equations as used for daily determination of the diffuse/direct ratio. This technique proved successful and was ultimately used in the model. Alternatively the individual daily values could have been calculated using the original technique of the SUCROS model and then the same

value used for all hours of the day, but this would potentially overestimate the direct component in the morning and evening and underestimate it during the middle of the day. The remaining changes to the SUCROS model were simply a matter of directly scaling parameter values from daily to hourly. The techniques and equations used remained the same.

Being a mechanistic model, the backbone of SUCROS is its parameters. Rather than rely on regression modeling of large data sets (that would be difficult to obtain for all the conditions of interest) SUCROS uses an understanding of the processes behind crop growth and development coupled with parameter values that can either be measured or estimated. The variables used in SUCROS are not solely for use in SUCROS, rather they are parameters that have been developed over the years by horticulturalists to better understand and compare how different crops develop and grow. Unfortunately, the parameter values needed for this particular growth model were not directly available from the literature, required an understanding of how they are derived and how the values would change for a crop harvested before it is fully mature. This selection and adaptation of parameter values is not ideal, but the alternative of directly measuring values from baby spinach crops would require special apparatus and measurement techniques, the benefits of which would be questionable. Especially since the values would then be used in a relatively imprecise model. In addition, the purpose of developing this model is to facilitate comparisons between different growing conditions. The accuracy of the end result is less important than the relative response of the model to differing input.

The parameter developed from observed growth, namely, the relation between the biomass of the crop and leaf area index, maintained a clear linear relationship through all ages of the crop. In the original SUCROS model the development stage of the crop is important, because the characteristics of the plant growth change as the



crop matures. For this reason the original SUCROS model uses a more complicated model to relate biomass to leaf area index and is a function of the development stage. By focusing on a baby leaf crop, it was possible to simplify the original SUCROS model and use the observed linear relationship.

As a part of validating the model, biomass growth data from a parallel growing experiment was compared to the simulated biomass growth. The model has a tendency to overestimate growth through most of the crop cycle, but underestimate production in the final few days. This error is most likely due to the values of the parameters that were determined from the literature, which were taken from other crops, grown under quite different conditions. The conditions of hydroponic production are quite different from those encountered in field production. Growth rates higher than those measured in the field are expected in hydroponics, where conditions are optimally controlled. An ample supply of water and nutrients, coupled with temperature and light control, reduce the stress on a crop and allow it to grow closer to its theoretical optimum.

The scalability of the model due to light is an important property that is perhaps more valuable than the absolute accuracy of the model. Differences in variations of spinach varieties could potentially have different responses and parameter values, resulting in different final harvest weights. However, the ultimate purpose of this model is to simulate production under varying conditions with the primary variable being light. Fortunately, this model mimics the linear response to light integral that has been observed in spinach production.

There are, however, several limits to the use of this model that could potentially appear in greenhouse production. The model was developed based on a single plant spacing of approximately 1200 plants  $\text{m}^{-2}$  coupled with a harvest at day 16. This spacing was found during previous spinach work (Albright et al., 2005) to be

optimal for baby spinach production. Closely related to the spacing is the selection of the harvest time. At the Cornell university greenhouses, light levels have typically been controlled to between 15 and 20 mols m<sup>-2</sup> per day. These light levels are based on previous experience with other crops, the Ithaca, NY climate, and the cost of supplemental lighting. With this amount of lighting (and a density of 1200 plants m<sup>-2</sup>) it was found that harvesting from 12 to 16 days after floatation produced a good crop. Extrapolating to different spacings would be difficult as this would potentially change the grow rates of the crop through more or less competition for light. The model was also not developed for extremes of temperature and humidity. Usually, extremes of temperature are not a problem in a greenhouse as long as the temperature control equipment in place, such as heaters, shades and evaporative coolers, are functioning properly. Some protection from temperature extremes is due to the pond itself, acting as a temperature buffer. High humidity conditions can be encountered in the winter months when venting is restricted to conserve energy; however, this parameter is usually monitored by the control system, and not permitted to reach levels that would seriously impact crop growth.

## CHAPTER 6: *PYTHIUM APHANIDERMATUM* GROWTH AND TRANSMISSION MODEL

### 6.1 INTRODUCTION:

In the original description of the SUCROS plant growth model, a major assumption was the model is for use in conditions where the crop is not limited by water and nutrients, and not hindered by the presence of pests or disease (Goudriaan and van Laar, 1994). This condition is not met for the case of modeling the growth of a crop in the presence of *Pythium aphanidermatum*, a water borne root mold which spreads rapidly through the nutrient solution and attacks plant roots. However, SUCROS is well suited for modification to model the disease interaction, as its mechanistic form separates individual growth of root and shoot due to photosynthate production.

In her doctoral dissertation entitled “Influence of Plant Age, Inoculum Dosage, and Nutrient Solution Temperature on the Development of *Pythium aphanidermatum* in hydroponic Spinach (*Spinacia Oleracea* L.) Production Systems”, Katzman (2003) documented much of the data necessary to expand the SUCROS growth model to account for *Pythium* processes. Reported data include the effects of initial zoospore concentration in the nutrient solution, time of inoculation and nutrient solution temperature on root and shoot damage, zoospore concentrations in solution, and mycelium development. Her data coupled with experiments performed during the NYSERDA spinach disease project (Albright et al., 2007), forms the basis of the *Pythium* growth and transmission model and modification of the SUCROS model. The developed model was validated by comparing predicted multi-crop cohort growth and development under disease conditions, to those observed during the NYSERDA spinach disease project. Continuous production experiments with multiple aged crops in the same pond were carried out and a part of these studies involved inoculating the

multi-cohort system with known concentrations of zoospores and measuring the growth of the cohorts as the disease progressed. Also during these experiments, an uninoculated control pond became infected. Data from this condition was used to help quantify the development of an infection under natural conditions, as would be most likely to occur in a commercial setting.

The objectives of this chapter are to:

1. develop the framework and parameters for a model to predict the growth and spread of *Pythium aphanidermatum* in hydroponic baby spinach production under controlled environment conditions,
2. modify the SUCROS based baby spinach crop growth model to take into account the damage done to the crop's roots due to *Pythium aphanidermatum* zoospores and mycelium,
3. validate the functionality of the model by comparing predicted crop growth and *Pythium* disease damage to experimentally collected data.

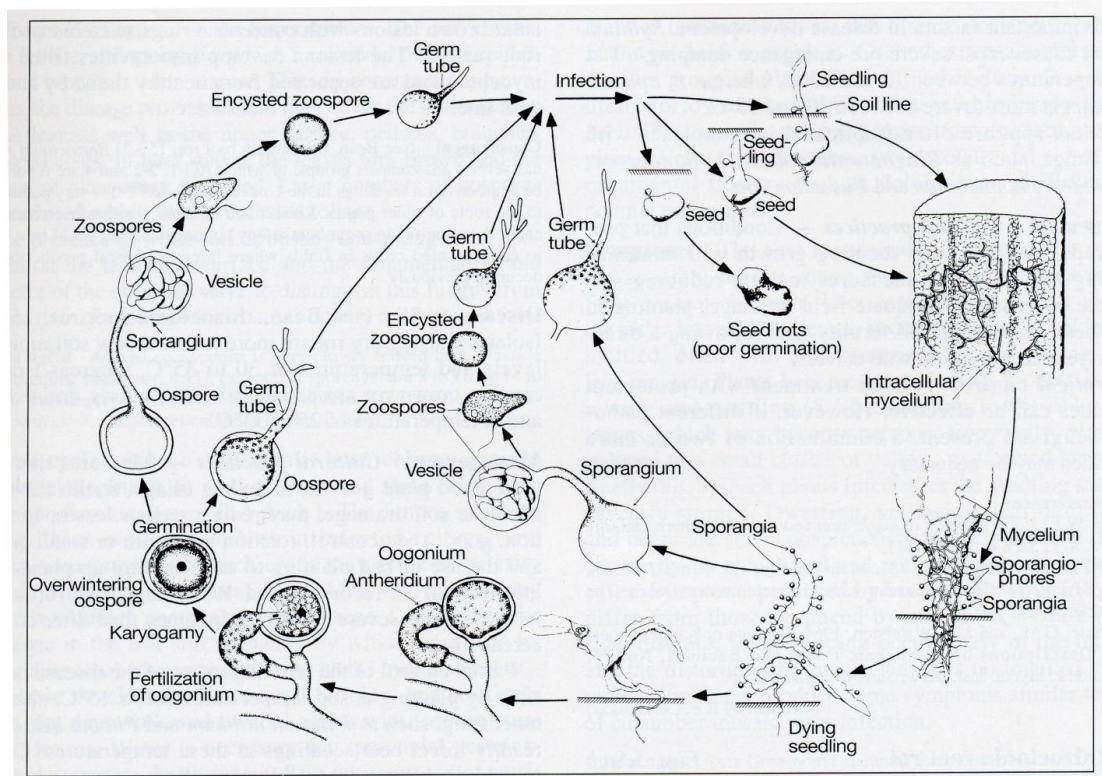
## **6.2 LITERATURE REVIEW:**

### **6.2.1 *Pythium aphanidermatum* lifecycle:**

*Pythium* species are a widespread and major pathogen that affect many different crops in both field and hydroponic culture and have been identified as one of the most common and destructive pathogens found in hydroponic systems (Stanghellini and Rasmussen, 1994). *Pythium aphanidermatum* was also identified as the causative agent of the root disease that plagued earlier research into hydroponic spinach production at Cornell (Katzman 2003).

*Pythium* species can cause seed rot, seedling damping off, and root rot of most plants (Agrios 1978), however, spinach is particularly susceptible (Howard et al.

1994). As described in Agrios, (1978) *Pythium* is a type of Peronosporales, which in turn, is a type of Oomycetes. This fungus has elongated mycelium, produces oospores as resting spores and zoospores or zoosporangia as asexual spores. The life cycle of a typical *Pythium* species is illustrated in Figure 6.1.



**Figure 6.1** Disease cycle of *Pythium* sp. (Agrios 1978).

The primary mode of infection of roots is through zoospores, which possess flagella and allow them to move through continuous films of water. Shortly after release, zoospores lose their flagella, encyst and grow a germ tube, which allows them to infect roots. Spore germ tubes or saprophytic mycelium, come in contact with the tissue of the host plant either by chance, or because exudates from the plant act as a chemotropic stimulant, which causes zoospores to move to, or mycelia to grow towards the plants. It is this mobility which exacerbates the problem of *Pythium* in

hydroponic production. Because the crop is grown in a common nutrient solution (whether the production system is Nutrient Film Technique or pond) zoospores quickly disperse through the system infecting roots throughout the entire crop.

Once the germ tubes or saprophytic mycelium come in contact with root material the fungus enters either through direct penetration or cracks, and further penetrates the tissue through mechanical pressure and enzymatic dissolution. Breakdown of the tissue occurs as the fungus grows both through and around the cells and cellulolytic enzymes digest the cell walls. The fungus consumes the plant cell substances, using them as both a source of energy and as raw material for its own growth. Infected tissue decays until only a frail remnant of indigestible material is left behind with the mycelium.

As infection develops and mycelium spreads through the root, the reproductive stages of *Pythium* begin to grow and form sporangia and oospores. Sporangia are the bodies that release zoospores, whereas, oospores are a hardier form of spore which tolerate both high and low temperatures as well as low moisture conditions. In field production oospores allow *Pythium* to survive harsh winter conditions where there may be limited plant material available, and temperatures are too cold for the survival of the mycelium. When conditions are amenable again, oospores develop a sporangium independent of plant material, and release zoospores. However, this process is considerably slower than direct production of sporangia from mycelium. However, the oospore stage of *Pythium* is still an important consideration in hydroponic production. Oospores present a means of infecting the hydroponic system via medium (Katzman 2003) and make cleaning up of a hydroponic system difficult. With the exception of oospores, the various *Pythium* stages are relatively fragile and can be inactivated through a number of commercially available technologies (as detailed in chapter two).

### 6.2.2 *Pythium aphanidermatum* in hydroponic systems:

For her dissertation, Katzman (2003) examined a number of factors necessary to model the impact of a *Pythium* outbreak on a crop. She examined sources of inoculum, influence of plant age at time of inoculation on disease development, relationships between inoculum dosage and disease development, and influence of nutrient solution temperature on plant growth and disease development. Though the focus of Katzman's work was on more mature spinach plants (as opposed to the baby leaf spinach of this research) many of her findings are still applicable. The most important finding of Katzman's research is that lethality of *Pythium* is strongly related to temperature. By examining spinach crops grown under identical conditions, but with nutrient solution temperatures of 18C, 24C and 30C respectively, and inoculated with *Pythium* at 25 zoos ml<sup>-1</sup>, Katzman found crops grown at 18C did not differ significantly from the uninoculated control crops grown at the same temperature. She also found crops grown at 24C and 30C suffered a 20% and 70% reduction in shoot dry mass, respectively. This follows other work, such as Bolton (1980), which suggests *Pythium aphanidermatum* has an optimal temperature of 30C. Bates and Stanghellini (1984) found when nutrient solution temperatures were above 23 C, *Pythium aphanidermatum* was the dominant oomycete (as opposed to *Pythium dissotocum* whose range was 17 to 22 C). This finding has important implications for hydroponic spinach production. By reducing the temperature of the nutrient solution, the time required for the mycelia to develop the sporangia necessary to further spread the infection is increased. This, in turn, may allow the removal of the root material before the release of zoospores occurs, provided the crop is grown with a short crop cycle. Removal before zoospore release prevents new material from becoming infected, and stops the infective cycle.

## 6.3 METHODS AND MATERIALS:

### 6.3.1 Disease model Framework

The data for the development of the spinach disease model comes primarily from Katzman (2003). Throughout her series of experiments on age of plant, inoculum concentration, and temperature effects on disease development, Katzman always categorized the condition of the roots. Though the categorization of the roots into classes was somewhat subjective, the fact that it was carried out consistently in all of her experiments provides a common method of comparison, and allows the effects she investigated to be integrated into a common model. A summary of the symbols and variables used is presented in Table 6.1.

**Table 6.1** Summary of variables and symbols used in the *Pythium aphanidermatum* growth and transmission model.

$\alpha_{\text{conc}}$	Concentration effect intercept
$\alpha_{\text{grow}}$	Mycelial growth rate intercept
$\alpha_{\text{temp}}$	Temperature effect intercept
$\beta_{\text{conc}}$	Concentration effect slope
$\beta_{\text{grow}}$	Mycelial growth rate slope
$\beta_{\text{temp}}$	Temperature effect slope
$\delta_{\text{root}}$	fraction of the root that is dark brown/black
$\epsilon_{\text{mycelium}}$	Reference amount of mycelium @ 25 zoos ml <sup>-1</sup> (kg ha <sup>-1</sup> )
$\eta_{\text{photo}}$	Coefficient of Photosynthetic Rate reduction
$A_{\text{mature}}$	Maturation age of mycelium (hr)
$A_{\text{release}}$	Zoospore release age (hr)



**Table 6.1 continued**

c	Cohort number
E(C)	Concentration adjustment factor
E(T)	Temperature adjustment factor
GPHOT	Gross photosynthetic production ( $\text{kg ha}^{-1}$ )
GPHOT <sub>da</sub>	Gross photosynthetic production, adjusted for disease ( $\text{kg ha}^{-1}$ )
gr	Mycelium growth rate ( $\text{kg kg}^{-1} \text{ hr}^{-1}$ )
gr <sub>b</sub>	Base growth rate at 24 C ( $\text{kg kg}^{-1} \text{ hr}^{-1}$ )
gr <sub>p</sub>	Mycelium growth rate on a per plant basis ( $\text{g plant}^{-1}$ )
m <sub>f</sub>	Final mass of mycelium infected root ( $\text{kg ha}^{-1}$ )
m <sub>0</sub>	Starting Mass of mycelium ( $\text{kg ha}^{-1}$ )
m <sub>c</sub>	Mass of zoospore releasing mycelium for each cohort c, (kg)
m <sub>equiv</sub>	Equivalent mass of mycelium added due to zoospores in solution (kg)
n	Number of periods over which growth is accrued
n <sub>zoos</sub>	Zoospore release amount ( $\text{zoos kg}^{-1}$ )
PRAD	Adjustment to gross photosynthetic production as a function of root damage.
T	Temperature (C)
vol	Volume of nutrient solution (ml)
URM <sub>avg</sub>	Average mass of root that is uninfected ( $\text{g plant}^{-1}$ )
URM <sub>avg,kg ha-1</sub>	Average amount of infected root ( $\text{kg ha}^{-1}$ )
z <sub>c</sub>	Number of zoospores released by each particular cohort (zoos)
Z <sub>conc</sub>	Concentration of zoospores in solution ( $\text{zoos ml}^{-1}$ )

The basis of the spinach disease model is the growth of mycelium within, or on the roots of the crop. This process is tracked for each cohort of the crop on an hourly basis. Each hour the amount of new mycelium growth on each cohort is stored in an array, which is the size of the age of the oldest mycelium material on the cohort, in hours. In addition to growth of existing mycelium, zoospores in solution attack available healthy root material, and the mass of mycelium created is a function of their concentration in solution.

$M_{j,k}$  is the mycelium array for each cohort

where:  $j, (1,2,\dots,j)$  is the age of the crop cohort in hours  
 $k, (1,2)$   $k = 1$  is the temperature adjusted age of the mycelium (hr)  
 $k = 2$  is the total mass of mycelium of this age (kg)

Equation 6.1 provides us with a means of calculating mycelium mass for the current hour.

$$M_{1,2} = \sum_2^j M_{j,2} * gr + m_{equiv}, \quad 6.1$$

All of the mycelium is aged by a temperature adjusted amount to account for the different developmental and growth rates of mycelium at varying temperature. (The mycelium, in addition to growing faster, also matures faster with increasing temperature).

The growth rate is adjusted for temperature in equation 6.2.

$$gr = gr_b * E(T) \quad 6.2$$

The developmental age of the mycelium is calculated in equation 6.3.

$$M_{j,1} = M_{j-1,1} + (1hr) * E(T) \quad 6.3$$

Based on the age of the mycelium, it is classified into one of two categories following Katzman (the third root category is white which indicates healthy root). The mycelium starts out as light brown or gray streaking in the roots until reaching age  $A_{\text{mature}}$  when the mycelium turns a darker brown/black color. This stage is later used to provide a measure of the damage to the root function and the shoot quality.

Each hour the mass of mycelium that is greater than or equal to the zoospore releasing age, ( $A_{\text{release}}$ ) releases zoospores which propagate the infection. The total number of zoospores released is calculated with equation 6.4.

$$z_c = m_c * n_{\text{zoos}} \quad 6.4$$

The number of zoospores released by the various cohorts of the crop are then summed and divided by the volume of the nutrient solution of the particular pond, to give the concentration of free zoospores in solution in the next hour (equation 6.5).

$$Z_{\text{conc}} = \frac{\sum_1^c z_c}{\text{vol}} \quad 6.5$$

These free zoospores then propagate the infection (provided there is uninfected root material present). The zoospores infect root material and form a mass of mycelium which is a function of the concentration of the zoospores in the solution (the higher the concentration of zoospores in solution, the larger the equivalent mycelium mass formed on the roots). This equivalent mass is determined with equation 6.6.

$$m_{\text{equiv}} = \varepsilon_{\text{mycelium}} * E(C) \quad 6.6$$

Damage to the growth of the crop is a function of the level of damage to the roots. Reduced crop growth is modeled by reducing the amount of photosynthate produced, described in equation 6.7.

$$GPHOT_{da} = GPHOT * PRAD \quad 6.7$$

The adjustment factor for the gross photosynthetic production is determined as a function of the percentage of the roots that are infected, in equation 6.8.

$$\text{PRAD} = 1 - \eta_{\text{photo}} * \delta_{\text{root}} \quad 6.8$$

Shoot quality is a function of the level of damage to the roots as well. However, rather than an absolute number, the quality is divided into two categories, saleable and unsaleable. When the level of root damage reaches a certain point (a function of the percentage of roots that are light brown/streaked and dark brown/black) the cohort is classified as unsaleable. The infection is initiated by assuming a specific amount of mycelium is present on new cohorts, representing natural infection due to oospores, and residual zoospores/mycelium.

The following sections further detail and expand on the processes outlined above and discuss how parameter values were estimated. A complete listing of the computer code developed is in Appendix A.

### *Growth of Mycelium*

The growth of *Pythium aphanidermatum* mycelium is the basis of the disease model. This process is linked to the damage caused to the roots, which in turn, affects the shoot growth and quality. As discussed previously, after infection with a zoospore, mycelium begins to spread through the root, consuming it as it goes. The mycelium spreads both up and down the root as much as 21 to 24 mm day<sup>-1</sup> (Guo and Ko 1993). Because the disease model developed for baby spinach is concerned with whole crop and cohorts within crop relations, infections at the individual root are not directly considered. This permits the simplification of the mycelium growth to a relative growth rate, based on the amount of mycelium already present, and the availability of healthy root material to infect.

As a part of her quantification of the extent to which spinach is damaged by *Pythium* infection, Katzman (2003) developed a “root rot rating” system and documented the state of the roots throughout her experiment. The “root rot rating” system is a categorization of the percentage of the roots falling into three categories based on physical appearance: white, light brown/streaked with gray, and dark brown/black. White roots were healthy root material. Light brown/streaked with gray roots were infected with growing mycelium and range from newly infected material to a more developed infection. The final stage of dark brown/black roots was the point where roots were heavily damaged/dead.

To estimate a rate at which mycelium grows and spreads through a root system, data from Katzman’s chapter entitled, “Influence of Plant Age at Time of Inoculation on Development of Disease Caused by *Pythium aphanidermatum* in Hydroponic Spinach Production “, was used. Of particular interest are her day 1 and day 9 (days following sowing) inoculations coupled with her day 9 and day 14 harvests (which correspond to times within the range of the usual 16 day baby spinach crop cycle. Katzman was not focused on baby spinach and so also conducted inoculations on days 14 and 21, and harvests on days 21 and 28 which are not of direct use for this model). The relevant data from Katzman has been summarized in Table 6.2 and Table 6.3. Table 6.2 is a summary of the effect of timing of *Pythium* inoculation on shoot dry matter, and Table 6.3, is a summary of the condition of the roots at the associated harvest times.

**Table 6.2** Influence of time of inoculation on shoot dry mass of spinach. Means and standard errors of shoot dry mass of plants inoculated with *P. aphanidermatum* zoospores on different days (1, 9, or 14) after sowing and harvested 9, 14, and 21 days after sowing. (From Katzman)

Inoculation day (D)	Mean shoot dry mass (g/plant) and standard error on harvest day		
	Harvest 9	Harvest 14	Harvest 21
Control	0.01 ± 0.00	0.051 ± 0.003	0.268 ± 0.014
Day-1	0.00 ± 0.00	0.008 ± 0.002	0.032 ± 0.012
Day-9	0.01 ± 0.00	0.061 ± 0.013	0.213 ± 0.051
Day-14	0.01 ± 0.00	0.051 ± 0.003	0.252 ± 0.017

**Table 6.3** Percentages (%) of roots within each root rot category (1-4). Spinach plants were inoculated with *P. aphanidermatum* zoospores on different days (1, 9, or 14) after sowing and harvested 9, 14, and 21 days after sowing. (From Katzman)

Treatment	Percentage (%) of roots within root rot rating on harvest day			
Inoculation day (D)	Root rot rating	Harvest 9	Harvest 14	Harvest 21
Control	1- white	100	100	96.7
	2 - light brown/streaked	0	0	3.3
	3 - dark brown/black	0	0	0
	4 – no root	0	0	0
Day-1	1- white	10.3	12.5	20
	2 - light brown/streaked	56.4	10	7.5
	3 - dark brown/black	25.6	62.5	57.5
	4 – no root	7.7	15	15
Day-9	1- white	100	90	5.1
	2 - light brown/streaked	0	10	76.3
	3 - dark brown/black	0	0	18.6
	4 – no root	0	0	0
Day-14	1- white	100	100	60
	2 - light brown/streaked	0	0	36.7
	3 - dark brown/black	0	0	3.3
	4 – no root	0	0	0

In developing a mycelium growth rate it was necessary to make several assumptions:

1. root damage is due to *Pythium* only,
2. root damage is uniform throughout the cohort,
3. mycelium development is visible in the root as a light brown discoloration and/or streaking after which it develops into a dark brown/black color,
4. root mass is directly proportional to shoot mass even when the roots are infected with mycelium, and follows a production rate as described in the SUCROS based spinach growth model,
5. the mycelium growth rate is only a function of available healthy root for infection,
6. the ageing of the mycelium for determining developmental stage is only a function of temperature,
7. *Pythium* zoospores infect the crop in a single hour. They are released and do not carry over hour to hour,
8. *Pythium* zoospores, when released, are uniformly spread throughout the pond.

Other organisms and mechanical damage can cause browning of the roots to some extent. This additional damage complicates the use of root damage as a measure of the level of mycelium present. However, to simplify the model it was assumed these contributions are minor. In Katzman's uninoculated control condition, after 21 days 3% of the root mass showed some damage, though it was not present in previous harvests on days 9 and 14. (This small amount of damage was thought to be due to unintentional *Pythium* infection).

Another simplification is to assume root damage caused by *Pythium* is uniformly spread throughout the cohort. This is not always the case when plants are exposed to high levels of *Pythium* at a young age when they are particularly vulnerable and liable to die outright. Moreover, it might happen in the case of an

initial infection, where only a single oospore may infect a single root of a single plant, which subsequently blossoms into a wider infection. However, since commercial interest is not in individual plants, but in the crop as a whole, averaging the damage across the entire crop is one way of accounting for this variation. Missing plants due to death are offset by surviving plants having less competition for light and space.

In her dissertation, Katzman categorized the conditions of the roots. An assumption is these root conditions also represent stages in the development of mycelium, and infected root material first progresses from lightbrown/streaked through to dark brown/black. Initial light brown streaking and graying occurs very quickly after infection as mycelium grows up the root. As mycelium develops it consumes root leaving behind only those compounds it cannot degrade such as lignins (Agrios 1978), darkening the root from its initial healthy white.

The fourth assumption (root mass is proportional to shoot mass) is necessary because direct measurements of the amount of root material present at the different harvests are not available. Shoot dry matter production was collected, along with root rot ratings (percentages of roots falling into four categorical ratings of health), and percentage containing mycelium. To determine the rate of mycelium development, the amount of root material as a function of the age of the crop is required, and to account for the reduced root material due to *Pythium* damage, the amount of root material is scaled according to the reduction in shoot mass.

It was assumed that growth rates of the mycelia are only a function of the availability of uninfected root. Infections in new cohorts with small root systems proceed slowly as there is not much root to infect. Infections in older plants with extensive root systems spread faster. Guo and Ko, (1993) published rates of growth of mycelia with units of  $\text{mm day}^{-1}$ . However, as we are dealing with a whole crop, it is more convenient to work on a mass basis. An overall rate simplifies the problem of



spread between adjacent roots, infections occurring at the top or bottom of the root (i.e. infection running out of places to spread to) and other complications of not using a mass basis approach.

To simplify the model, the assumption was made that rate of mycelium development varies only with temperature (increasing at higher temperatures). The data collected by Katzman in her experiments on inoculation timing and dosage was conducted at a single temperature, however, further experiments into the effect of temperature were also carried out by her in a separate chapter and were used to develop the relationship between mycelium ageing and temperature.

The basis for assuming that *Pythium* zoospores cause infections within the hour they are released is that zoospores are relatively short lived and will tend to rapidly infect available root material. The swimming stage only lasts approximately 30 minutes (Endo and Colt, 1974), after which the zoospores encyst and are unable to actively migrate towards roots. In floating pond hydroponics where the density of plants is great and the volume of nutrient solution relatively small, it is likely zoospores do not have to travel far before finding available roots to infect.

Using the same root to shoot ratio that was measured as a part of obtaining the data for the SUCROS spinach model (0.15) it is possible to estimate the root mass per plant from the measured shoot dry mass (Table 6.2). Then using the root quality data (Table 6.3) it is possible to estimate the amount of root that is infected with *Pythium* mycelium (Table 6.4).

**Table 6.4** Estimated influence of time of inoculation on mycelium infected root mass (g plant<sup>-1</sup>) of spinach based on measured shoot dry mass values, estimated root growth, and observed root condition. (Measured values from Katzman)

Inoculation time		Harvest Day 9	Harvest Day 14	Harvest Day 21
control	measured shoot dry mass (g plant <sup>-1</sup> )	0.01	0.051	0.268
	estimated root mass (g plant <sup>-1</sup> )	0.0015	0.00765	0.0402
	% root uninfected	100	100	96.7
	estimated mass infected (g plant <sup>-1</sup> )	0	0	0.00133
Day 1	measured shoot dry mass (g plant <sup>-1</sup> )	0.00157	0.008	0.032
	estimated root mass (g plant <sup>-1</sup> )	0.0002355*	0.0012	0.0048
	% root uninfected	10.3	12.5	20
	estimated mass infected (g plant <sup>-1</sup> )	0.00021	0.00105	0.00384
Day 9	measured shoot dry mass (g plant <sup>-1</sup> )	0.01	0.061	0.213
	estimated root mass (g plant <sup>-1</sup> )	0.0015	0.00915	0.03195
	% root uninfected	100	90	5.1
	estimated mass infected (g plant <sup>-1</sup> )	0	0.00092	0.03032
Day 14	measured shoot dry mass (g plant <sup>-1</sup> )	0.01	0.051	0.252
	estimated root mass (g plant <sup>-1</sup> )	0.0015	0.00765	0.0378
	% root uninfected	100	100	60
	estimated mass infected (g plant <sup>-1</sup> )	0	0	0.01512

\*Estimated root mass for the day 1 inoculated material harvested on day 9 is based on the same ratio as the day 1 inoculated material harvested on day 14. This ratio remained approximately the same for the harvest 21 and 28 data as well.

Assuming a constant growth rate of the form:

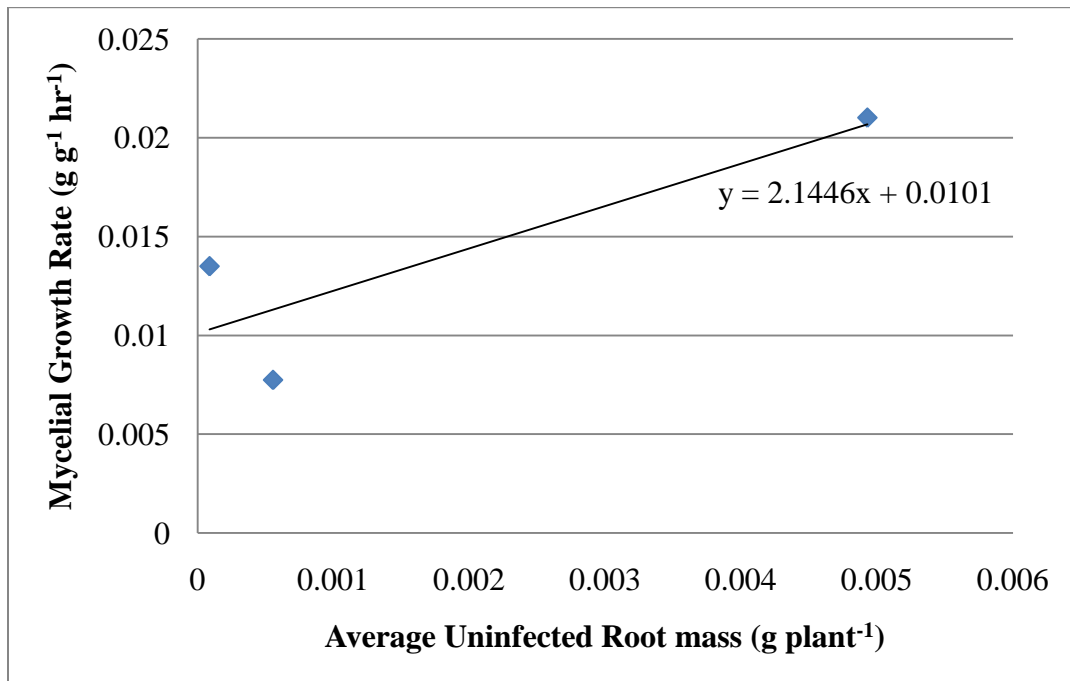
$$m_f = m_0(1 + gr_b)^n \quad 6.9$$

The growth rate ( $gr_b$ ) can be determined by rearranging equation 6.9 to:

$$gr_b = \left(\frac{m_f}{m_0}\right)^{\frac{1}{n}} - 1 \quad 6.10$$

Examining harvests on days 14 and 21 of the material inoculated on day 9, the estimated mass of infected roots increased from 0.00092 g plant<sup>-1</sup> to 0.03032 g plant<sup>-1</sup> over the course of the 168 hours between harvests. Using these values in equation

6.10 results in a growth rate of  $0.021 \text{ g g}^{-1} \text{ hr}^{-1}$ . The day 1 inoculated material has an estimated infected root mass of  $0.00021 \text{ g plant}^{-1}$ , on day 9, and  $0.00105 \text{ g plant}^{-1}$  on day 14, using equation 6.10 with 120 hours between harvests; a growth rate of  $0.0135 \text{ g g}^{-1} \text{ hr}^{-1}$ . On day 21 the day 1 inoculated material has an estimated infected root mass of  $0.00384 \text{ g plant}^{-1}$ . With the 168 hours between harvests this corresponds to a growth rate of only  $0.007 \text{ g g}^{-1} \text{ hr}^{-1}$ . The growth rate determined from the day 9 inoculation material is up to three times as large as that from the day 1 inoculation. However, the day 9 inoculation material was an infection spreading through a large established root system, whereas, the day 1 inoculation material was constrained by the lower availability of infectable root. To account for the increasing difficulty in finding new material to infect the mycelium growth rate was plotted vs. the average mass of uninfected root as illustrated in Figure 6.2.



**Figure 6.2** Mycelium growth rate as a function of the average uninfected root mass.

Assuming the growth rate is a linear function of the average mass of the roots that are uninfected gives the relation;

$$gr_p = 2.144 * URM_{avg} + 0.010 \quad 6.11$$

Scaling the average amount of root that is uninfected from g plant<sup>-1</sup> to kg ha<sup>-1</sup> (for use in the model), assuming a density of 1100 plants m<sup>-2</sup>, gives the relation;

$$gr_b = \beta_{grow} * URM_{avg,kg\ ha^{-1}} + \alpha_{grow} \quad 6.12$$

The growth of mycelium is limited such that it encompasses no more than 95% of the total root mass (the maximum observed infection percentage). This capping includes infection from free zoospores as well. In addition the growth rate is limited to maximum rate observed (temperature adjusted).

*Infection of healthy roots by zoospores:*

The *Pythium* infection cycle starts when a piece of mycelia, a zoospore or an oospore contacts living root material, and begins to spread. This model is primarily concerned with the infection of healthy root material with zoospores as this is the mode of infection of particular interest in a floating hydroponic system. Because the plants of a crop share a common well mixed pond, zoospores can quickly spread through the entire crop.

When a zoospore encounters healthy root material it quickly bonds to the root and begins to grow mycelia. The more zoospores in solution, the higher probability there is for healthy root material to become infected. However, not all zoospores are viable, not all form mycelia, and some may attack roots close to where mycelium already exists. To account for the variability in the infectivity of the zoospores, rather

than consider an absolute number of infections, a specific mass of healthy root is said to be infected. This mass is assumed to be a function of the concentration of the zoospores in solution and is limited by the available mass of healthy root to infect.

Using the mycelium growth rate determined previously and rearranging equation 6.10 to solve for the starting mass of mycelia, the equivalent mass of mycelia the infecting zoospores create in the roots can be calculated using equation 6.13.

$$\epsilon_{\text{mycelium}} = m_0 = \frac{m_f}{(1 + gr)^n} \quad 6.13$$

The equivalent mass of mycelium can be estimated from three starting points; day 1 inoculated material, day 9 inoculated material and day 14 inoculated material. From Table 6.4, plants inoculated on day 9 and harvested on day 14, developed a mycelium mass of  $9.2\text{E-}4 \text{ g plant}^{-1}$ , over the course of 120 hours. Assuming a mycelium growth rate of  $0.0105 \text{ g g}^{-1} \text{ hr}^{-1}$  (determined from equation 6.10), gives a starting mycelium mass of  $2.42\text{E-}4 \text{ g plant}^{-1}$ . Plants inoculated on day 14 and harvested on day 21, developed a mycelium mass of  $0.0151 \text{ g plant}^{-1}$ , over the course of 168 hours. Assuming a growth rate of  $0.0204 \text{ g g}^{-1} \text{ hr}^{-1}$ , gives a starting mycelium mass of  $5.06\text{E-}4 \text{ g plant}^{-1}$ . Material inoculated on day 1 and harvested on day 9 developed a mycelium mass of  $2.1\text{E-}4 \text{ g plant}^{-1}$ , over the course of 216 hours. Because the starting mass of roots is not known, the lowest mycelium growth rate of  $0.010 \text{ g g}^{-1} \text{ hr}^{-1}$  was assumed. This growth rate corresponds to a starting mycelium mass of  $5.88\text{E-}5 \text{ g plant}^{-1}$ . Averaging these three values results in a reference equivalent mass of mycelium of  $2.69\text{E-}5 \text{ g plant}^{-1}$ , or  $3.23 \text{ kg ha}^{-1}$ , when the concentration of zoospores is  $25 \text{ zoos ml}^{-1}$ .

### *Damage to Crop*

The basis for determining the effectiveness of root function and quality of shoot material is both the quantity and characteristics of damage to the roots. Previous sections have dealt with determination of growth and spread of mycelium which is primarily a quantity issue, i.e. the amount of the root infected. In her quantification of root damage, Katzman categorized roots according to their appearance. Besides the undamaged categorization she further broke the infected category down into roots that have light brown/gray streaking, and those that are dark brown/black. The light brown streaking corresponds to a recent mycelium infection of the roots, which eventually matures into dark brown/black root material. The presence of dark brown/black root material usually signified severe damage to the crop and significant biomass reductions. The presence of light brown streaking did not always correspond to a reduction in biomass of the plant. For this reason estimating when damaged root material matures into the dark brown/black stage provides a means to estimate both the quality and reduction in growth of the shoot.

It is assumed that maturation of root damage is strictly a function of the age of the mycelium, and is not a response to external triggers such as temperature changes or other stresses put on the mycelium.

To estimate the age at which mycelium matures into the dark brown/black stage ( $A_{\text{mature}}$ ), data from Katzman's time of inoculation experiment was used. It was necessary to make the assumption the dark brown/black stage is the final condition of the mycelium. In addition it was also assumed the "no root" category of Katzman also belongs to this final condition (essentially dead root material). Examining the day 9 inoculated material, on harvest day 14, 10 percent of the root is in the light brown/streaked stage. Seven days later on harvest day 21, 76.3 percent of the root was light brown/streaked and 18.6 percent was dark brown/black. By inserting values of

mycelium maturation age into the mycelium growth model in trial and error fashion it was found a mycelium maturation age of 128 hours (~5.3 days) produced a fraction of roots dark brown/black of 0.186.

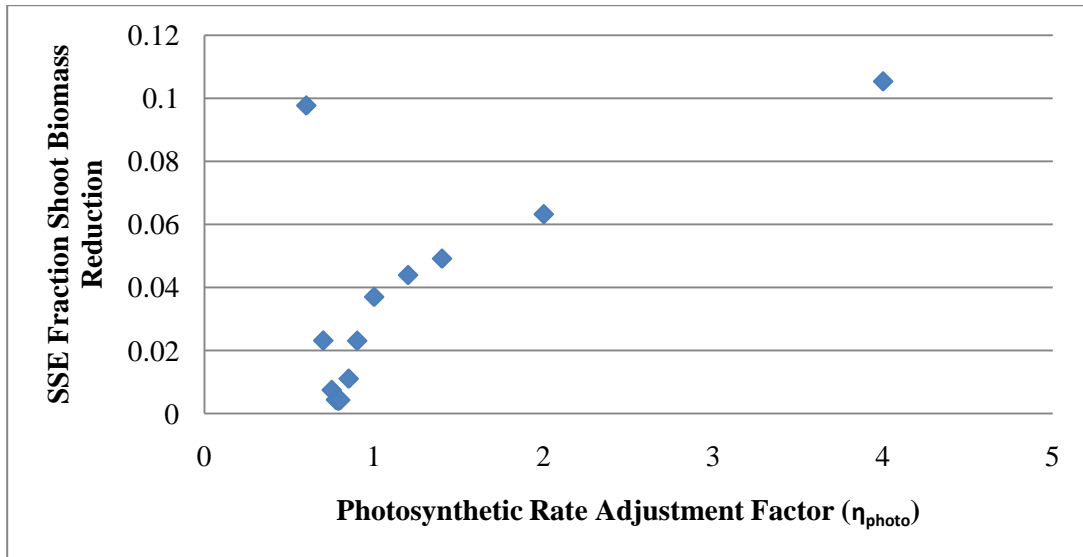
By attacking roots, *Pythium aphanidermatum*, reduces the ability of the crop to acquire water and nutrients necessary for normal plant function. Symptoms range from little to no reduction in growth, to complete death of the plant, depending upon the extent of the infection. Examining tables 6.1 and 6.2 which are shoot dry mass and root rot ratings for Katzman's time of inoculation experiment, it is apparent shoot biomass accumulation is only significantly impacted when root damage is heavy.

To reflect the effects of the infection, production of photosynthate by the leaves, can be decreased as a function of root damage. Reduced photosynthate production corresponds to less energy and building blocks for crop functions such as maintenance and new growth.

To scale the photosynthate production it was assumed the presence of infected root material decreased photosynthate production in a linear fashion.

$$PRAD = 1 - \eta_{photo} * \delta_{root} \quad 6.14$$

The value of  $\eta_{photo}$  was selected so the sum of the squared differences between the observed and predicted percent shoot mass was minimized (for the day 1 inoculation harvests on days 14 and 21 and the day 9 inoculation harvest on days 14 and 21. Day 1 inoculation harvest day 9 was not included, as this value was missing from the observed data). As can be seen in Figure 6.3 this resulted in an  $\eta_{photo}$  value of 0.79.



**Figure 6.3** Sum of the Squared Error of the fraction shoot biomass reduction as a function of the coefficient of photosynthetic rate reduction ( $\eta_{photo}$ ).

### 6.3.2 Adjustment to Mycelium Growth and Development:

Because the temperature of the nutrient solution may not always be 24 C, it was necessary to scale growth and development of the mycelium as a function of temperature. In addition, scaling is also required for the amount of mycelium formed by zoospores attacking roots, as it is unlikely a crop would be exposed to a level of 25 zoospores ml<sup>-1</sup>, such as was used to develop the reference infection mass.

#### *Concentration adjustment*

As reported in the fourth chapter of Katzman (2003) entitled “Relationship Between Inoculum-Dosage and Development of Root Rot Caused by *Pythium aphanidermatum* in Hydroponic Spinach Production,” she conducted an experiment where she inoculated ponds of spinach with known concentrations of zoospores and measured growth of the crop through periodic harvesting at days 9, 14, 21 and 28. In addition, Katzman also studied time of inoculation, introducing known concentrations of zoospores 9, 14 and 21 days after floatation. This data forms the basis for



developing a function to relate the concentration of zoospores in solution to the infection level of the crop roots.

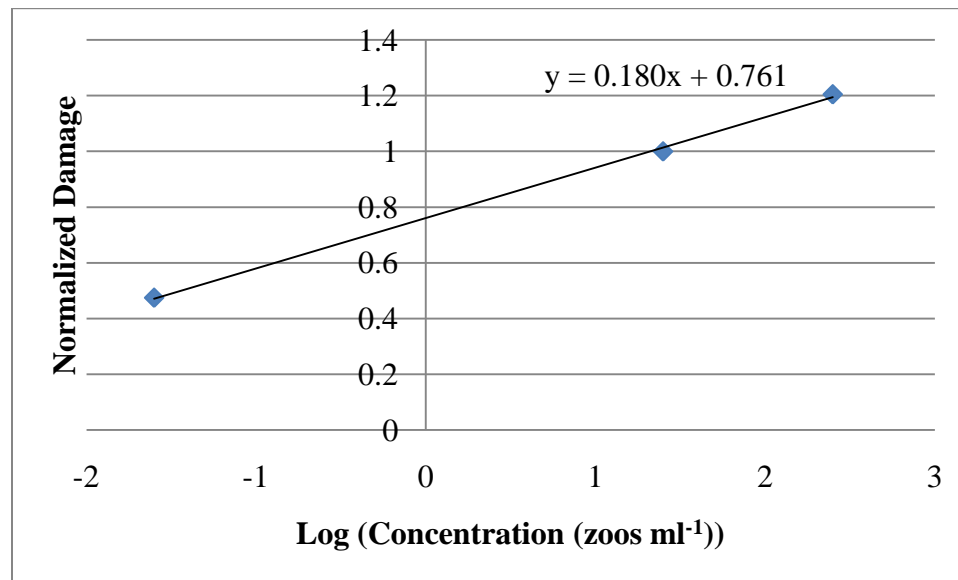
As was the case for the temperature effect, results were more noticeable if the root rot rating data is used, and if the data at harvest 21 is included. The results of this experiment are presented in Table 6.5.

**Table 6.5** Percentages (%) of roots within each root rot rating (1-3) at harvest. Spinach plants were inoculated with *P. aphanidermatum* zoospores (0.025 to 250 per ml in 10-fold increments) 9 days after seeding and harvested 14, and 21 days after seeding. (From Katzman)

Treatment	Percentage (%) of roots within Root Rot Rating at harvest		
Dosages- zoospores per mL	Root Rot Rating	Harvest 14	Harvest 21
Control	1- white	100	91.7
	2 - light brown/streaked	0	8.3
	3 - dark brown/black	0	0
0.025	1- white	96.7	33.9
	2 - light brown/streaked	3.4	50.8
	3 - dark brown/black	0	15.3
0.25	1- white	94.9	10
	2 - light brown/streaked	5.1	56.7
	3 - dark brown/black	0	33.3
2.5	1- white	83.3	6.7
	2 - light brown/streaked	16.7	76.7
	3 - dark brown/black	0	16.7
25	1- white	88.3	0
	2 - light brown/streaked	11.7	67.8
	3 - dark brown/black	0	32.2
250	1- white	80	2
	2 - light brown/streaked	20	59.2
	3 - dark brown/black	0	38.8

Unfortunately, there is confounding data when examining the 0.25 and 2.5 zoos ml<sup>-1</sup> treatments. The 0.25 zoos ml<sup>-1</sup> treatment appears to have as much damage

as the 25 zoos ml<sup>-1</sup>. And the 2.5 zoos ml<sup>-1</sup> treatment has a lower level of damage than would be expected looking at the other values. Leaving these two treatments out and considering the amount of dark brown/black root material, a relation for the effect of zoospore concentration on the initial infection amount can be calculated. If the damage at 25 zoos ml<sup>-1</sup> is used as the baseline and the log of the zoospore concentration vs. the normalized damage (damage at concentration divided by damage at 25 zoos ml<sup>-1</sup>) is plotted, as illustrated in Figure 6.4, a linear relationship is obtained.



**Figure 6.4** Normalized *Pythium* damage as a function of initial concentration of zoospores. (Data from Katzman).

The normalized damage correction factor is thus:

$$E(C) = \beta_{\text{conc}} * \log(Z_{\text{conc}}) + \alpha_{\text{conc}} \quad 6.15$$

The factor E(C) is then used to adjust the reference equivalent mass of mycelium ( $\epsilon_{\text{mycelium}}$ ) (equation 6.6) which is used to relate the concentration of zoospores in solution to mycelium mass.

*Temperature effects:*

In Katzman (2003) dissertation her fifth chapter was entitled “Influence of Nutrient Solution Temperature on Plant Growth and Disease Development.” For this study Katzman grew spinach crops with nutrient solution temperatures of 18, 24 and 30 C, and inoculated them with *Pythium aphanidermatum* at a level of 25 zoos ml<sup>-1</sup>. The inoculation occurred on day 9 after sowing, and harvests occurred on days 9, 14, 21 and 28. Summary results are presented in Table 6.6.

**Table 6.6** Influence of nutrient solution temperature and *P. aphanidermatum* on shoot dry mass. Means and standard errors of shoot dry mass (g per plant) for harvests at 9, 14, and 21 days after seeding. Treatments varied nutrient solution temperatures (18, 24, and 30 C) and presence or absence (+/-) of *P. aphanidermatum* (*P.a.*). (From Katzman, 2003)

Treatment	Mean shoot dry mass (g/plant) and standard errors at harvest		
Temperature C +/- P.a.	Harvest 9	Harvest 14	Harvest 21
18 - P.a.	0.028 ± 0.00†	0.23 ± 0.01	1.32 ± 0.04
24 - P.a.	0.028 ± 0.00	0.22 ± 0.01	1.48 ± 0.05
30 - P.a.	0.028 ± 0.00†	0.23 ± 0.01	1.13 ± 0.02
18 + P.a.	0.028 ± 0.00†	0.23 ± 0.01	1.47 ± 0.05
24 + P.a.	0.028 ± 0.00	0.24 ± 0.01	1.34 ± 0.06
30 + P.a.	0.028 ± 0.00†	0.15 ± 0.01	0.34 ± 0.02

† Temperature treatment has not yet been applied.

Unfortunately, the 25 zoos ml<sup>-1</sup> dose Katzman used is relatively low and so the temperature effects are not as apparent when examining the shoot dry mass data. In addition, inoculation was carried out on day 9, rather than upon initial floatation, which would have provided the infection more time to develop. The only notable difference between the shoot dry masses on harvest day 14 is for the high temperature, *Pythium* inoculated treatment. Fortunately, differences are more apparent in the root rot rating data for the same experiment, which are presented in Table 6.7.

**Table 6.7** Influence of nutrient solution temperature and inoculation with *P. aphanidermatum* on root rot rating (RRR). Percentages (%) of roots within each RRR category per treatment for plants harvested 14, and 21 days after seeding. Treatments varied the nutrient solution temperature (18, 24, and 30 C) and presence or absence (+/-) of *P. aphanidermatum* (*P.a.*). (From Katzman)

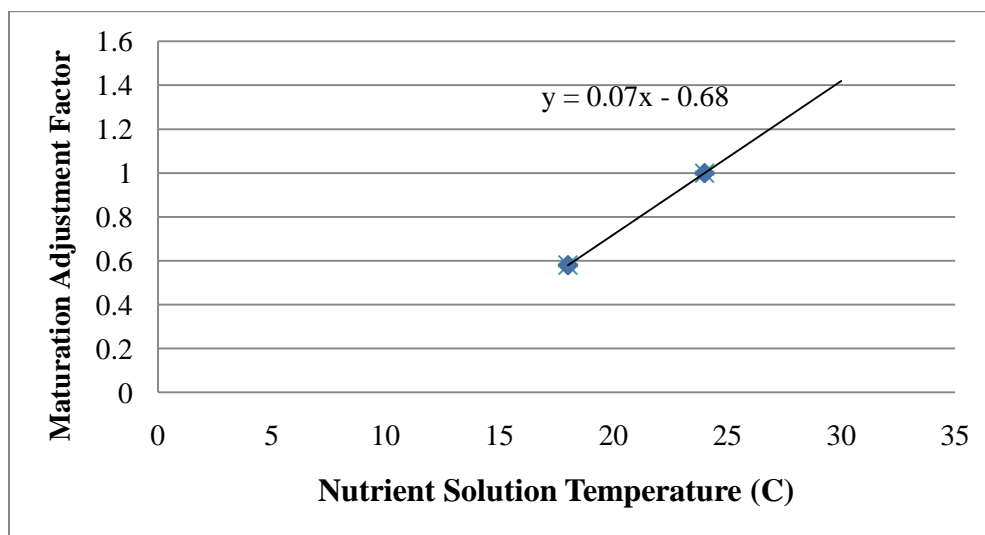
Treatment	Percentage (%) of roots within each RRR category at harvest day		
Temperature C +/- P.a.	RRR Category	Harvest 14	Harvest 21
18 C - P.a.	1- white	100	100
	2 - light brown/streaked	0	0
	3 - dark brown/black	0	0
24 C - P.a.	1- white	100	100
	2 - light brown/streaked	0	0
	3 - dark brown/black	0	0
30 C - P.a.	1- white	97.6	53.8
	2 - light brown/streaked	2.4	44.9
	3 - dark brown/black	0	1.3
18 C + P.a.	1- white	100	82.1
	2 - light brown/streaked	0	17.9
	3 - dark brown/black	0	0
24 C + P.a.	1- white	57.1	6.3
	2 - light brown/streaked	42.9	79.7
	3 - dark brown/black	0	13.9
30 C + P.a.	1- white	47.6	0
	2 - light brown/streaked	21.4	1.3
	3 - dark brown/black	31	98.8

In the temperature trials Katzman performed it was clear the amount of root damage increased as a function of temperature. Katzman also found zoospores were released from the infected roots earlier in the high temperature conditions. These observations mean the mycelium grows and matures faster and causes increased damage by releasing zoospores, which further infect the root. Another assumption to simplify the temperature effect of the nutrient solution is temperature does not affect the plant itself. This is not entirely the case as can be seen in Table 6.6, comparing shoot mass of the uninoculated treatments at harvest day 21. According to this data,

there is a penalty for growing at temperatures that are too high. Potentially exacerbating this effect is the fact the crop might be stressed at higher temperatures, making it more vulnerable to *Pythium* damage. The increase in mycelium development and growth rate at higher temperatures encompasses this effect.

A further assumption is there is no deleterious effect from temperature changes on the growth of mycelium. Temperatures found in the ponds are within a relatively constant range and any changes are gradual due to the relatively large volume.

With only three temperatures to use, the mid value of 24C was selected as baseline (as this was the temperature common to all of Katzman's experiments). To find the adjustment factor for 18C, the conditions of Katzman's experiment were used as input to the model (25 zoos ml<sup>-1</sup> inoculated day 9). Through trial and error an adjustment factor of 0.58 was found to produce the observed 17.9% light brown/streaking in the roots at harvest day 21. Because it is likely that damage in the roots at 30C was also partially due to reinfection, this point was not used in the development of the temperature relation. The 18 and 24 C points are plotted in Figure 6.5.



**Figure 6.5** Age adjustment factor as a function of nutrient solution temperature.

Assuming a linear relation results in:

$$E(T) = \beta_{\text{temp}} * T - \alpha_{\text{temp}} \quad 6.16$$

### 6.3.3 Sources of inoculum

Natural *Pythium* infections must begin at some point. Usually a few spores will infect a single plant, and the subsequent release of zoospores will spread the infection to the rest of the crop. Stanghellini and Rasmussen (1994), identified growing medium such as peat and sand, along with greenhouse dust and even water supplies, as potential sources of inoculum. In addition, they found common greenhouse pests such as shore flies and fungus gnats can potentially transmit *Pythium* spores. In her dissertation, Katzman (2003) investigated sources of *Pythium* inoculation in the Cornell greenhouse. Potential sources examined included tap and reverse osmosis supply water, greenhouse medium, and dust, as well as the spinach seeds themselves. Unfortunately, she was not able to isolate *Pythium* from any of these sources. One explanation for this was the number of *Pythium* propagules was below her detection limit. Regardless, spontaneous *Pythium* infections occurred, confirming that in hydroponic situations, a very small number of *Pythium* propagules can quickly mature into a full blown infection.

For the disease model it is necessary to have a starting point to simulate a spontaneous inoculation. In Katzman's various studies she noticed browning of roots on mature plants always seemed to occur typically at 21 to 25 days after sowing. Examining the control data in Table 6.2 we can see 21 days after sowing, 3.3% of the roots were showing signs of the browning associated with *Pythium*. The experiments carried out to study the progress and characteristics of infections, relied on dosing with zoospores at concentrations many orders of magnitude greater than would be expected for any naturally occurring infection. One option is to assume a set baseline

concentration of zoospores are always present (a very low concentration). However, a problem with this technique is concentration damage relationship developed previously has an extrapolated lower limit concentration of 0.015 zoospores ml<sup>-1</sup> (found by solving equation 6.15 for the zoospore concentration at which the adjustment factor equals 0).

Another option is to assume the seedlings are already inoculated to a certain level before they enter the pond, such as would be the case if zoospores/other infectious material were found in the growing medium. To estimate a value for this initial mass of mycelia, the developed disease model was used, along with the control data for Katzman's time of inoculation experiment. Using the same conditions Katzman used (air temperature 24C and 16 mols PAR m<sup>-2</sup> day<sup>-1</sup>) as input to the growth and disease model, it was found that a starting mycelial mass of 1.5 E-3 kg mycelium ha<sup>-1</sup> resulted in damage to 3.3% of the roots at day 21, which is the level Katzman observed. There was no corresponding shoot biomass reduction at this low level of damage. This starting mass ( $m_0$ ) is thus assumed to exist as a baseline in every new cohort.

#### 6.3.4 Release of zoospores

Infected roots develop sporangia, which in turn release zoospores. Under laboratory conditions, the entire reproductive process of *Pythium* can be as short as 24 hours, zoospore to zoospore, but can take significantly longer in a hydroponic system. Benchtop evaluations and data from Katzman indicate this reproductive cycle time is dependent on temperature. In Katzman's work, conducted at 24C, she monitored the level of zoospores present in solution. Unfortunately, her techniques did not allow an accurate means of measuring these levels, as illustrated by the fact immediately following inoculation she could not measure the same number of zoospores as were

inoculated (Table 6.8). A complicating factor in making such a measurement is that zoospores actively migrate towards roots and in the relatively root dense environment of floating hydroponics it is likely that the zoospores released, rapidly find new roots to infect and are not measured as free zoospores.

**Table 6.8** Influence of inoculation time, zoospore concentration and temperature on zoospores detected in nutrient solution at time of harvest. (From Katzman)

Experimental Condition	Treatment means and standard errors of zoospores per ml nutrient solution on harvest day			
	Harvest 1	Harvest 9	Harvest 14	Harvest 21
Innoculation Time				
day 1	0.87 ± 0.25	0.13 ± 0.03	0.01 ± 0.00	0.04 ± 0.03
day 9		0.99 ± 0.16	0.29 ± 0.02	0.06 ± 0.02
day 14			1.00 ± 0.22	0.09 ± 0.03
day 21				0.82 ± 0.13
Zoospore conc (zoos ml <sup>-1</sup> ) (innoculated day 9)				
0		0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
0.025		0.0 ± 0.0	0.001 ± 0.001	0.012 ± 0.003
0.25		0.021 ± 0.007	0.004 ± 0.002	0.011 ± 0.003
2.5		0.165 ± 0.031	0.007 ± 0.005	0.008 ± 0.002
25		1.091 ± 0.115	0.246 ± 0.066	0.067 ± 0.020
250		8.146 ± 1.362	1.269 ± 0.132	0.080 ± 0.011
Temperature (C) (innoculated day 9)				
18		3.61 ± 0.18	0.03 ± 0.01	0.00 ± 0.00
24		5.19 ± 0.71	0.01 ± 0.00	0.02 ± 0.01
30		4.72 ± 0.77	0.61 ± 0.21	1.19 ± 0.22

Looking at the day 1 inoculated material, on day 9 it is apparent zoospores are being released into solution. Encysted zoospores were found to survive up to 7 days in moist soil conditions (Stanghellini and Burr 1973), but since these zoospores were detected 9 days after inoculation it is likely they are the results of reproduction from the original infection. Similarly, with the day 9 through day 21 inoculated material zoospores were detected after 7 days.



Stanghellini and Rasmussen (1994) determined the reproductive capacity of a zoosporic pathogen similar to *P. aphanidermatum* and estimated that a single mature lettuce plant with 2,000 cm<sup>2</sup> of root surface area, infected with the zoosporic pathogen *Plasmopara lactucae-radicis*, is capable of producing 8 million zoospores. The release of zoospores can be characterized by two variables; the age at which mycelia develops sporangia capable of releasing zoospores, and the number of zoospores released. For the purposes of this model it is assumed the timing of release is only a function of the age of the mycelium (and not some external trigger). It is also assumed the number of zoospores released is only a function of the total mass of mycelium tissue that is of zoospore releasing age.

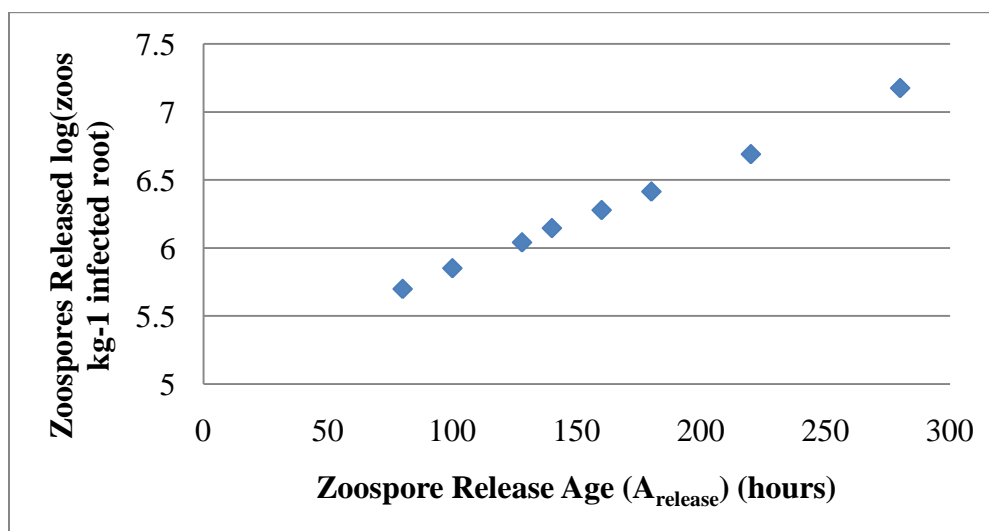
To simplify the model the assumption is made that all zoospores released are viable and find root material to infect (if there are uninfected roots present). This simplification means zoospores that are released, but not viable, and zoospores that do not find roots to infect do not have to be accounted for. This also implies the assumption that the proportion of zoospores falling into the category of unviable, remains constant.

The assumption is also made that mycelium of a certain age release zoospores and continue to release zoospores until the root material is removed at harvest time. The total number of zoospores released by a cohort of the crop can be determined by multiplying the mass of zoospore releasing mycelium by a constant factor with units of zoospores per kg root.

To determine the number of zoospores released, the spinach growth and disease model was run with the parameters determined from Katzman's temperature data. In the development of the temperature relation, the 30 C condition was not used because it is likely that a significant amount of the mycelium growth was due to reinfection from zoospores released from the same root material. Instead this data was

used to determine the zoospore release age and number of zoospores released per kg of mature mycelium. At 30 C, with no inoculation, 46.2% of the roots were infected at harvest 21.

In determining the zoospore release age, and the number of zoospores released per kg of mature mycelium it became apparent these two variables are highly correlated to one another. By selecting a younger zoospore release age, a smaller number of zoospores released per kg would produce the 46.2% damage in the roots at harvest day 21. Similarly an older zoospore release age required a larger number of zoospores to cause the same amount of damage. This relationship is plotted in Figure 6.6.



**Figure 6.6** Zoospore release amount as a function of zoospore release age

To select a pair of zoospore release age and amount, the model was run with Katzman's data for the 24C uninoculated control. This condition was common to the three experiments she conducted. In the temperature experiment, no damage was apparent in harvests 14 or 21; in the concentration experiment, at harvest day 21, 8.3% of the roots were light brown/streaked. In the time of inoculation experiment the control showed 3.3% light brown/streaking at harvest 21. It was found that a

combination producing 8.3%, corresponded to a zoospore release age of 360 hours and a zoospore release amount of  $8.6E7$  zoos  $kg^{-1}$   $A_{release}$  mycelium. However, this is longer than the assumed mycelium maturation age, and so a  $A_{release}$  and corresponding release amount of 128 hours and  $1.1E6$  zoos  $kg^{-1}$   $A_{release}$  mycelium was selected. This produces light brown streaking in 17.6 % of the roots at harvest day 21, which is larger than observed, but not unreasonable.

### 6.3.5 Damage to shoot quality

Because there is a harvestable mass of spinach shoot, does not mean that the material is saleable. Root damage may not completely kill a plant, but it may cause various levels of wilting, discoloration or other unappealing characteristics which would prevent consumers purchasing it. To account for the true lost product due to *Pythium* damage, it is necessary to incorporate a shoot quality factor to account for quality losses in addition to reduced growth.

In chapters three and four of her dissertation, Katzman (2003) documented shoot quality as a part of her study of the effects of plant age at time of inoculation, and inoculation dosage on disease development. Shoot quality was visually assessed and placed into 6 categories:

- 1.) Green turgid leaves and cotyledons;
- 2.) Cotyledons yellowing or necrotic;
- 3.) Several true leaves or entire shoot yellowing;
- 4.) Shoot slightly wilted;
- 5.) Shoot completely wilted;
- 6.) Shoot completely dry and appearing “dead.”

The categories progress from healthy to dead plants. For commercial purposes only material in categories one and two are acceptable. Yellowing or necrotic cotyledons

are not desirable, but this yellowing is also a condition found in healthy plant growth (though usually in more mature plants). The cotyledons (grown from seed reserves) provide the first photosynthetic structure of the plant. Once true leaves have developed, cotyledons are no longer necessary and plants will generally begin to translocate their resources into other growth. A fraction of yellowing or necrotic cotyledons was also seen in Katzman's control data. Yellowing in the true leaves is unattractive to the customer, and wilting cannot be tolerated, as such tissues quickly decay and spoil. Though some shoots in the crop might appear fine it is impossible to commercially selectively harvest plants in a densely packed baby spinach crop. If a percentage is bad, the whole crop cohort is considered lost. An assumption of the model is that damage is spread uniformly through the crop.

While categorizing the condition of the shoots, Katzman also documented the root rot rating, which has been described and used previously in this model. As in the determination of the effectiveness of the root due to root damage, the quality of the shoots is related to the root rot rating, and the percentage of the root that is dark brown/black.

Examining Katzman's data it can be seen that shoot quality becomes unacceptable when there is *any* dark brown/black root material present (indicating an advanced stage of infection). More difficult to assess is the case of slight wilt in a small percentage of the crop. When above 10 to 15% of the root material is light brown/streaked, slight to complete wilting of the plant occurs in a small percentage of plants in the crop.

The shoot quality factor allows the determination of whether to terminate a crop, because it is unsaleable even if not totally dead. It is important to be able to simulate shoot quality as losses are both biomass and quality related, and in

commercial production a grower would remove the material damaged beyond recovery as soon as it reaches that state, rather than at the usual time of harvest.

#### 6.3.6 Validation of the *Pythium* disease model:

In the estimation of several parameters an assumption was made that effects from second generation zoospores were negligible. (I.e. zoospores released from mycelium formed from the initial infection). The first set of validations checks this assumption by running the entire model with a single crop for 21 days and comparing the growth and root damage predicted, to that observed in Katzman's time of inoculation, inoculum concentration, and nutrient solution temperature experiments. The results were compared by normalizing the individual plant growth measurements of Katzman by dividing the condition biomass by the control biomass and the crop predictions by dividing the expected growth, by growth under identical conditions without *Pythium* effects. Fraction root damage is directly comparable and fraction of the roots infected and fraction dark brown/black were also examined.

The second set of validation checks examined the performance of the model in predicting multi-cohort disease effects. For these checks results of chapter three were used. Model and observed harvest biomasses were compared for known inoculation concentration and timing experiments with both cool (20C) and warm (27.5C) nutrient solution temperatures.

An additional check compared the effect of crop duration and nutrient solution temperature on the number of saleable harvests before *Pythium* damage became too severe. As a crop spends longer in the pond, there is greater opportunity for zoospores to reproduce and spread to further cohorts. Similarly, higher temperatures produce this same effect by reducing the amount of time it takes for mycelium to grow, mature and release zoospores to further the infection.

### 6.3.7 Sensitivity analysis of model parameters:

To determine how sensitive the crop disease model is to the values of the parameters chosen, each parameter was individually varied by plus and minus 10%, and the model run using the original conditions from which it was derived, namely inoculation on day 1 and day 9, and harvests on days 9, 14 and 21. Also as in the validation, the output examined included normalized harvest biomass (infected/uninfected), fraction of roots infected and fraction of roots dark brown/black. The results of these runs were then summarized in 18 tornado plots (1 plot for each inoculation (2), harvest (3) and output type combination (3)).

A similar process was also conducted for evaluating the parameters estimated for temperature, initial zoospore concentration, zoospore release age, zoospore release number and concentration parameters. The parameters were again varied plus and minus 10% and input to the model, which was run with the uninoculated conditions used in their estimation. The model used nutrient solution temperatures of 24 and 30C, no inoculation, and harvests on day 21. As before, normalized harvest biomass, fraction of the roots infected and fraction of the roots dark brown/black were examined. The results from these runs were summarized in 6 tornado plots. Concentration of zoospore effects were included in this tornado plot as the uninoculated temperature experiment relied on reinfection which uses concentration effect parameters.

## **6.4 RESULTS:**

### 6.4.1 Validation of the *Pythium* disease model:

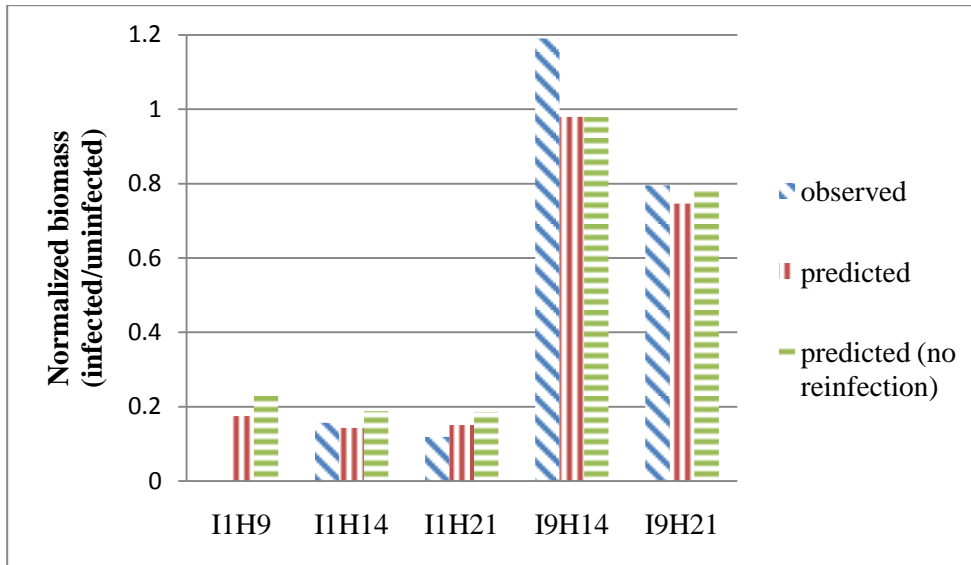
The validation of the *Pythium* disease model was divided into three parts. The first part is concerned with validating the model parameters derived from Katzman's data, which focused on single crop effects. The second part is concerned with the

spread of the *Pythium* infection in a multi-cohort system and uses the data collected and presented in chapter three. The third check is not compared against any data, but is presented to illustrate how the model performs in predicting multi-cohort system performance as a function of temperature and harvest time.

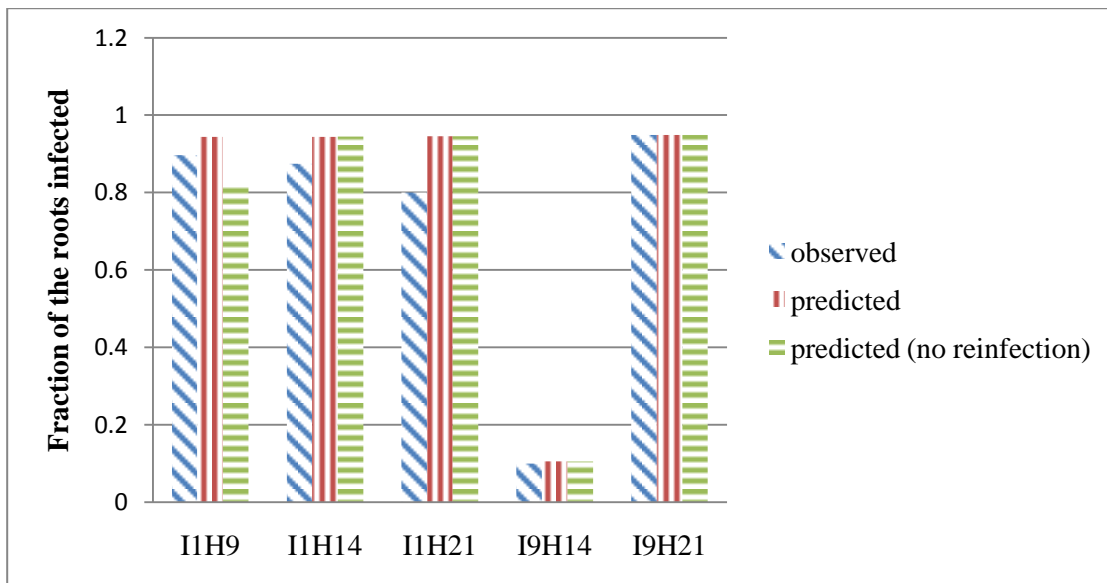
#### *Validation of single crop Pythium disease model parameters*

The spinach disease model was developed with observed shoot biomass and root rot rating data from Katzman. Parameters describing mycelium growth rate, the initial infection amount due to zoospores, the maturation of the mycelium and the effect of root damage on shoot biomass were based on data from Katzman's time of inoculation experiment.

An assumption used in estimating these parameters was that effects of second (and more) generation zoospores released from the crop were negligible compared to the initial inoculation. To facilitate comparisons between Katzman's observed single plant data, and the model's predicted whole crop values, the results were normalized by dividing the infected values by the uninfected values. This assumption allowed the effects of the *Pythium* infection to be expressed as fractions which could be compared side by side. The effects compared include the harvest biomass reduction, fraction of the roots infected, and fraction of roots appearing dark brown/black (fraction of roots light brown/streaked can be determined by subtraction). Data for inoculations on days 1 and 9 (I) and harvests at days 9, 14 and 21 (H) are presented in Figure 6.7, Figure 6.8 and Figure 6.9. Figure 6.7 is a plot of the biomasses, Figure 6.8 is a plot of the fraction of the roots infected, and Figure 6.9 is a plot of the fraction of the roots that are dark brown/black.

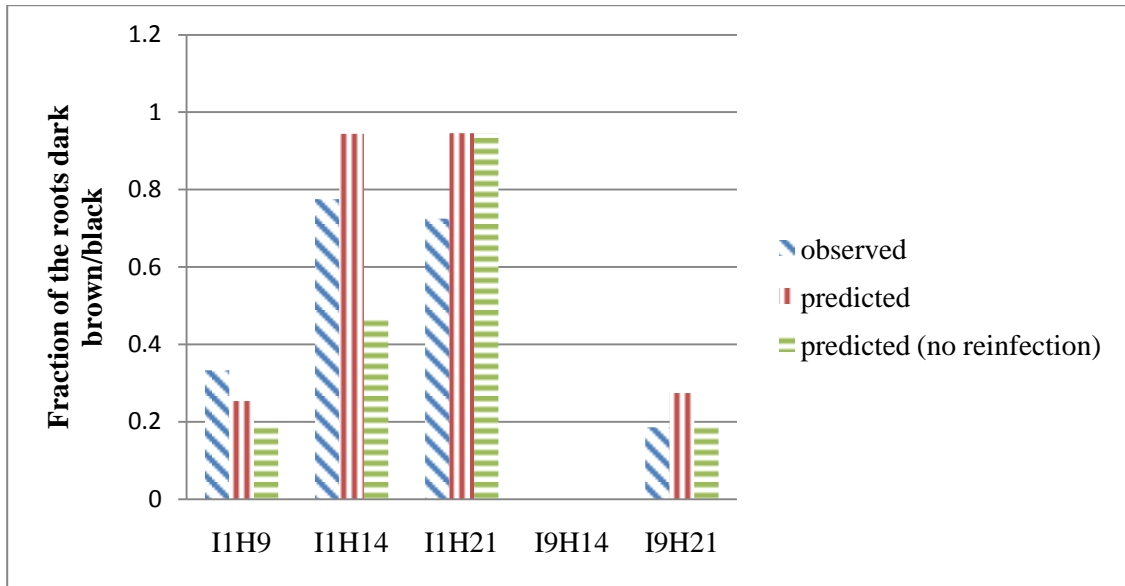


**Figure 6.7** Yield ratio (infected biomass/uninfected biomass value). From Katzman (2003) observed time of inoculation experiment and model predictions (with and without reinfection) at a root zone temperature of 24C. Code is I, followed by inoculation day, and H, followed by harvest day.



**Figure 6.8** Fraction of roots infected. From Katzman's observed time of inoculation experiment and model predictions (with and without reinfection). Code is I, followed by inoculation day, and H, followed by harvest day.



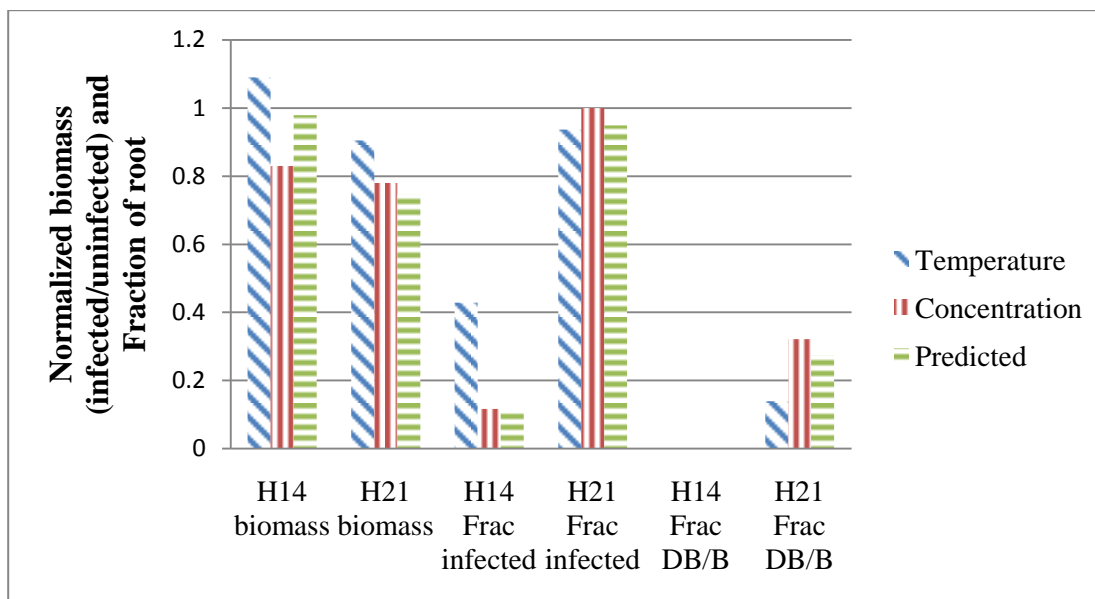


**Figure 6.9** Fraction of roots dark brown/black. From Katzman's observed time of inoculation experiment and model predictions (with and without reinfection). Code is I, followed by inoculation day, and H, followed by harvest day.

As can be seen from these three figures, the inclusion of reinfection of the crop roots with released zoospores has no major effect on the predicted biomass, and the predicted root infection amount. The only large difference in the fraction of roots dark brown/black was in day 1 inoculated material at harvest 14, where the inclusion of reinfection improves the accuracy. This finding supports the assumption there was little to no effect of second (or more) generation zoospore effects on parameters describing mycelium growth rate, initial infection amount due to zoospores, maturation of the mycelium, and effects of root damage on shoot biomass.

To validate the single crop portion of the model itself and the selection of parameter values, data from Katzman's further experiments concerned with concentration of initial inoculation and temperature of nutrient solution were used. During these later two experiments, Katzman had conditions that corresponded to the conditions used in her time of inoculation experiment; specifically an inoculation of 25 zoos ml<sup>-1</sup> at day 9 with a nutrient solution temperature of 24 C. These conditions

are the same as those used in the experiments on which data the model is based. The observed and predicted results are presented in Figure 6.10.

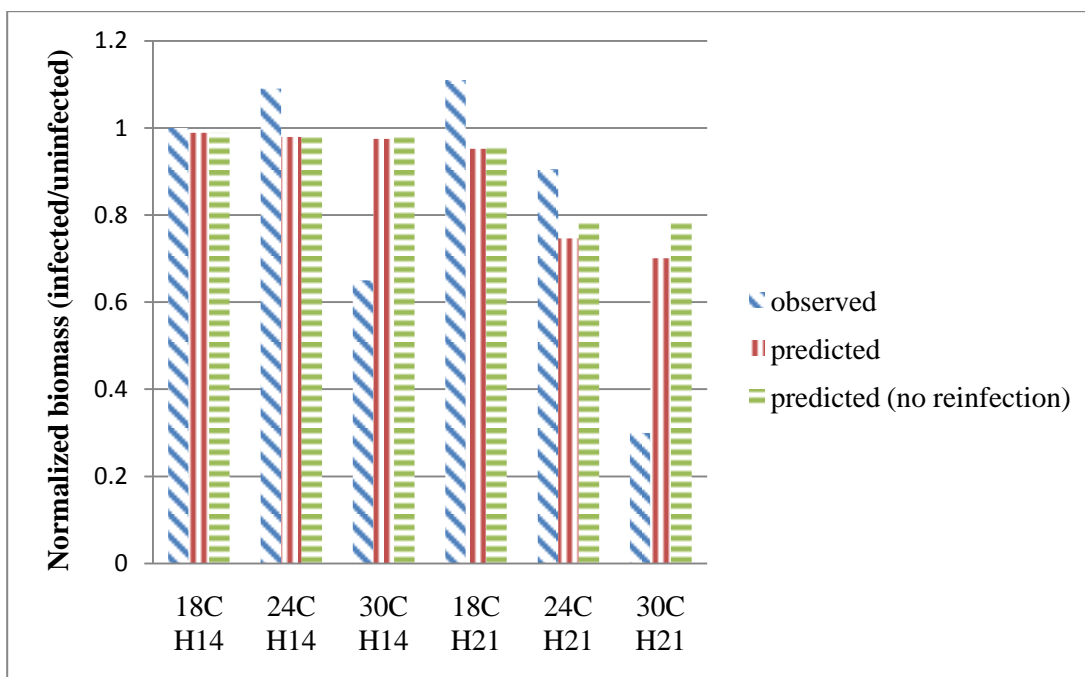


**Figure 6.10** Yield ratio (infected biomass/uninfected biomass value), fraction of roots infected (Frac infected), and fraction of roots dark brown/black Frac DB/B). From Katzman's observed temperature and zoospore concentration experiments (24 C and 25 zoos ml<sup>-1</sup>) and model predictions.

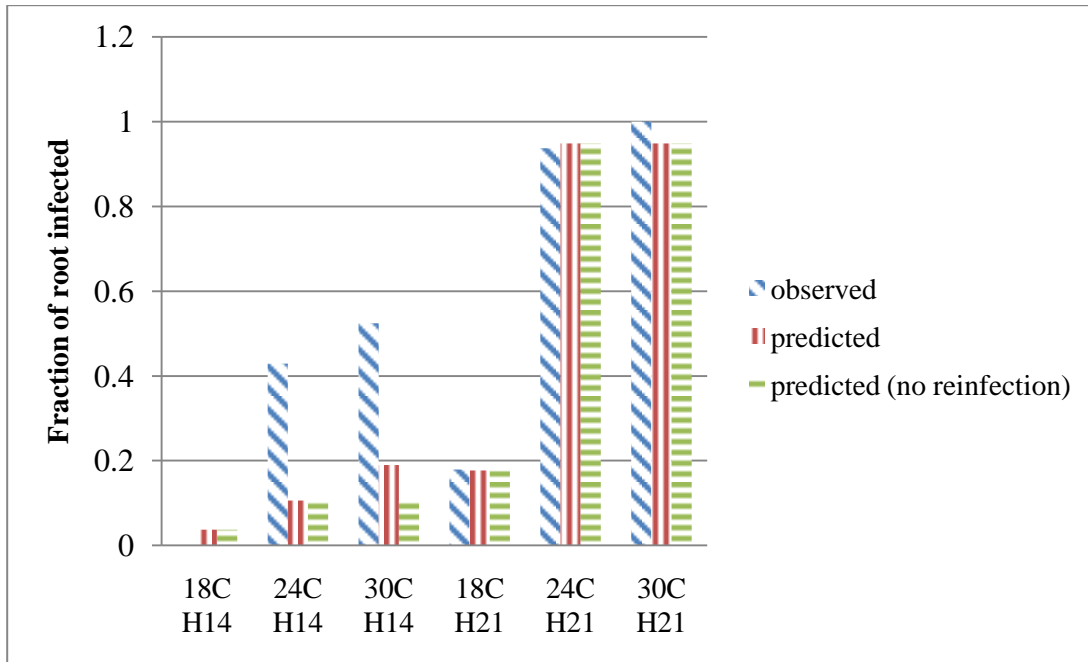
The model performs well with the harvest 14 and harvest 21 predictions of biomass, and fraction of roots dark brown/black, with predicted values falling between or close to the observed data. The prediction of fraction of the roots infected is also very good, with the predicted fraction falling between the two observed fractions for harvest 21. The prediction for the fraction infected at harvest 14 did not fall between the two observed fractions; however, it seems likely the observation for the temperature experiment is significantly different from the time of inoculation and concentration experiment values. The time of inoculation observation showed that

0.10 of the roots were infected at harvest 14, which agrees very closely with the concentration observation of 0.117 and the predicted value of 0.106.

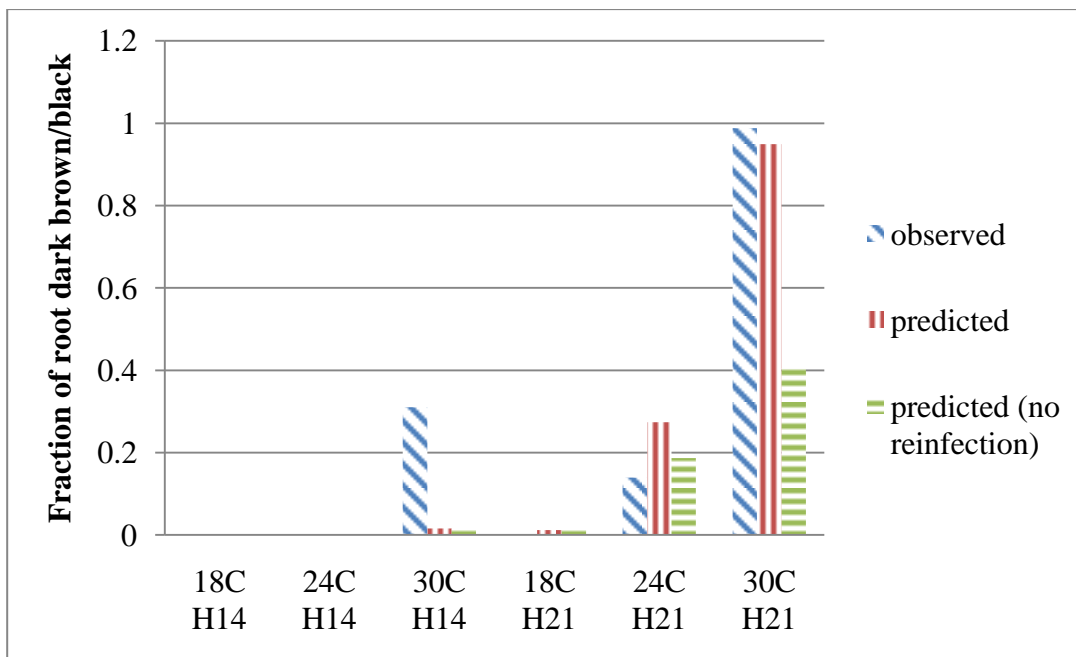
To validate the portion of the model concerned with the effects of nutrient solution temperature on the growth of mycelium and effect on crop biomass, the model was run with the same conditions as Katzman's nutrient solution temperature experiment. The results are presented in Figure 6.11, Figure 6.12 and Figure 6.13. Predicted values both with and without reinfection were calculated to illustrate the effect of secondary infection, particularly at higher temperatures.



**Figure 6.11** Yield ratio (infected biomass/uninfected biomass value) at harvest. From Katzman's nutrient solution temperature experiment and model predictions. Code is temperature followed by harvest day.



**Figure 6.12** Fraction of root infected. From Katzman's nutrient solution temperature experiment, and model predictions. Code is temperature followed by harvest day.



**Figure 6.13** Fraction of root dark brown/black. From Katzman's nutrient solution temperature experiment, and model predictions. Code is temperature followed by harvest day.

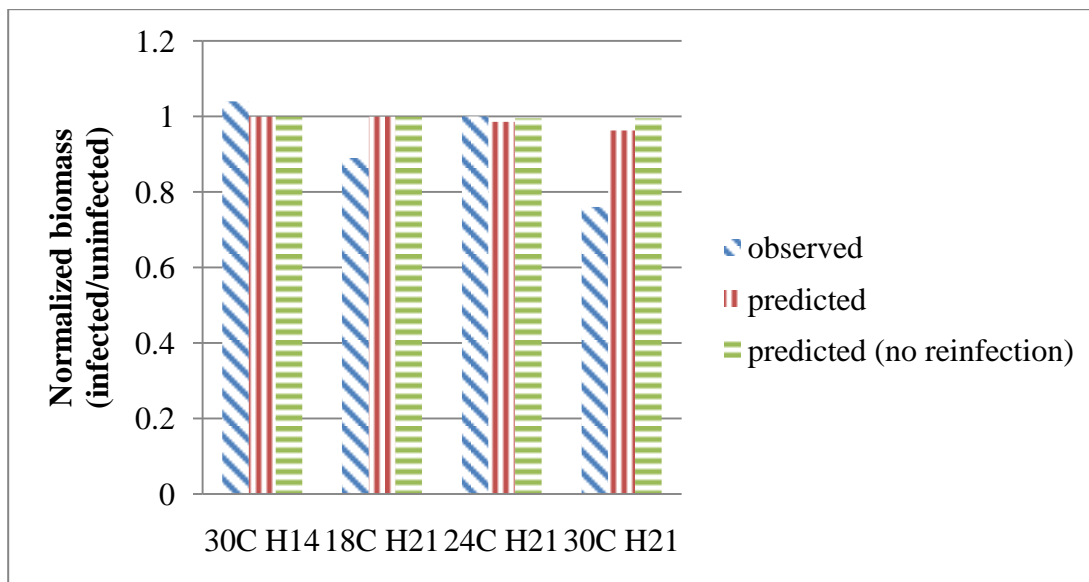
The model reasonably predicts harvest biomasses (Figure 6.11) for the two lower temperature conditions, 18 and 24C, underestimating them by 1 to 2% at harvest 14, and 4 and 15% at harvest day 21 (assuming the observed harvest biomass fractions at 24C Harvest 14 and 18C harvest 21 are 1 and not 1.09 and 1.11 respectively). However, for the 30C harvest day 14 condition it predicted a harvest biomass fraction of 0.975, as compared to an observed biomass fraction of 0.65. The 30C harvest 21 condition prediction was even worse with a predicted fraction of 0.701 versus an observed fraction of 0.3. There was very little difference between the predictions with and without reinfection.

The model predicted well the fraction of the roots infected (Figure 6.12) at harvest 21, for all three temperature conditions. However, it overestimated the values at harvest 14 for both the 24 and 30C condition (though as has been described previously the observed value is quite different and so the prediction is actually closer to the other observed values. The 30C fraction was considerably underestimated (33%).

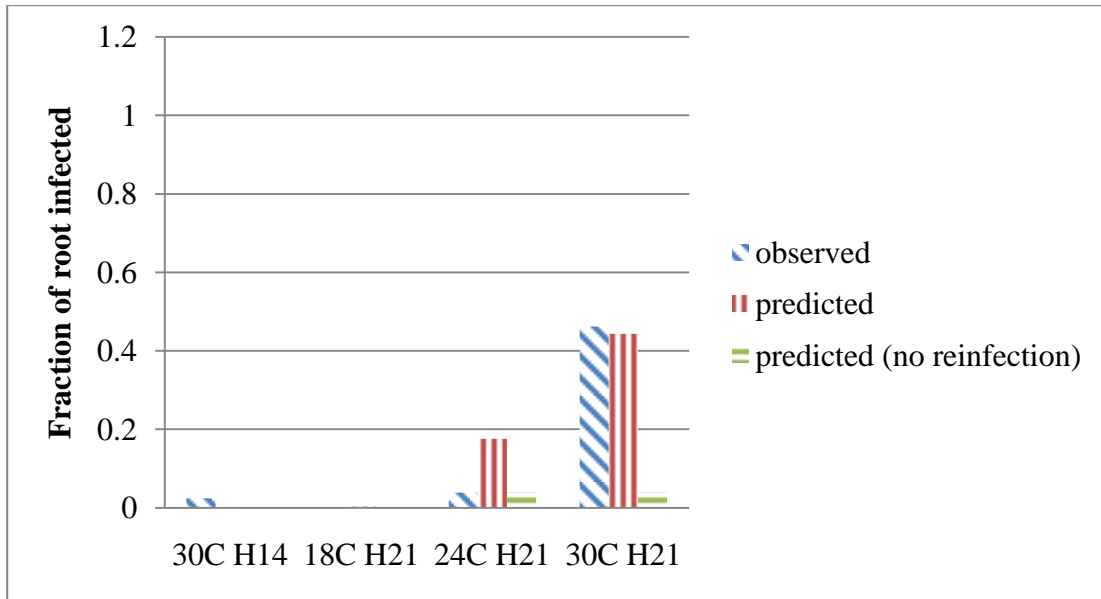
The model also predicted quite well the fraction of the roots dark brown/black (Figure 6.13) for the 18 and 24C temperature conditions at both harvest 14 and 21. The model also significantly underestimated the values for the harvest day 14, 30C condition, however the day 21 condition only differed by 4% (though this level is close to the upper limit the model permits).

In these results there was little difference between the predictions using reinfection and predictions without. This lack of difference is because the dose of 25 zoospores ml<sup>-1</sup> was large enough that it reduced the possibility for further zoospores to cause any more damage; second generation zoospores could not make things much worse.

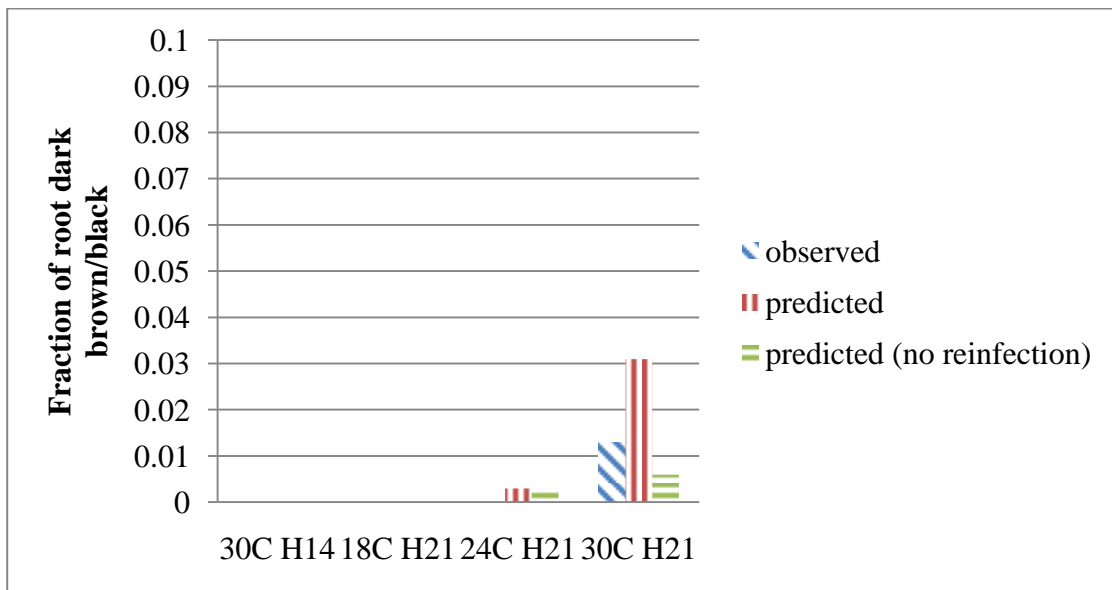
A reason for the error in the 30C predictions is the model was based on the uninoculated effects at 30C, which were felt to be more representative of conditions likely to be found in commercial production. An inoculation dose of 25 zoospores ml<sup>-1</sup> would not occur in a commercial setting and so it is of less concern the model does not predict these effects as well as other ranges. To illustrate the temperature performance of the model under more natural conditions, it was compared to the observed uninoculated control ponds Katzman used in her temperature experiments (Figures 6.14, 6.15 and 6.16).



**Figure 6.14** Yield ratio (infected biomass/uninfected biomass value) at harvest. From Katzman's uninoculated nutrient solution temperature experiment, and model predictions. Code is temperature followed by harvest day.



**Figure 6.15** Fraction of root infected. From Katzman's uninoculated nutrient solution temperature experiment, and model predictions. Code is temperature followed by harvest day.



**Figure 6.16** Fraction of root dark brown/black. From Katzman's uninoculated nutrient solution temperature experiment, and model predictions. Code is temperature followed by harvest day. (Note scale is different from other figures.)

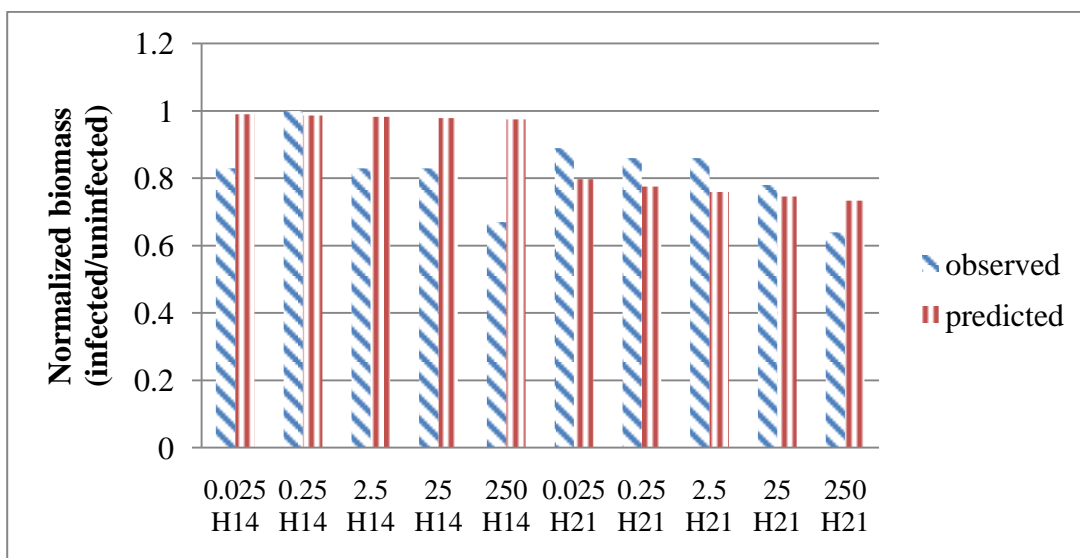
The performance of the model is better under this comparison. Predictions of the fraction of biomass is good for all the harvest day 14 predictions (18 and 24C

conditions were not shown as there was no *Pythium* damage apparent at that point). Harvest day 21 predictions are also good for the 18 and 24 C temperatures, however, the biomass fraction for the 30 C condition was overestimated by 20%.

The fraction of roots infected was a good fit, although this is also due to the fact some of the parameters used to form the model were taken from these observations. It is clear from Figure 6.15, the effects of reinfection are very important in high temperature conditions.

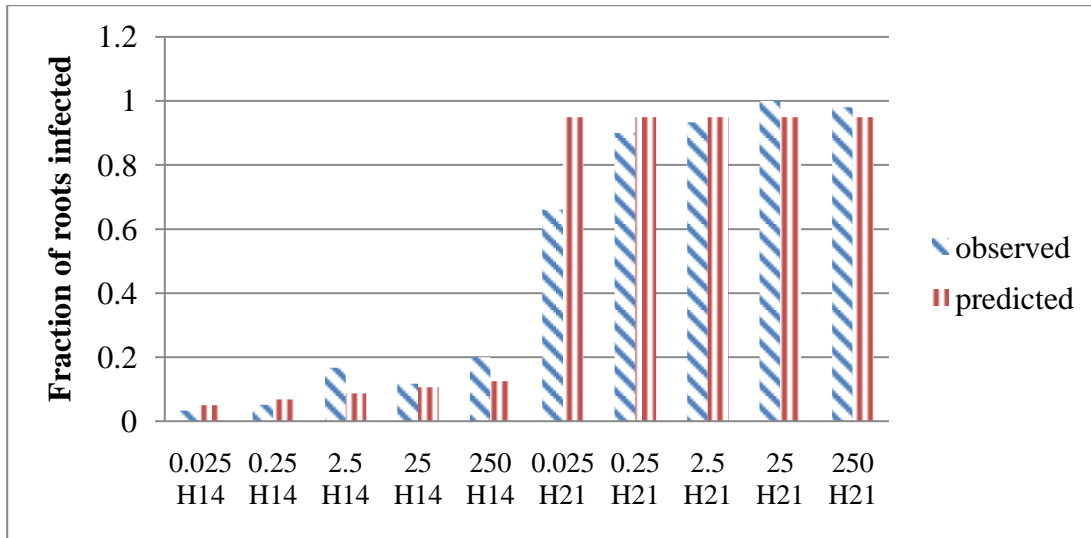
The fraction of roots that are dark brown/black are also predicted very well under all conditions, though this is partially due to the fact the values are low compared to other inoculated infections (note scale of Figure 6.16 is adjusted to assist in making comparisons).

To validate the portion of the model concerned with the effects of inoculum concentration effects on the growth of mycelium and effect on crop biomass, the model was run with the same conditions as Katzman's inoculation dosage experiment. The results are presented in Figure 6.17, Figure 6.18 and Figure 6.19.

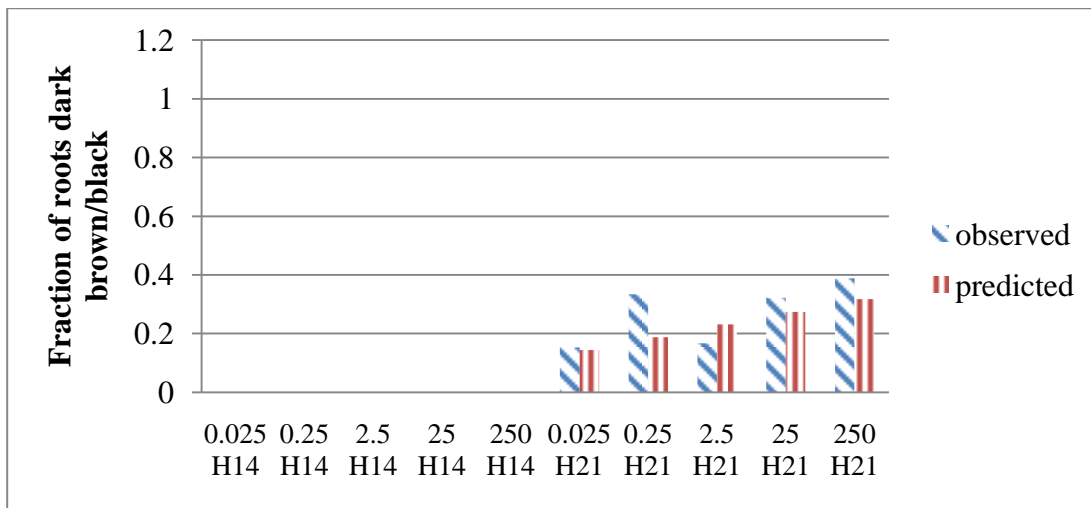


**Figure 6.17** Yield ratio (infected biomass/uninfected biomass value) at harvest. From Katzman's inoculation concentration experiment, and model predictions. Code is zoospore concentration (zoos ml<sup>-1</sup>) followed by harvest day.





**Figure 6.18** Fraction of roots infected. From Katzman's inoculation concentration experiment, and model predictions. . Code is zoospore concentration (zoos ml<sup>-1</sup>) followed by harvest day.



**Figure 6.19** Fraction of root dark brown/black. From Katzman's inoculation concentration experiment, and model predictions. . Code is zoospore concentration (zoos ml<sup>-1</sup>) followed by harvest day.

The model overpredicts harvest biomass at all concentrations of starting inoculum for harvest 14 (with the exception of 0.25 zoos ml<sup>-1</sup>), whereas, it underestimates harvest biomasses for harvest day 21 with the exception of the highest concentration, 250 zoos ml<sup>-1</sup> where the estimate is 0.094 higher. Furthermore, the

model does not demonstrate as much of an effect of concentration, as is apparent in the observed data. The observed biomass data of harvest 14 is difficult to use as any differences (but for the highest concentration) were within the experimental error. For the harvest 21 observed data, there is a difference of 0.25 between the lowest and highest concentrations of inoculation, whereas, the model only predicts a difference of 0.104.

The model is better at predicting the fraction of root infected. It slightly over and underpredicts the fraction of roots that are infected for the day 14 harvest. For the day 21 harvest it is close in all concentrations but for the lowest where it overestimates the fraction of roots infected by 0.288. As with biomass, the differential between the highest and lowest concentrations are smaller in the predicted than the observed data. The observed data had a differential of 0.167 as compared to a prediction of 0.075 in the harvest day 14 conditions. In harvest day 21 conditions, excluding the lowest concentration, the observed differential was 0.08 (0.32 with the lowest concentration) whereas, the model predicted no difference between the highest and lowest concentrations, as the prediction was at the model limit of 0.95 infection.

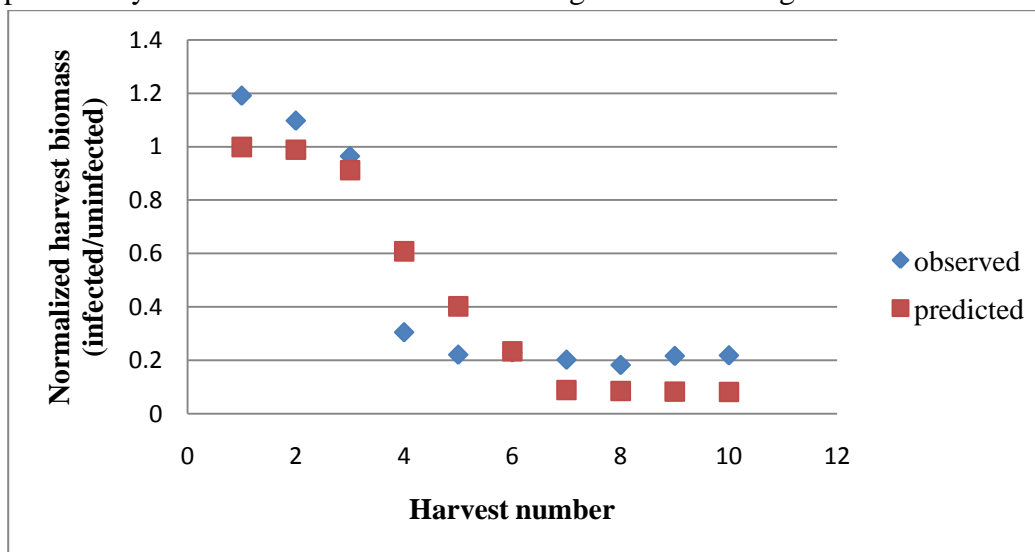
For the fraction of the root that is dark/brown black, the model performed well for the harvest day 14 data, where no material was observed to be dark/brown black in any condition, and the model predicted the same ( $< 0.01$ ). For harvest day 21 predictions, the model underpredicted the fraction (but for the 2.5 zoos  $\text{ml}^{-1}$  condition) and increased from 0.144 at 0.025 zoos  $\text{ml}^{-1}$  to 0.317 for the 250 zoos  $\text{ml}^{-1}$  condition. The observed fractions ranged from 0.153 to 0.388.

#### *Validation of multi cohort Pythium disease model parameters*

The multi-cohort performance of the disease model was validated with data from the purposely inoculated conditions presented in chapter 3. Two sets are used;

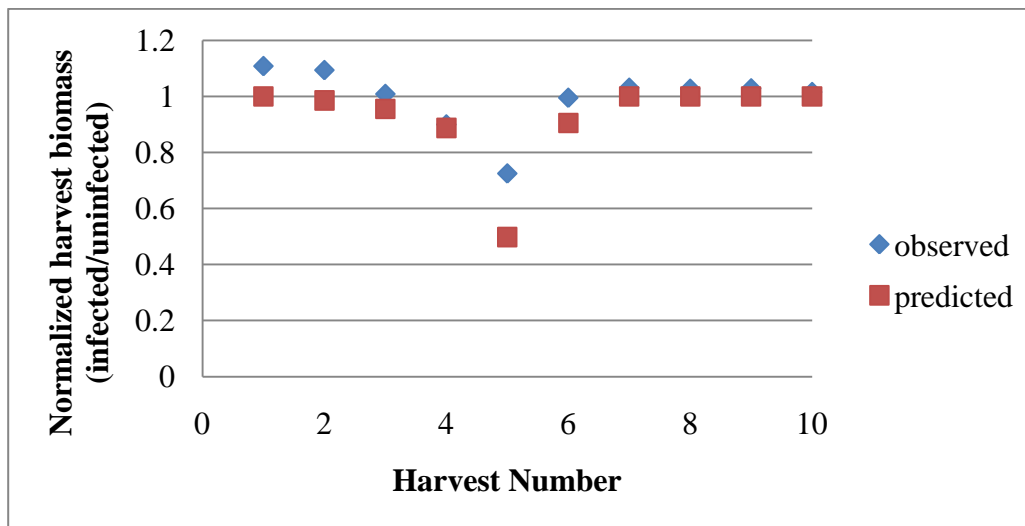
high temperature inoculation, and temperature suppression inoculation. In the high temperature inoculation, the system was run with a nutrient solution of 27.5C until a stand of 5 cohorts (aged 1, 4, 7, 10 and 13 days from floatation) was established. At this time, an inoculum dosage of 100 zoos ml<sup>-1</sup> was added. The plants were harvested when each cohort was 14 days old, and new cohorts were added every three days. The temperature suppression examination started out the same, however, 24 hours after inoculation with 100 zoos ml<sup>-1</sup>, the temperature of the nutrient solution was lowered from 27.5C to 20C. The harvesting and addition of new cohorts followed the same pattern as the high temperature inoculation set.

To validate the multi cohort portion of the *Pythium* disease model, the conditions used in these two studies were input into the disease model, which was extended to handle multiple cohorts in a common pond of nutrient solution. The observed experimental results were normalized by dividing the infected harvest biomasses by the corresponding uninfected control biomasses and plotted with the predicted yield ratio harvest biomasses in Figure 6.20 and Figure 6.21.



**Figure 6.20** Yield ratio (infected biomass/uninfected biomass value) for a warm temperature nutrient solution (27.5C) inoculated with 100 zoos ml<sup>-1</sup> for one sequence of harvests. (A new cohort was added every 3 days, harvested after 14 days for a total experimental time of 41 days.)

In Figure 6.20, the predicted harvest biomasses follow the same trend as the observed data, showing a steep decline followed by a stable harvest biomass amount as the disease stabilizes in the crop. Differences between observed and modeled data are the model predicts a less severe drop off, and the level of disease settled out to about one third of that observed, but close in absolute values.



**Figure 6.21** Yield ratio of biomasses (infected/uninfected) for an initially warm temperature nutrient solution (27.5C) inoculated with 100 zoos ml<sup>-1</sup>, switched to a cold (20C) temperature nutrient solution, 24 hours after inoculation.

The performance of the model to predict the response of the system to a switch to a cold nutrient solution temperature (Figure 6.21) appears to be very good. Though the model predicts a higher level of damage in the fifth harvest material, it correctly predicts the harvest biomasses of material not directly exposed to the initial inoculation dose would be close to the uninoculated control. However, the model still predicts some level of damage in the crop as the harvest six biomasses do not immediately return to an uninfected state as would be represented by a normalized value of one. As in the high temperature inoculation simulation shown in Figure 6.20, the differences of Figure 6.21 may also be explained by temperature. The reduction at harvest 5 is the youngest material present in the system when the inoculation was

applied. It was exposed to high temperatures in the presence of a high inoculation dosage and, as in the situation previously described, the model predicted more damage than was observed. The fact the harvests did not return to a completely uninfected state following the dropping of the temperature could be due to the fact the cool temperature applied was 20C (again, not a temperature Katzman used). From the Katzman data it is impossible to say what the safe cut-off temperature would be for a 14 day crop cycle. A temperature of 18 appears to be safe, but as observed in the multi-cohort work 20C, also appears adequate.

A further check on the functionality of the model is how the length of the crop cycle and temperature of the nutrient solution, affect the number of harvests before the harvested material is too badly damaged by *Pythium* to sell (Table 6.8). This measure is different from merely observing the drop in normalized biomass as was done in figures 6.20 and 6.21. Though biomass may not be largely affected, if the roots are 10 to 15% infected, there is some wilting present in the crop, which in turn makes the material unsaleable. To determine the values presented in Table 6.9, the model was run with different crop lengths, and nutrient solution temperatures. Harvests were counted as successful, if they had less than 15% of their roots infected.

**Table 6.9** Number of harvests (based on a 3 day harvest schedule in a continuous production pond) before crop material is too badly damaged by *Pythium* to market, as a function of nutrient solution temperature. Grown at 18 to 32 C for 14 to 22 days.

		Temperature C							
		18	20	22	24	26	27.5	30	32
Crop Length (days)	14	>50	>50	>50	>50	>50	>50	>50	3
	16	>50	>50	>50	>50	>50	>50	2	1
	18	>50	>50	>50	>50	4	2	1	0
	20	>50	>50	>50	>50	1	0	0	0
	22	>50	>50	>50	0	0	0	0	0

The trend presented in Table 6.9 is as expected. As the length of time the crop spends in the pond increases (increasing crop length), there is increased time for reproduction and release of zoospores to newer cohorts of the crop. Increasing nutrient solution temperature accelerates this process, and at higher temperatures the number of successful harvests before collapse is reduced. Because of the form of the model, it appears to be very sensitive to critical points, where below a specific temperature infections do not develop, and only a degree or half a degree higher and major problems result. This sensitivity is also apparent in the results of table 6.8 where several combinations of temperature and crop length result in no crop losses (at least in the observed period of 50 harvests). This lack of outbreaks does not seem as plausible as other results of the model. However, the model is assuming growing and disease conditions that are unlikely to exist in real world production. Constant temperatures, and constant disease and crop growth factors are an assumption of the model that may not be satisfied in the real world. In addition, it is assumed the crop is never under any stresses, which could make it more susceptible to infection. Though the results presented in Table 6.9 may not exactly correspond to what one would expect, the trends presented do, and as such the model may be useful for making comparisons of various production techniques.

#### 6.4.2 Sensitivity analysis of *Pythium* disease model parameters

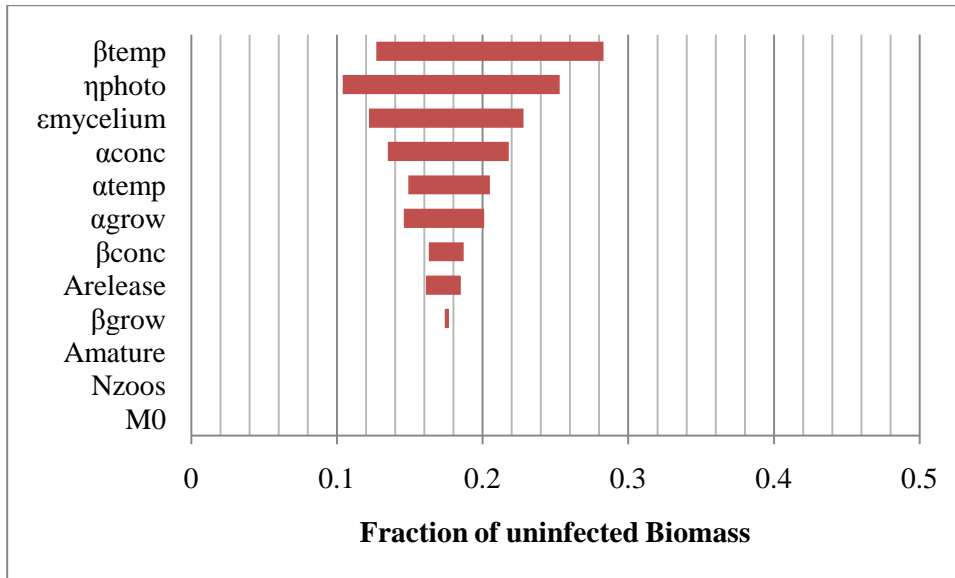
By plotting the parameters of the model in tornado style, it is possible to see the effect of each parameter on the final outcome. To generate the tornado plots, the model was run twice for each parameter; once with the value of the parameter increased 10% and once with the value decreased 10%. The output of the model was recorded for each run, and then summarized in tornado plots. A complicating factor is the model is developed to function from a number of different starting points and

different conditions. Because certain variables become more important under different conditions it was necessary to create several tornado plots.

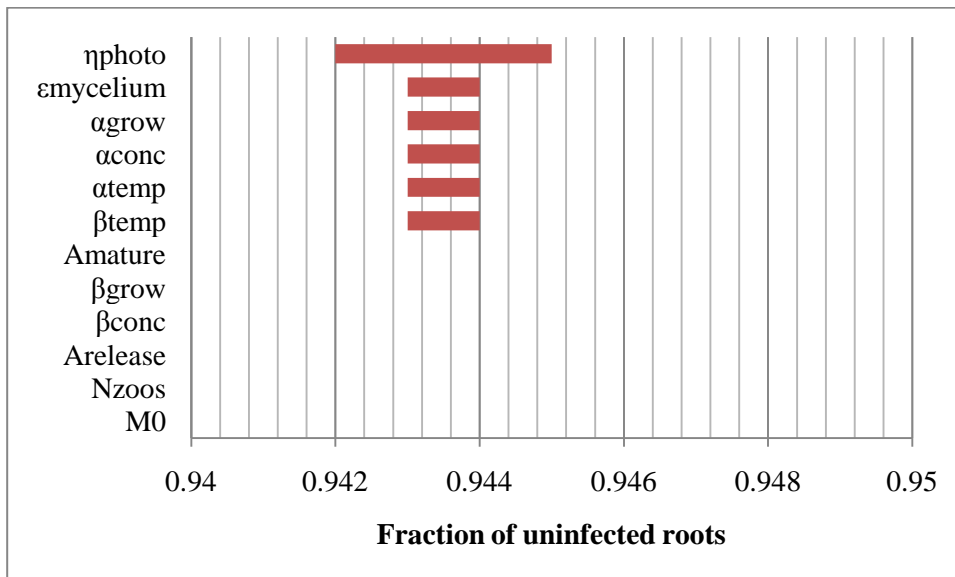
The parameters examined are:

1.  $\beta_{\text{conc}}$ , Concentration effect slope
2.  $\alpha_{\text{conc}}$ , Concentration effect intercept
3.  $\epsilon_{\text{mycelium}}$ , Reference amount of mycelium @ 25 zoos ml<sup>-1</sup>
4.  $M_0$ , Starting Mass of mycelium
5.  $\beta_{\text{grow}}$ , Mycelial growth rate slope
6.  $\alpha_{\text{grow}}$ , Mycelial growth rate intercept
7.  $A_{\text{mature}}$ , Maturation age of mycelium
8.  $\eta_{\text{photo}}$ , Coefficient of Photosynthetic Rate reduction
9.  $\beta_{\text{temp}}$ , Temperature effect slope
10.  $\alpha_{\text{temp}}$ , Temperature effect intercept
11.  $A_{\text{release}}$ , Zoospore release age
12.  $N_{\text{zoos}}$ , Zoospore release amount

To examine the effect of each variable under the conditions used to develop the model in the first place, 15 tornado plots were necessary. These plots represent the output of the model with two inoculation days (day 1 and day 9), three harvest days (days 9, 14 and 21) and for the three output values of the model, fraction of uninfected biomass, fraction of the roots uninfected, and fraction of the roots dark brown/black. The day 9 inoculation and day 9 harvest combination were dropped as no significant effects of *Pythium* were observed. The results of this examination are shown in Figure 6.22 to Figure 6.36.

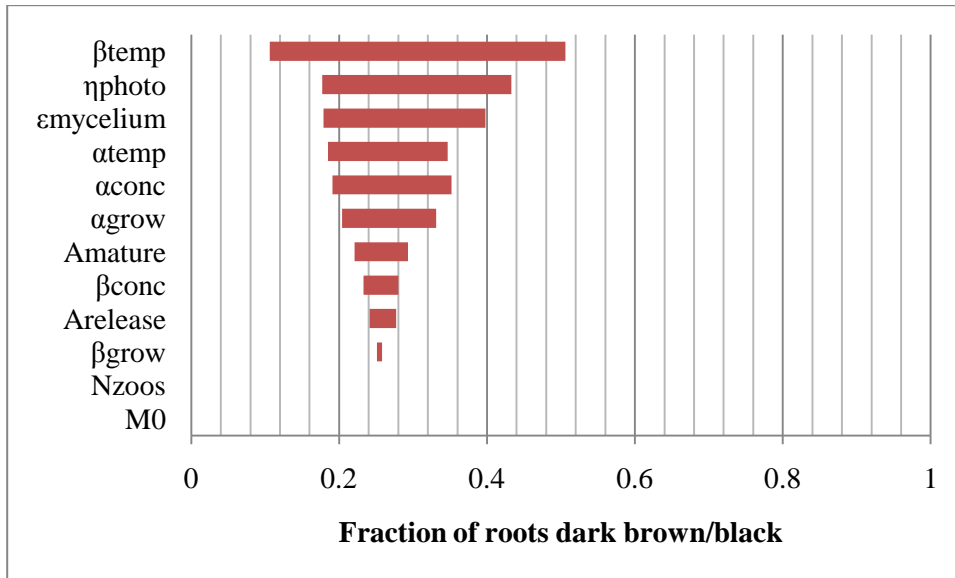


**Figure 6.22** Sensitivity of the fraction of uninfected biomass to model parameters with inoculation on day 1 and harvest on day 9 with a nutrient solution of 24 C.

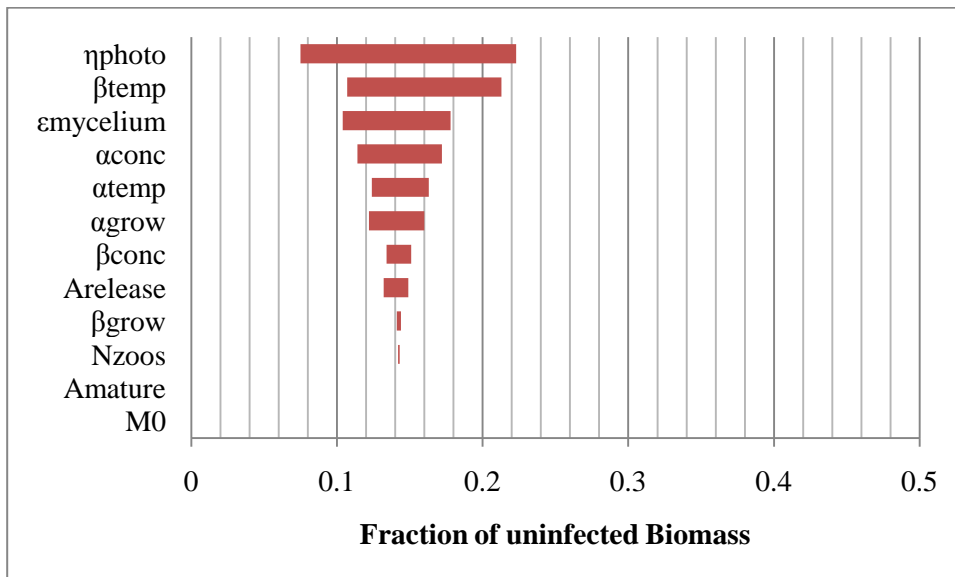


**Figure 6.23** Sensitivity of the fraction of uninfected roots to model parameters with inoculation on day 1 and harvest on day 9 with a nutrient solution of 24 C.

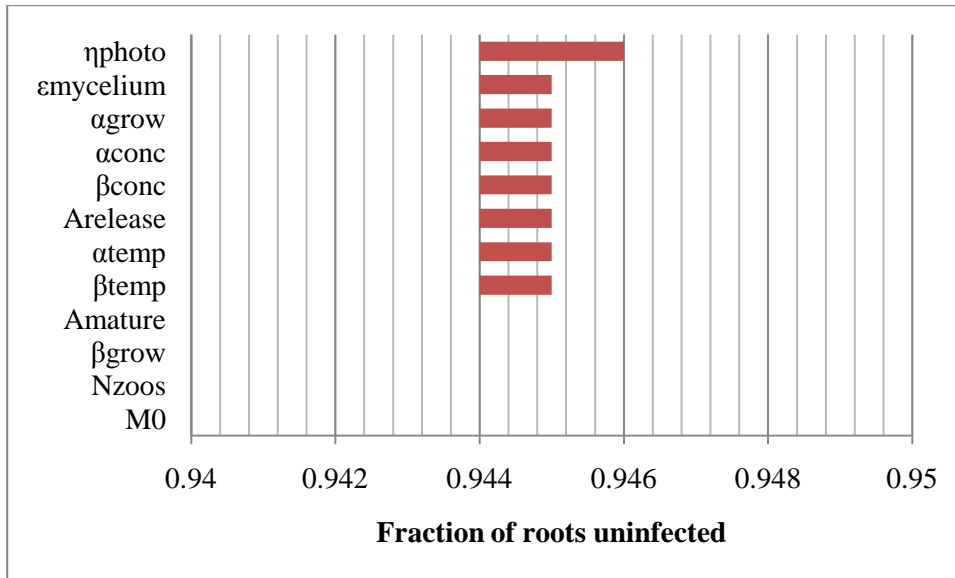




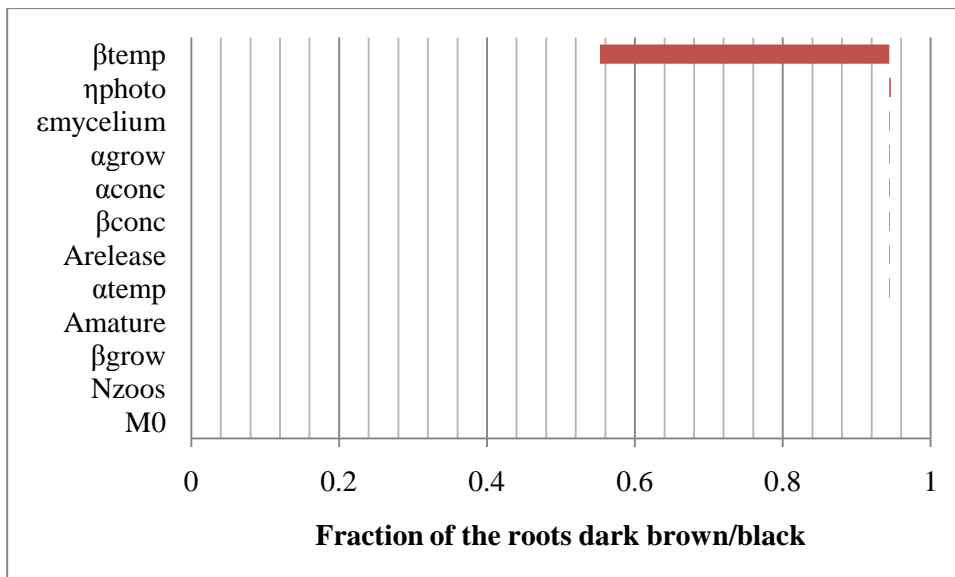
**Figure 6.24** Sensitivity of the fraction of roots dark brown/black to model parameters with inoculation on day 1 and harvest on day 9 with a nutrient solution of 24 C.



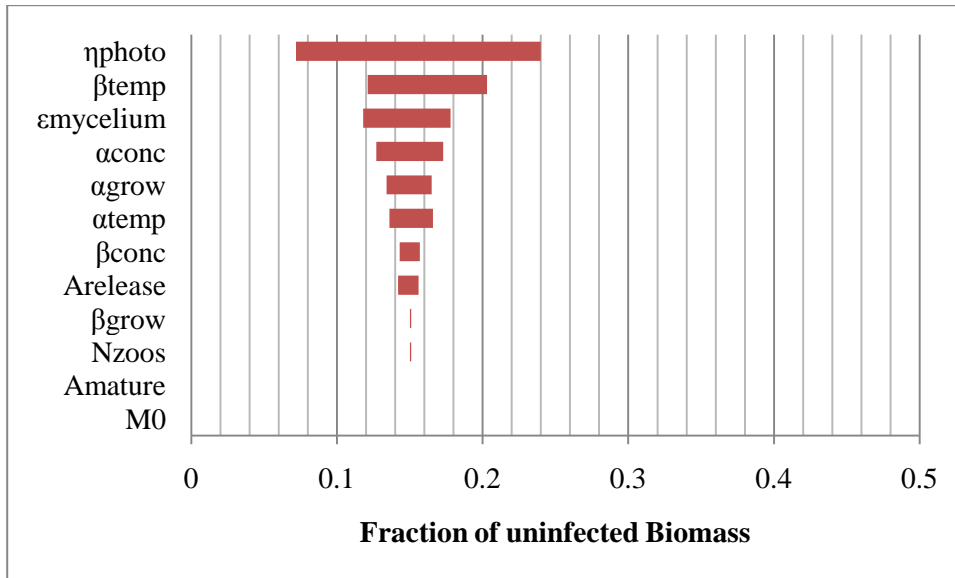
**Figure 6.25** Sensitivity of the fraction of uninfected biomass to model parameters with inoculation on day 1 and harvest on day 14 with a nutrient solution of 24 C.



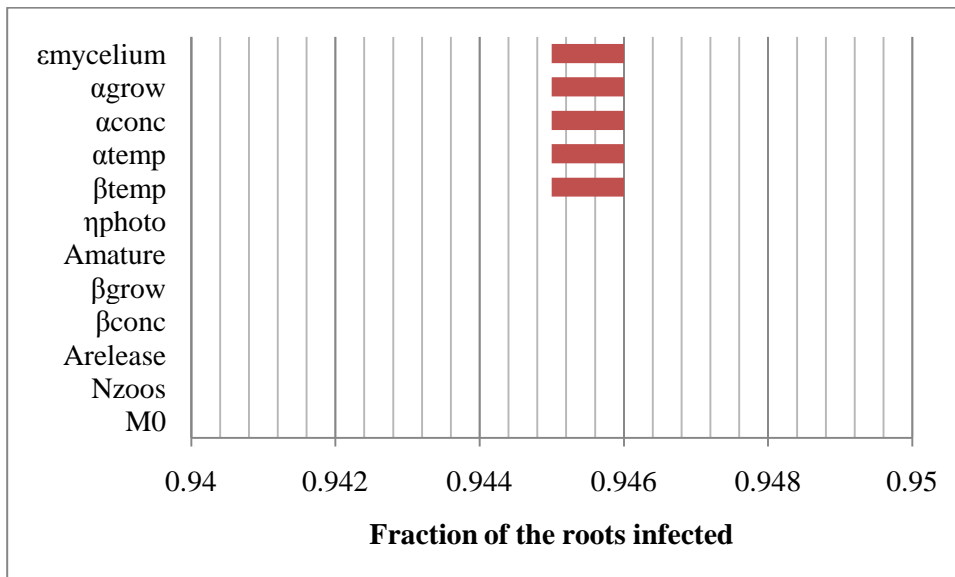
**Figure 6.26** Sensitivity of the fraction of uninfected roots to model parameters with inoculation on day 1 and harvest on day 14 with a nutrient solution of 24 C.



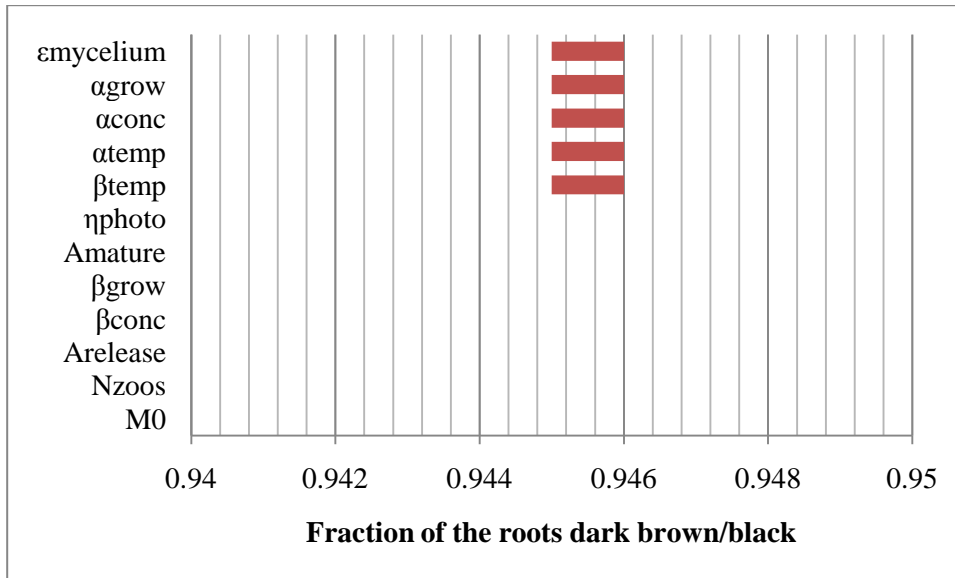
**Figure 6.27** Sensitivity of the fraction of roots dark brown/black to model parameters with inoculation on day 1 and harvest on day 14 with a nutrient solution of 24 C.



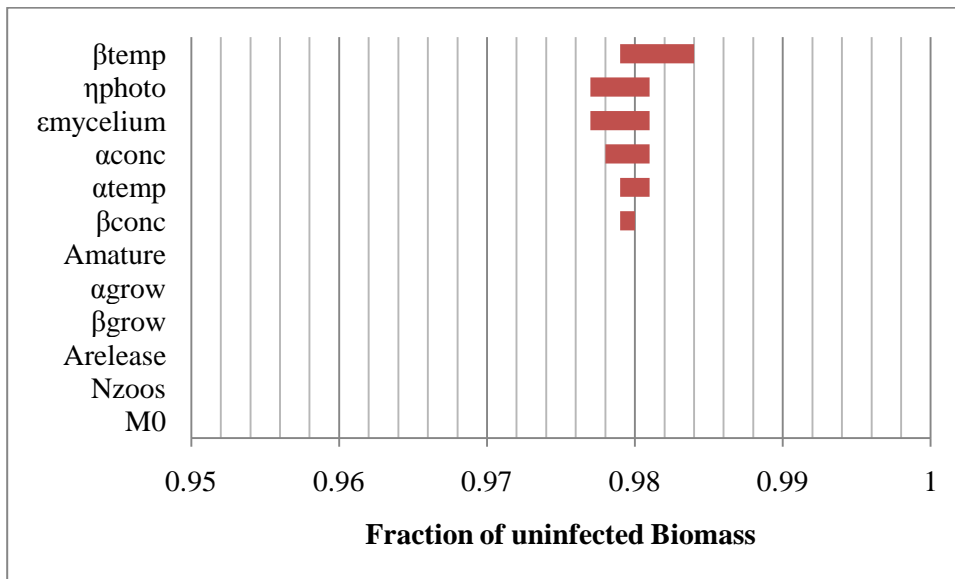
**Figure 6.28** Sensitivity of the fraction of uninfected biomass to model parameters with inoculation on day 1 and harvest on day 21 with a nutrient solution of 24 C.



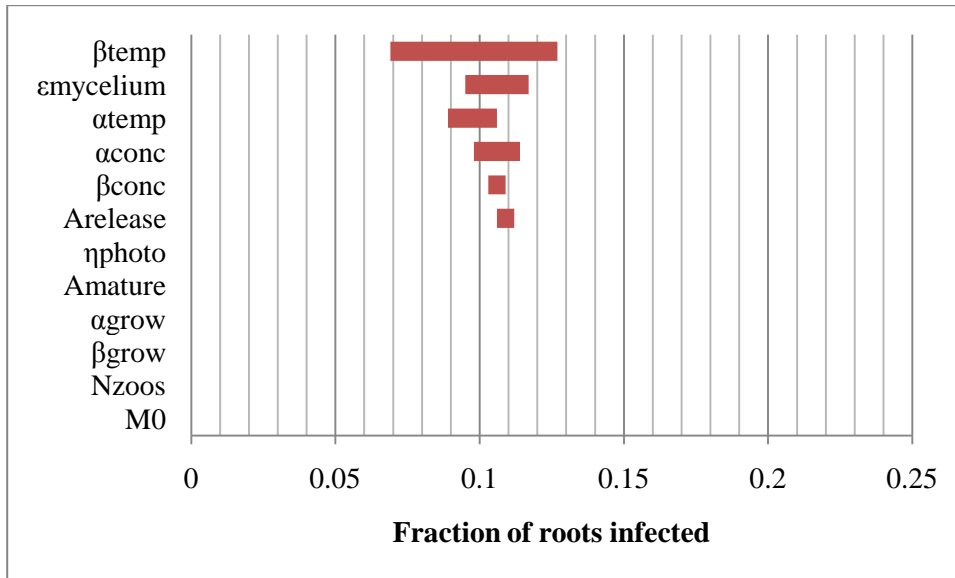
**Figure 6.29** Sensitivity of the fraction of uninfected roots to model parameters with inoculation on day 1 and harvest on day 21 with a nutrient solution of 24 C.



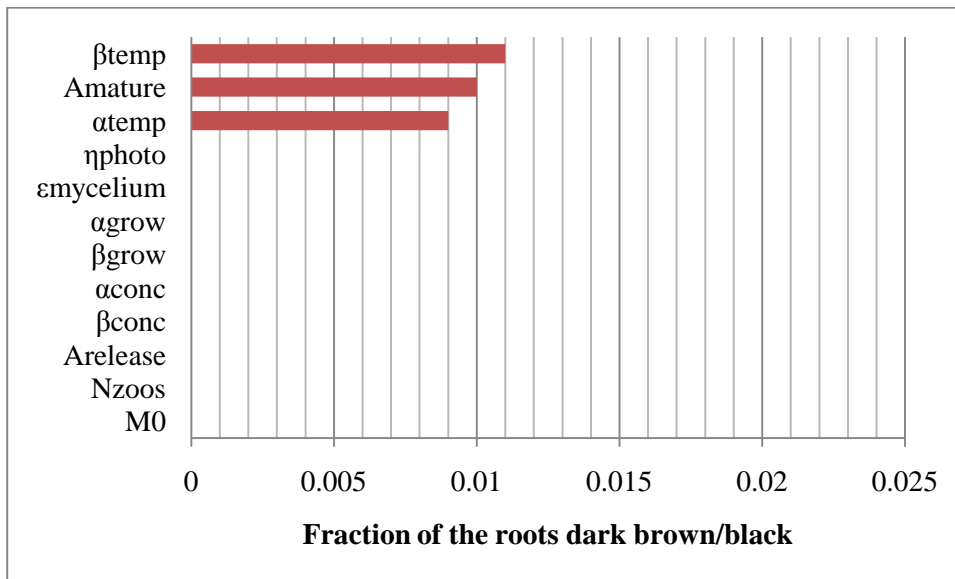
**Figure 6.30** Sensitivity of the fraction of roots dark brown/black to model parameters with inoculation on day 1 and harvest on day 21 with a nutrient solution of 24 C.



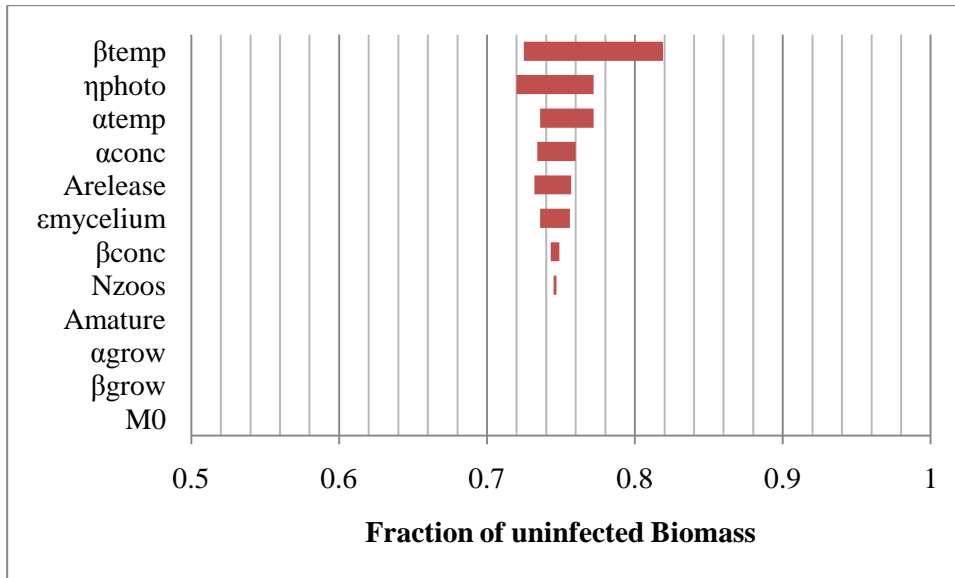
**Figure 6.31** Sensitivity of the fraction of uninfected biomass to model parameters with inoculation on day 9 and harvest on day 14 with a nutrient solution of 24 C.



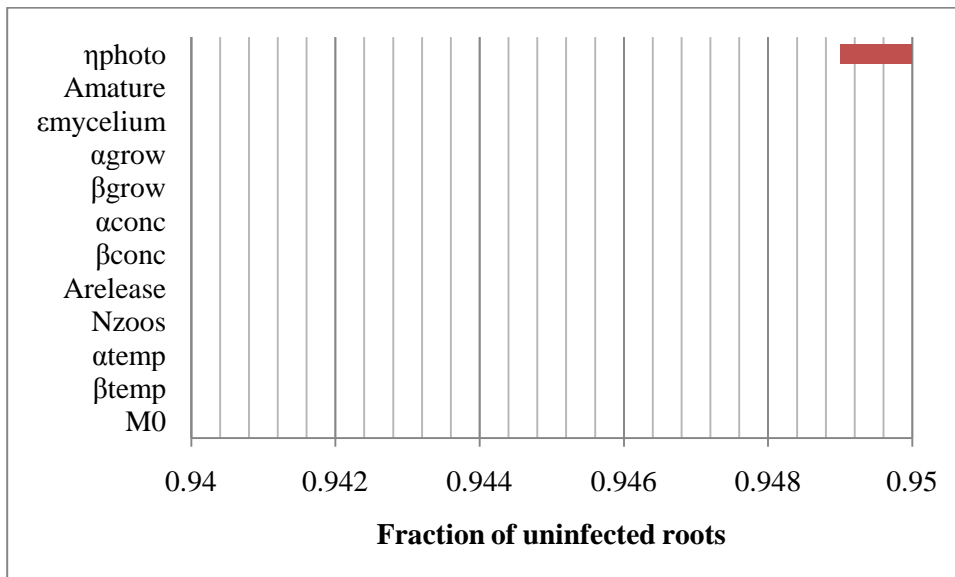
**Figure 6.32** Sensitivity of the fraction of uninfected roots to model parameters with inoculation on day 9 and harvest on day 14 with a nutrient solution of 24 C.



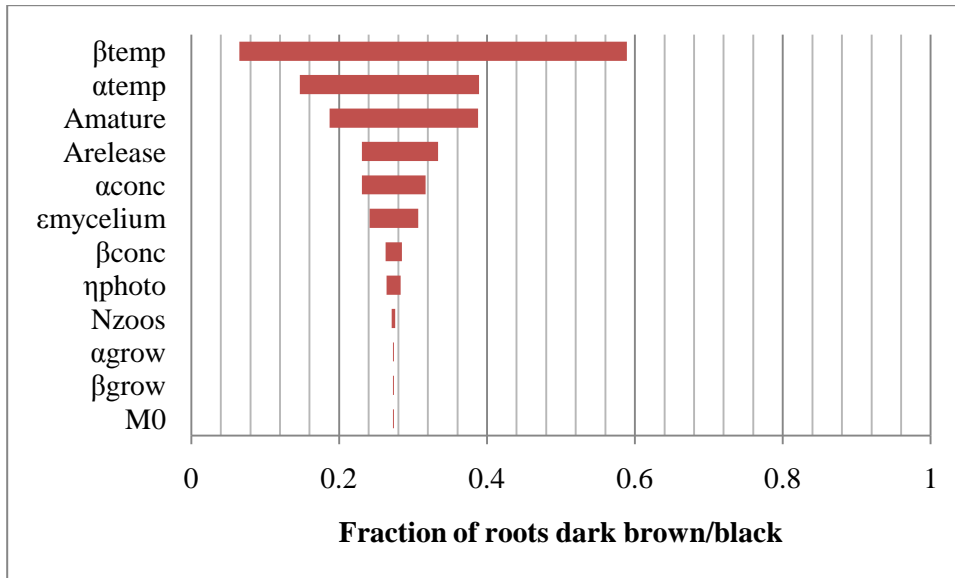
**Figure 6.33** Sensitivity of the fraction of roots dark brown/black to model parameters with inoculation on day 9 and harvest on day 14 with a nutrient solution of 24 C.



**Figure 6.34** Sensitivity of the fraction of uninfected biomass to model parameters with inoculation on day 9 and harvest on day 21 with a nutrient solution of 24 C.



**Figure 6.35** Sensitivity of the fraction of uninfected roots to model parameters with inoculation on day 9 and harvest on day 21 with a nutrient solution of 24 C.



**Figure 6.36** Sensitivity of the fraction of roots dark brown/black to model parameters with inoculation on day 9 and harvest on day 21 with a nutrient solution of 24 C.

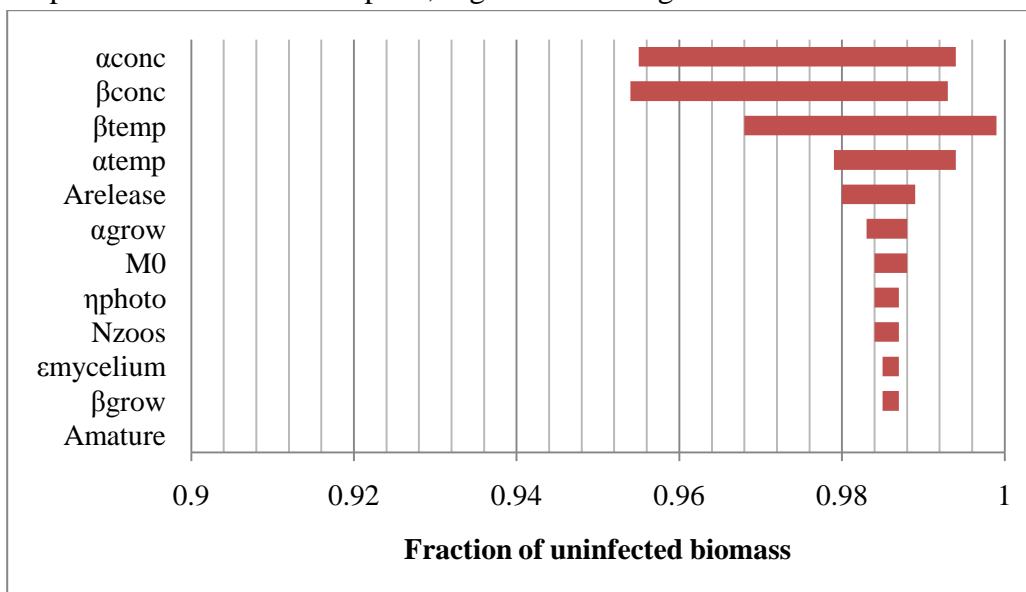
It is not surprising the variable to which the model is the most sensitive is the slope of the temperature effects,  $\beta_{temp}$ . In the model, it is assumed the temperature affects not only the growth rate of the mycelia, but the developmental rate as well. The intercept of the temperature effects,  $\alpha_{temp}$ , is also important, though it is not as powerful as the slope,  $\beta_{temp}$ .

Another expected effect is the sensitivity of the model to the coefficient of Photosynthetic Rate reduction,  $\eta_{photo}$ , particularly examining the fraction of the uninfected biomass.  $\eta_{photo}$  relates the amount of infection in the roots to the reduction in photosynthate production, which directly affects the accumulation of plant biomass.

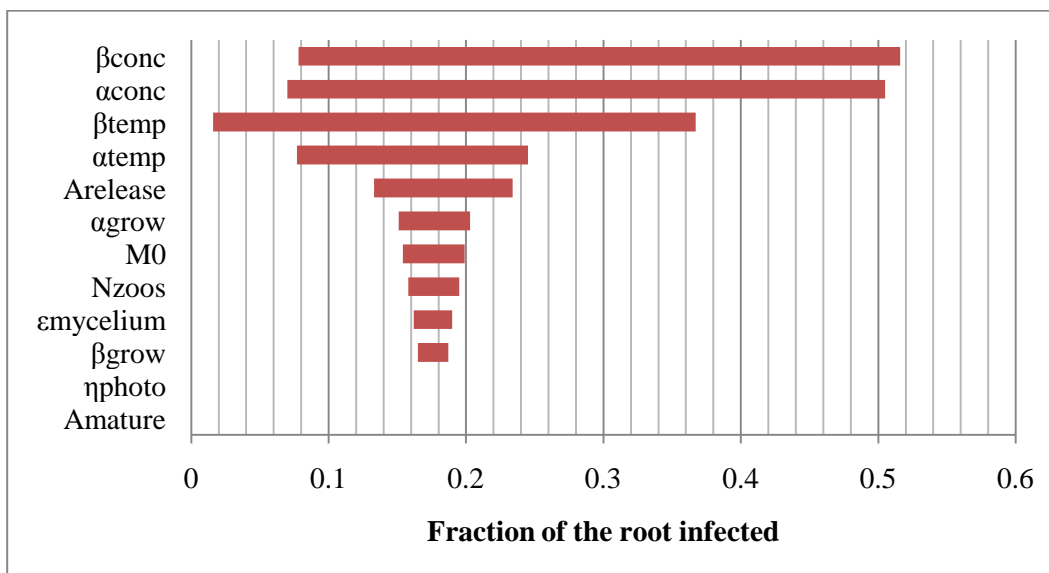
The initial amount of mycelium,  $M_0$ , appears to have no effect on the model at all. However, this lack of effect is because these plots examined conditions in which the crop was inoculated with a large single dose of zoospores, causing massive damage and overwhelming any effect the natural inoculum present might have had.

To examine the sensitivity of the model to parameters under conditions more likely to be found in a commercial setting, the sensitivity analysis was run with no

inoculation at 24 and 30C; conditions under which Katzman observed the development of *Pythium* damage. The observed (and predicted) damage was only apparent in harvest day 21 material. The results of this part of the sensitivity analysis are presented in six tornado plots, Figure 6.37 to Figure 6.42.

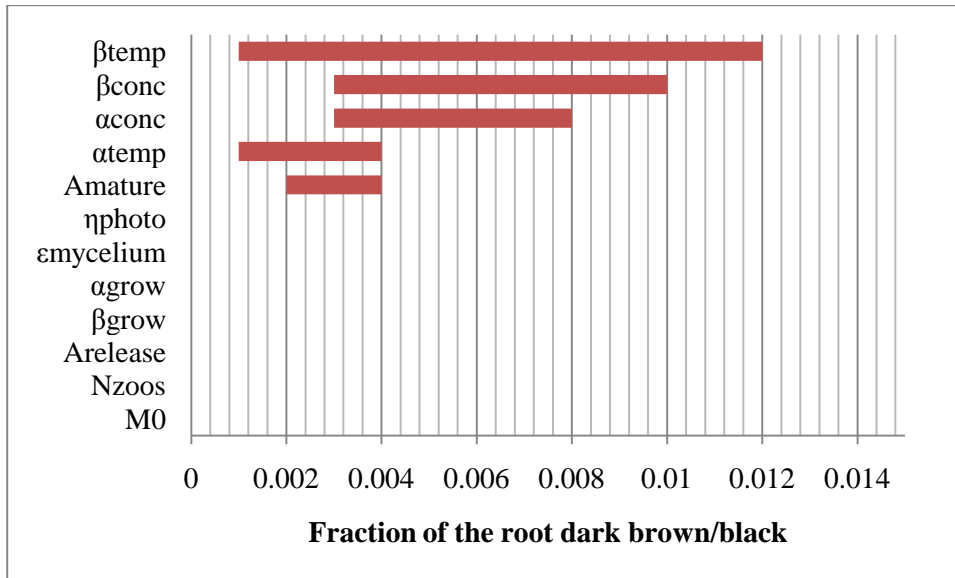


**Figure 6.37** Sensitivity of the fraction of uninfected biomass to model parameters at harvest day 21 with no inoculation and a nutrient solution of 24 C.

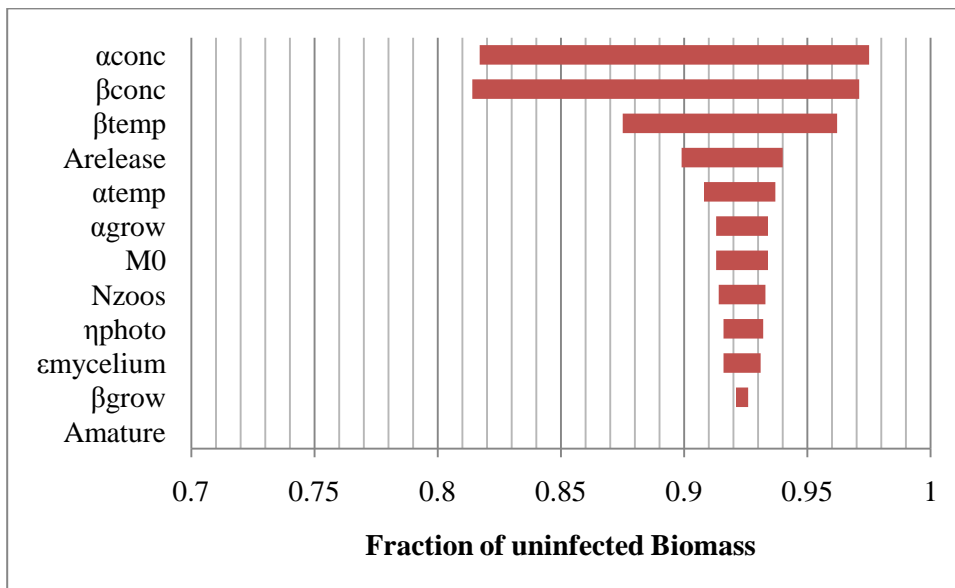


**Figure 6.38** Sensitivity of the fraction of the root infected to model parameters at harvest day 21 with no inoculation and a nutrient solution of 24 C.

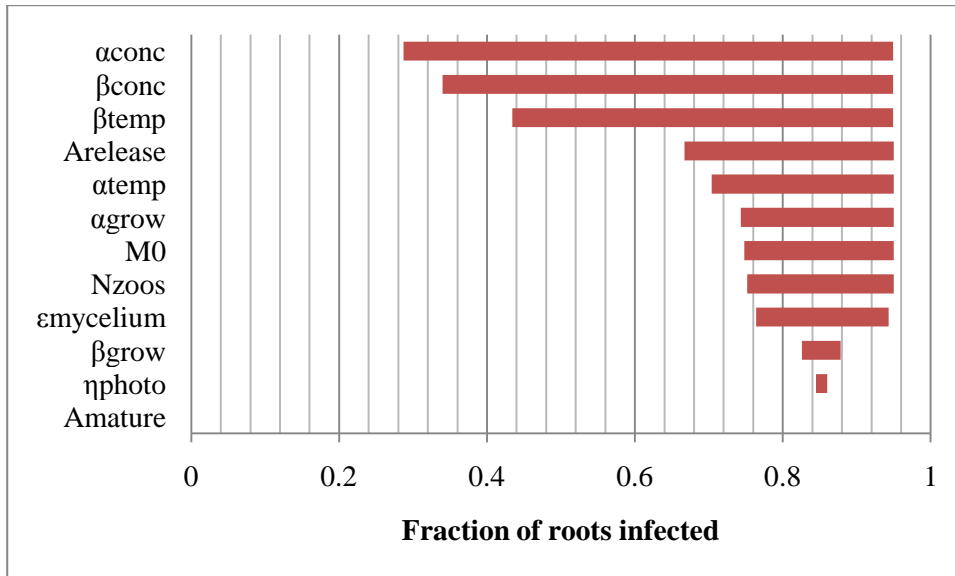




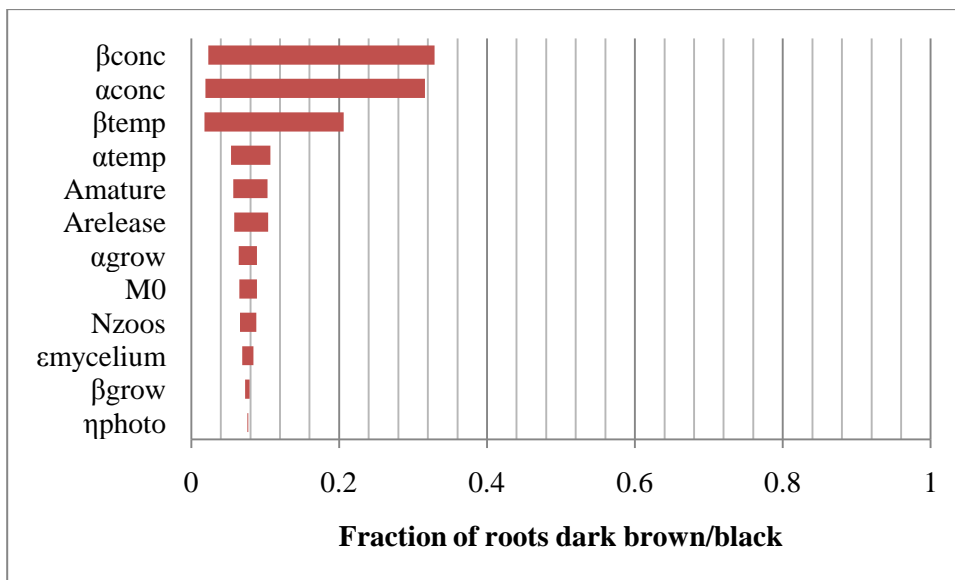
**Figure 6.39** Sensitivity of the fraction of the root dark brown/black to model parameters at harvest day 21 with no inoculation and a nutrient solution of 24 C.



**Figure 6.40** Sensitivity of the fraction of uninfected biomass to model parameters at harvest day 21 with no inoculation and a nutrient solution of 30 C.



**Figure 6.41** Sensitivity of the fraction of the root infected to model parameters at harvest day 21 with no inoculation and a nutrient solution of 30 C.



**Figure 6.42** Sensitivity of the fraction of the root dark brown/black to model parameters at harvest day 21 with no inoculation and a nutrient solution of 30 C.

The parameter to which the model is most sensitive, when considering a naturally developing infection (uninoculated), is the concentration effects on slope and intercept. This variable defines how much damage free zoospores in solution do to the roots. It becomes important under these conditions because at harvest day 21 under

both temperatures *Pythium* has had enough time to complete its reproductive cycle and release zoospores from the initial infection amount. At the higher 30C temperature, this effect is even more apparent as the mycelium develops and grows faster as temperature increases.

As in the case of the inoculated conditions temperature effect variables continue to be important to the model. Speeding or slowing the rate of growth of the mycelium and regulating the time of release of the zoospores results in variables that are very important in a naturally developing infection.

The zoospore releasing age,  $A_{\text{release}}$ , also plays a noticeable role under these conditions. This variable controls when the mycelium has matured enough to release zoospores, which furthers the infection. The number of zoospores released,  $n_{\text{zoos}}$ , plays less of a role.

## **6.5 DISCUSSION:**

The first objective of this chapter was to develop a framework and parameters for a model to predict the growth and spread of *Pythium aphanidermatum* in a hydroponic baby spinach crop grown under controlled environment conditions. To accomplish this goal, data from Katzman's study of *Pythium* (Katzman 2003) in individual spinach crops grown under a variety of conditions was combined with observations made during the NYSERDA spinach disease project (Albright et al., 2007). Katzman's data formed the basis for the model, whereas, the NYSERDA data was used to validate the developed model in multi-cohort production. The core of the disease model is the hour by hour tracking of the growth and development of the *Pythium* mycelia on the roots of each cohort of spinach. Through the examination of Katzman's data it was possible to deduce a growth rate for the mycelium. The remainder of the parameters of the model followed; using the estimated growth rate

and further data to expand the functionality of the model piece by piece, until it encompassed the anticipated scope of its utility.

The second objective was to modify the SUCROS based baby spinach growth model to take into account the damage done to the roots due to *Pythium aphanidermatum* zoospores and mycelia on crop growth. SUCROS was chosen as a model because it tracks growth of both the roots and shoots. The shoots are of primary interest to a grower as that is the part of the crop harvested and sold. The roots are of particular interest because they are the target of *Pythium*, and impact the ability of a crop to transpire and grow. The disease model was developed so a variable, the coefficient of photosynthetic rate reduction, linked the damage in the roots to the reduction in the crop's ability to generate photosynthates, which are the building blocks of the crop.

The final objective of the chapter was to validate the model by comparing predicted crop growth and *Pythium* disease damage to experimentally collected data. The model quite accurately predicts the growth of the crop, and the spread of *Pythium* under the conditions from which it was developed in Katzman's experiments. It also accurately predicts the development and spread of a *Pythium* infection in a multi-cohort situation, where several differently aged cohorts of spinach are present within a common nutrient solution, such as was studied in the NYSERDA disease project.

The model successfully captures the sensitivity of the spread of *Pythium* to the temperature of the nutrient solution. In the sensitivity analyses the variables controlling the response of the growth rate, and the maturation of the mycelium to temperature, exerted the most influence over the outcomes of the model under the conditions examined. This result follows the major finding of Katzman, and the NYSERDA disease project.

A major assumption of the model was that damage due to second generation zoospores was not a significant part of the observed damage in the 18C and 24C temperature conditions. This assumption simplified the development of the mycelium growth rate, and the temperature response curve. This assumption was shown to be valid when the output of the model both with and without reinfection was compared to the observations made in Katzman's experiments.

A shoot quality index was included in the model because Katzman showed small amounts of root damage (10 to 15% infected root) lead to some wilting in the crop. Even a small amount of wilting is unacceptable to a grower as it can lead to rapid decay following harvest. Using strictly biomass as a metric for crop failure would miss this important aspect as biomass does not decrease as soon as *Pythium* appears. In the calculation of the number of harvests until crop failure as a function of temperature and crop duration, it was apparent by examining biomass data only, one would miss the wilting aspect of crop production. Inclusion of crop quality index also allows better modeling of grower response to disease. Material that is wilting and showing signs of infection would be removed as soon as or shortly after it is apparent, as there is no commercial value in continuing that particular cohort.

A primary discrepancy in the model is the number of harvests until failure seems too forgiving of high temperatures and longer crop cycles. However, the number of harvests might be deceiving as it does not represent successive harvests, rather harvests of separate cohorts spaced three days apart. The model is based on data from Katzman and her growing conditions which were primarily concerned with single crop production, and the extension to multi-cohort systems goes beyond the scope of her work. What is more problematic is the sensitivity of the model to the temperature and crop length. At one temperature, production collapses within four harvests, but only two degrees cooler in the nutrient solution and production appears

safe. One would expect rather than be safe, the number of harvests until collapse would merely increase, perhaps by a large amount, but failure would be inevitable. However, it is possible this result is more a function of the conditions the crop is exposed to rather than a serious flaw in the model. The model assumes the rather unlikely condition that the parameter values of the crop growth and the *Pythium* disease model are constant in time, and environmental conditions remain constant as well. The model also neglects the effects any temporary stresses might have on the susceptibility of the crop to *Pythium*. In many of the combinations of temperature and crop duration where the model predicts no problem, it is likely a slight imbalance or temporary change in conditions could push the crop into runaway failure. Monte-Carlo simulation of the model, where the parameters of the model are randomly varied, would present a more realistic summation of the performance of the model by having probability distributions in place of the all or nothing results generated by the constant values of this model.

A potential problem with the model is it was primarily developed from data where the experimental ponds were inoculated with artificially high levels of zoospores, unlikely to be encountered in a natural setting except in the cases of extremely progressed infections. The purpose of the model, however, is to predict how infections develop in commercial settings where large scale inoculations will not occur. Thus it was necessary to extrapolate the performance of the model in a multi-cohort naturally occurring infection from the unintentional damage observed in Katzman's control crops. However, even if the absolute timing of an infection due to natural inoculation is not very accurate, the model provides a means of comparing production strategies; not a definitive prediction of when an outbreak will occur.

The model is also not directly concerned with the accuracy of the prediction of the final harvest weight of highly damaged crops. Though it may reasonably predict

such values, in a commercial setting such knowledge is unnecessary. If the crop is unsaleable, the absolute biomass of the unsaleable crop is unimportant other than from a waste management perspective.

Another consideration is the data on which the model is based was from a highly regulated and frequently cleaned experimental setup. Such conditions are unlikely to be found or maintained in a commercial setting, and as such the model is most likely underestimating the rapidity with which a *Pythium* infection would develop in an uninoculated commercial setting.

A major assumption of the model is zoospores do not carry over from hour to hour. This model would predict that removing a crop and waiting an hour before putting new material in, would break the disease cycle, however, in reality this is certainly not the case. As such caution must be taken in using the model in situations for which it was not developed.

A simplification was the model only considered light integrals have been done in previous spinach work. Extending the use of the model to conditions outside of this range will require the assumption that slowed (or increased) crop growth will not affect the spread of *Pythium* beyond that which the model already accounts. The model was developed using data from harvests conducted 21 days from seeding, however, the light on the crop over the 21 days remained constant.

The temperature effects of the model were derived from only three points. Though they span the range of temperatures usually encountered in greenhouses, care should be taken not to extrapolate too far beyond this range. Further data collected at additional temperatures would better characterize the temperature response.

Another major assumption of the model is the disease organism is *Pythium aphanidermatum*. This is the organism identified by Katzman as present in the Cornell greenhouses. However, Bates and Stanghellini (1984) found in Arizona the

disease organism of interest changed as the temperature of the nutrient solution shifted with the season. *Pythium dissotocum* was found to become prevalent in the ponds of the growing system when the temperature of the nutrient solution was below 23C. However, a complicating factor is that lettuce was grown in the same ponds as the spinach. Though seemingly unaffected by the presence of *Pythium dissotocum*, the lettuce could have served as a reservoir of zoospores, accelerating the development of the disease in the spinach crop. In addition, their production did not focus on a babyleaf crop, where a shortened crop duration could have potentially overcome *dissotocum*. Though *dissotocum* might function at temperatures below that of *aphanidermatum*, there is the possibility that its rate of growth might be such it too can be overcome by shortened crop cycles and perhaps even further temperature reductions of the nutrient solution. Before the model is extended to encompass *Pythium dissotocum*, further experiments with this organism must be carried out.



## CHAPTER 7: SIMULATION OF SELECT PRODUCTION STRATEGIES

### 7.1 INTRODUCTION:

Though *Pythium aphanidermatum* is a major obstacle to commercial hydroponic spinach production, it is not an insurmountable barrier. Commercial hydroponic spinach production is practiced in Japan; however, their technique, namely single use of the nutrient solution followed by extensive cleanout between crops, is very expensive, both in terms of financial and environmental cost. To investigate alternate means of production, NYSERDA (New York State Energy Research and Development Authority) funded a project to examine both existing and new water treatment technologies to remove *Pythium* from nutrient solutions and to evaluate these treatment systems in both pond and aeroponic production (Albright et al. 2007). Chapters two and three of this dissertation describe our work on this issue. The primary findings of the NYSERDA *Pythium* project were there are many effective means of killing and/or removing *Pythium* zoospores from the nutrient solution. A better alternative, however, was to take advantage of crop cultural practices to prevent infection from propagating.

One of the findings of Katzman in her dissertation entitled, “Influence of Plant age, Inoculum Dosage, and Nutrient Solution Temperature on the Development of *Pythium aphanidermatum* in Hydroponic Spinach Production Systems” (2003) was the *Pythium* disease reproductive cycle is highly temperature dependant. Similar results were observed in the NYSERDA Spinach disease project where it was found that by reducing the nutrient solution temperature, the reproductive time of *Pythium* was increased. If the crop cycle is shorter than the time it takes for *Pythium* zoospores to reach maturity and generate new zoospores, the infection would die out as infected roots are removed at harvest without having a chance to release zoospores to further the infection.

Using these findings, production strategies for commercial production of hydroponic spinach have been developed. Various combinations of nutrient solution cooling, two pond system, batch production and nutrient solution sterilization, result in growing techniques with quite different risks of crop losses due to *Pythium* outbreaks that vary under different climate conditions. However, the cost of implementing these different strategies in terms of capital, labor and energy, are also quite different and the decision of which system a grower should select is a combination of the estimated risk of loss for their particular conditions and the cost of their chosen growing technique.

Before investing in a large scale commercial production system, an examination of the expected risk of loss due to *Pythium* is needed. Pilot scale testing of production systems is one option, however, there are drawbacks associated with this approach. Results of a pilot scale test would not necessarily translate to other locations or regions where differences in climate could affect both the disease cycle and/or the growth of the crop itself. Even though controlled environment systems seek to manage environmental conditions within set targets, the cost of doing so may be prohibitive. Pilot scale testing also has the drawback that conditions vary from year to year, and extreme events could strain a system and natural variability in the climate and disease parameters, may not be encountered. In addition, all the combinations of alternative production methods would be difficult to fully test individually on a meaningful scale and would be complicated by disease processes that are likely seasonal, requiring year round studies.

An alternative, or at least a prelude to pilot scale testing, is to model the environment-greenhouse-plant-disease relationship and use a stochastic simulation to determine expected outcomes of different production strategies. The individual variability of the parameters within this complex system can be estimated and

ultimately combined in a Monte-Carlo style simulation to provide an estimate of the expected frequency of *Pythium* outbreaks.

To simulate the greenhouse response to environmental conditions, a stepwise steady state model was developed (described in chapter four). This model takes environmental inputs of solar radiation, outdoor temperature, and outdoor humidity and, combined with typical greenhouse operational control strategies (light and temperature), predicts the indoor PAR levels and air and pond temperatures. Simulation of spinach growth is achieved by using the output of the greenhouse simulation model as input to a modified SUCROS growth model adjusted for hourly growth of spinach under both natural and supplemental lighting (described in chapter five). Working in concert with the growth model is a *Pythium* disease model that simulates the growth and spread of a *Pythium* infection in hydroponic pond production and its effect on the growth of the spinach crop (described in chapter six). This combined greenhouse-spinach-*Pythium* model system is used to explore the effects of different production strategies and growing conditions on crop and *Pythium* disease growth in this chapter.

***Objectives:***

1. simulate the production of hydroponic spinach in the presence of *Pythium aphanidermatum*, under different production strategies,
2. develop quantitative measures of comparing the risk of *Pythium* damage between the different production strategies,
3. present the results of the quantitative measures of the various production strategies to assist in the selection of which strategies to investigate further at pilot scale.

## **7.2 LITERATURE REVIEW:**

### **7.2.1 Previous attempts to grow spinach hydroponically**

One of the first attempts to grow commercial spinach hydroponically in the United States was in Arizona, where both spinach and lettuce crops were grown in the same ponds (Bates and Stanghellini, 1984). Production was carried out year round with 2 week old seedlings (both spinach and lettuce) transplanted into floatation boards and harvested 4 to 6 weeks later. The project commenced in September 1981 and by October 1981 extensive root damage, wilting and even death was apparent in the spinach seedlings after they had spent 1 week in the system. Subsequent investigation of the cause of the root damage identified *Pythium aphanidermatum* as the causal agent of infection. At this time the temperature of the nutrient solution was approximately 26C. Because *Pythium aphanidermatum* was known to be associated with high temperatures the researchers felt as the season progressed into the winter and nutrient solution temperatures dropped, the problem would go away. As the season progressed and the nutrient solution temperature dropped to 22C, root rot and plant death continued. However, another organism, *Pythium dissotocum* was isolated from the roots of the infected crops. Though not as virulent as *Pythium aphanidermatum*, *Pythium dissotocum* was able to infect and kill plants at the lower temperatures and so the commercial spinach production part of the experiment was abandoned.

Studies, however, were conducted to better understand the dynamics of the infections. Throughout the course of a year 100 spinach seedlings were introduced daily to a commercial lettuce pond. The percentage of plants showing stunting or death was recorded every week. Two weeks after placement in the pond, the plants were removed and 10 plants showing either death or infection had their roots analyzed for the presence of *Pythium aphanidermatum* and *Pythium dissotocum*. The

temperature of the pond was recorded daily. Bates and Stanghellini found disease due to *Pythium* occurred year round, however, the lowest disease incidence (< 25% of plants stunted or dead) occurred in January and February when the nutrient solution temperatures were below 20 C. For the remainder of the year when the nutrient solution temperature was above 20 C, high disease incidence was apparent (>50% of plants stunted or dead). Isolation of the causal agents from infected roots showed *Pythium dissotocum* was the predominant or sole pathogen at temperatures below 23 C, whereas *Pythium aphanidermatum* was the predominant or sole pathogen above 23C. Both pathogens were seldom isolated from the same diseased root or plant. A cycling of the pathogen make up with the seasonal temperature of the nutrient solution was apparent. Subsequent investigations of metalaxyl (a fungicide primarily used for soil borne pathogens) had some success in limiting root rot, but showed signs of phytotoxicity at concentrations 10 µg a.i. ml<sup>-1</sup>. Similarly treatments with chlorine, zineb and captan were ineffective at controlling root rot and/or showed extreme phytotoxicity, besides being not legal for use on crops for human consumption.

To date the only successful commercial spinach hydroponic production systems have operated in Japan. It proved possible to grow spinach without succumbing to root rot disease by using a batch production system where a single crop is grown in a single pond, or NFT system and the nutrient solution discarded after each crop and growing apparatus thoroughly cleaned. Of course, a major drawback with this production system is the high cost. Nutrient solution is expensive, primarily in terms of the cost of the clean water itself, and the cost of disposing of used solution. In many locations, disposal of the nutrient solution is tightly regulated to prevent contamination of ground and surface waters. Another disproportionately high expense when compared to other production styles is the high labor associated with cleaning between crops. Thoroughly cleaning the ponds, associated pumps and piping with

disinfectant, and rinsing everything, is a labor intensive process. The relatively short crop cycle of spinach, coupled with the loss of growing area while cleaning is conducted, lead to a very high cost of production. Perhaps only in Japan where a premium price for quality and freshness can be realized, could such a system break even. It is not clear whether the system is still operational, or if it was only run for display purposes.

### 7.2.2 Cornell Experience with Hydroponic Spinach production

The investigation of hydroponic spinach production at Cornell started with the use of a Nutrient Film technique (NFT) production system (Both et al 1996). Spinach seedlings were started in rockwool slabs and transplanted to the NFT system after 8 days. 33 days after seeding a final harvest was conducted. The daily light integral was varied to develop a growth curve for spinach. The results of this experiment were confounded by Chlorine in the tap water (levels above 1 ppm were found to be phytotoxic) and a root rot was identified as *Fusarium*. Treatment with fungicides and potassium silicate were tried, but none proved successful in overcoming the root disease problem.

The cause of the *Fusarium* infection was suspected to be contaminated seed (Katzman 2003). Subsequent experiments in both NFT and pond systems exhibited slightly different symptoms, which proved to be consistent with *Pythium* species. Katzman (2003) investigated not only the possible sources of the infection, but also examined the effects of plant age, inoculum dosage, and nutrient solution temperature on the development of the disease. Concurrent with Katzman's research, a New York State Energy Research and Development Authority (NYSERDA) grant (Albright et al., 2005) allowed the examination of pond production of spinach. The NYSERDA

research was hampered by the fact pond solutions could not be reused and strict hygiene had to be maintained at all times to prevent infection with *Pythium*.

Following the submission of the NYSERDA project final report, a second NYSERDA grant was awarded to investigate both new and existing methods for removing *Pythium* zoospores from the nutrient solution as well as how an alternate growing system, aeroponics, would affect the development and spread of disease (Albright et al., 2007). Aeroponics is a form of hydroponic production where the nutrient solution is misted or sprayed onto the roots of the crop which dangle free in the air. The theory behind selecting aeroponics for investigation with *Pythium* and also examining nutrient solution treatment technologies is movement of zoospores from infected plants could be stopped, or at least severely slowed. *Pythium* zoospores move through the nutrient solution seeking out roots to infect. In aeroponics there is no direct water connection between roots. Unused nutrient solution that drips off the roots would be collected and treated to remove any zoospores present before being resprayed onto the crop. Infected plants may suffer, but the infection should theoretically not be able to progress from plant to plant. However, cultivating spinach aeroponically proved to be very difficult. Nozzles were susceptible to clogging and required frequent inspection to ensure an adequate flow of water to the roots. During the day under high transpirational loads, wilting and death of plants could occur rapidly if the spray was cut off. Other difficulties were due to the high plant densities required for baby spinach production. High plant densities corresponded to dense root mass, which made uniform distribution of the nutrient spray difficult. Root zone temperature is difficult to control in aeroponics. The relatively small volume of nutrient solution present in aeroponics systems is subject to relatively rapid changes of air temperature. During the summer months, wilting and temperature stress were apparent in the aeroponic system. Further investigation into aeroponics was

abandoned when it became apparent temperature control of the nutrient solution in pond systems presented a viable means of production (as described in chapter three).

### 7.2.3 Cornell Experience with Hydroponic Pond production

The hydroponic system selected for the lettuce research module Cornell CEA designed and operated was a pond system, on which 11 day old seedlings were transplanted into floating Styrofoam boards at a spacing of  $1.35\text{E-}2\text{ m}^2$  (21 square inches) per plant. Following 11 days the crop is then transplanted to a final density of  $2.71\text{E-}2\text{ m}^2$  (42 square inches) per plant, where it remains for 14 days until harvest. There are many reasons a pond system was chosen over other systems such as NFT (with which extensive lettuce work has been conducted at Cornell). One major benefit is the materials handling aspect of production. Due to the floating nature of pond production it is relatively easy for a single person to move a very large growing area. Typically plants are respaced at one end of a pond and harvested at the other end. This constant movement through the greenhouse helps to ensure a more uniform growth as individual plants spend a limited amount of time in any spots that may receive more or less light than others. Another major benefit is the buffering action of the pond itself, in terms of water, pH, nutrient, oxygen availability and the ability to control solution temperature. Because the ratio of volume of nutrient solution to plants is high there is a large reservoir of nutrients for the crop.

A major drawback of the NFT system is that, by the time the film of nutrient solution passes over the last plants in a row, it is possible that many of the nutrients and oxygen may have been stripped out. NFT systems are also susceptible to high root zone temperatures as the root zone is usually within two degrees of air temperature. In terms of temperature, the pond is a large thermal mass with a long thermal time constant that takes a long time to heat and cool. This protects the crop



from rapid changes in root temperature that could take place during the summer months when solar radiation is most intense.

All of these benefits of pond systems apply to spinach production as well, however, with the major disadvantage of allowing easy spread of *Pythium* zoospores. However, as has been demonstrated in chapter three, provided temperature control is maintained, it is possible to grow a series of successive spinach crops successfully and continuously without replacing the nutrient solution for each crop. Even with the requirement of replacing or sterilizing the nutrient solutions between batches, the advantages associated with a pond system likely outweigh the disadvantages of other styles of production such as NFT, particularly when the crop is baby leaf spinach. The ability to moderate and control the nutrient solution temperature is a prime reason to select pond hydroponics for spinach production.

#### 7.2.4 Hydroponic Spinach Production Strategies

In the Japanese system of hydroponic spinach production, following harvest, the nutrient solution is discarded and the growing ponds thoroughly sterilized. Though expensive, this system is successful as any infection present is essentially removed after every crop cycle. The key to this style of production is the infection does not have a chance to spread from one crop to the next, building up with each successive generation as would occur if the nutrient solution was reused without treatment between crops. This process not only requires clean nutrient solution, but also batching of the crop; one pond, containing only a single age cohort and then not growing past about 24 days, else there is an outbreak. Deviation from this would potentially allow an infection to spread. Because replacing the nutrient solution every crop, and sterilization of the growing apparatus is costly, the NYSERDA spinach disease project (Albright et al., 2007) examined different treatment technologies for

removing/killing zoospores from the nutrient solution. Effective treatment would potentially allow the nutrient solution to be reused.

The primary finding of Katzman (2003) and the NYSERDA Spinach disease project final report was the *Pythium* life cycle is highly temperature dependant and by controlling the temperature of the nutrient solution, it is possible to produce hydroponic baby-leaf spinach even with *Pythium* zoospores present in the solution. The findings were, provided the crop cycle was shorter than the time it takes for infected material to release zoospores to propagate new infections, the infection would die out as diseased root material is removed at harvest and has no chance to release zoospores. However, certain differences between the study and real life production could potentially lead to problems with this strategy. In the NYSERDA study the crop was harvested on a strict schedule regardless of the size of the individual plants. In commercial production a grower would like to control the amount of growth and could potentially leave the crop in for a few days more to increase its worth. At the end of the crop cycle, when baby-leaf spinach is usually harvested, growth is nearly exponential, so a few extra days of growth can lead to a significant increase in biomass production. (Though this will lead to an increased risk of loss due to *Pythium* by providing more time for zoospores to develop and spread).

A two (or more) pond system is a potential solution for extending the crop cycle, or decreasing the risk associated with a set harvest schedule. The crop is initially started in one pond and upon reaching a certain age (for example half of the expected crop cycle length), is moved to a separate pond. This strategy takes advantage of the time it takes an infected plant to release zoospores into solution. If there is an infection present in the initial pond, the infected material is moved to the second pond before it has a chance to release its zoospores to further the infection. This means new seedlings added to the first pond won't be infected by the material

moved to the second. Though zoospores might be released into the second pond infecting roots there, the initial pond will essentially clean itself. When material is moved to the second pond (which may or may not contain zoospores in solution) it might become infected, however, again if the shortened duration it spends in the second pond is shorter than the reproductive cycle of the disease, the second pond would essentially clean itself. The two pond system depends on the reproductive cycle of the *Pythium* being longer than the duration the crop spends in either pond.

### **7.3 METHODS AND MATERIALS:**

#### **7.3.1 Production strategies modeled**

The focus of this simulation is on pond production. Other types of hydroponic production may be possible, however, the advantages of hydroponic pond production make it a prime candidate for further investigation. There are strategies that can be employed within pond production to overcome its major drawback of susceptibility to *Pythium* infection. The strategies modeled for exploration purposes are:

1. continuous production of multiple aged crops in a single pond with nutrient solution temperature control at 18, 20, 22 and 24 C,
2. continuous production of multiple aged crops in a single pond without temperature control. (Included for comparison with the other strategies, even though it has been demonstrated to not work in practice),
3. continuous production of multiple aged crops using a two pond system with temperature control of the nutrient solution at 18, 20, 22 and 24 C. Plant cohorts older than half the harvest age are moved to a separate pond,
4. continuous production of multiple aged crops using the two pond system without temperature control

Within these production strategies there is an additional factor to model: the biomass of the crop at harvest. Under normal production without the threat of disease, harvest biomass is essentially controlled by two factors; the duration of the crop and the amount of light it receives. Desired size of the individual plants at harvest is a decision the grower must take into account. To reduce the amount of time a crop must spend to reach a certain biomass, supplemental lighting can be used. An added benefit of supplemental lighting and shading is the amount of light a crop receives can be controlled to a consistent year round level, which allows precise scheduling of harvests, and predictability of the harvest biomass. In previous lettuce and spinach research, it was found a daily light integral of 17 mols m<sup>-2</sup> PAR (photosynthetically active radiation) was a reasonable target to avoid tip-burn, so it was also used for this simulation. Tip-burn is less of a concern in baby spinach production, because of the very open canopy and good air movement. Spinach crop durations of 12, 14 and 16 days were investigated. To demonstrate the importance of light control on the reduction of damage due to *Pythium*, conditions with no supplemental lighting were also examined. For the case of single pond production with no supplemental lighting, spinach crops remained in the pond until they matched the average harvest biomasses corresponding to 12, 14 and 16 days of growth under 17 mols m<sup>-2</sup> of daily PAR (or until they are disease damaged to a point that they must be removed). To simplify the model, the two pond system was only modeled with supplemental lighting control and defined harvest scheduling. Using a two pond system without consistent predictable growth would require crop growth prediction and handling that is beyond the scope of this study.

Several assumptions were needed to simulate some of a growers production decisions. The first assumption deals with when *Pythium* damaged crops are removed from the growing system. If a crop is clearly damaged beyond recovery, the grower

would remove it as soon as possible as space and time in a greenhouse are expensive. If some of the plants of the crop are visibly wilted (which is more likely to be noticed by a grower than root damage), it usually signifies a *Pythium* infection severe enough to trigger the removal of the entire crop (all cohorts).

The second assumption was seedlings are floated on a strict three day schedule. This was the schedule followed during the multi-cohort experiments detailed in chapter three, which parts of the disease model were developed from. It takes approximately two days from seeding for germination to occur, at which point the seedlings are floated.

### 7.3.2 Simulation of Spinach Production Strategies

#### *Integration of greenhouse, spinach growth and disease model.*

To evaluate the performance of the production strategies, a greenhouse simulation model was coupled with a spinach growth and *Pythium* disease model. The greenhouse simulation model was necessary to provide a means of calculating the nutrient solution temperature, given climate data. The nutrient solution temperature is of primary importance in the disease model. The greenhouse model also calculates air temperature and amount of PAR reaching the crop, based on climate data, time of day and day of the year, as well as estimates of the greenhouse transmittance. All three models were necessary to represent the complete system of spinach production.

#### *Input to the model*

Input to the model was the hourly climate data collected from the Cornell Northeast Regional Climate Center weather station located on Game Farm Road, near Ithaca, NY. Variables included air temperature, relative humidity and solar radiation. Other weather sets could be used to analyze performance of the different production

strategies in different regions of the country. The weather data set encompasses 12 years of data, from 1983 to 1994. Simulation of weather data including extreme events and typical mean year data, are other alternatives not presented here. Because the temperature variation within the pond system varies only seasonally and is highly resistant to shorter duration temperature swings (a major benefit), using actual data from a 12 year period would adequately represent the natural variability, expected from year to year. A simulation of weather, while perhaps better able to represent extreme events, would require considerable development. Temperature, light and relative humidity, which are all important in determining pond temperatures, are also highly correlated.

Control of PAR within the greenhouse follows the rules for supplemental lighting and shading specified in the Light and Shade System Implementation (LASSI) algorithm (Albright, 1998). For conditions where integral control was used, a daily target PAR of  $17 \text{ mol m}^{-2}$  was applied. The shade curtain was closed for thermal protection of the crop at night (night being defined as the hour beginning after sunset to the end of the hour before sunrise).

Aerial temperature setpoints were 24 C during the day, and 19C during the night and assumed values for the greenhouse parameters follow typical greenhouse characteristics as described in detail in chapter four.

### *Initial Conditions*

The yearly weather data sets are based on day of year; i.e., they start with January 1<sup>st</sup> (day 1) and end with December 31<sup>st</sup> (day 365 or 366). To start the model, typical values of temperatures within the greenhouse for January 1<sup>st</sup> were used as the initial conditions. These typical values were determined by running the model for a full year and recording the temperatures on the last hour of the last day of the year. To

start the spinach production simulation, spinach crop cohorts were added to the production ponds on a three day schedule. At the appropriate time or age, each cohort was harvested. If a *Pythium* outbreak occurred, all cohorts in the system were culled, and production restarted following the addition of new crop cohorts on the same three day schedule.

### *Model Output*

Of primary interest to a potential commercial grower is the frequency of *Pythium* outbreaks. The output of each model run was the successful harvest data, and the *Pythium* cull data. The harvest and cull data included harvest shoot and root dry biomass, cohort ID (used to determine the values of the random variables used for the growth model of this specific cohort), mass of mycelium, mass of the roots dark brown/black, *Pythium* cohort ID (used to determine the values of the random variables used for the *Pythium* disease model), age of the crop, and hour of the simulation at which the harvest occurred.

### *Monte-Carlo Simulation*

The greenhouse, growth, and disease model is a representation of a complex system that links the model's inputs (climate data) to the model's output (spinach production data). By modifying the control and production system decision aspects of the model, it is possible to explore the effects of these variables on the output; and provide insight into which systems and decisions might yield the best performance.

However, not all parameters of the model are deterministic. Many of the values of the model parameters are estimates with an error associated with their estimation. Another source of error is in the form of the model itself; i.e. that the model does not adequately represent the processes under consideration.

To account for the parameter estimation and model error a stochastic method was used. Stochastic simulation is drawing random values from these input distributions, calculating the output of the model with these values, storing the results and repeating the process for a number of iterations. Each iteration is a possible combination of the random variables, and by performing enough iterations, a stable distribution of the output variable will be generated (Hardaker et. al. 2004).

### *Variability of parameters*

The reason for using stochastic simulation is there are uncertainties associated with the parameter values within the system model. A greenhouse is a complicated environment with considerable uncertainty in the manner of interaction with the climate. To simplify the analysis, these aspects of the system were treated as deterministic.

To calculate hourly temperatures within the greenhouse for each iteration would add considerable computation time (approximately 10 minutes per simulated year). A better use of the greenhouse model was to determine the temperatures for each set of control decisions, and store the required hourly data values for use by the simulation when needed. Therefore, the greenhouse simulation needed to run a total of ten times; with uncontrolled and controlled nutrient solution temperatures of 18, 20, 22 and 24C, with and without supplemental lighting.

Within the modified SUCROS growth model (Goudriaan and van Laar, 1994) there are seven parameters used to predict growth of the spinach crop under varying environmental conditions. They are:

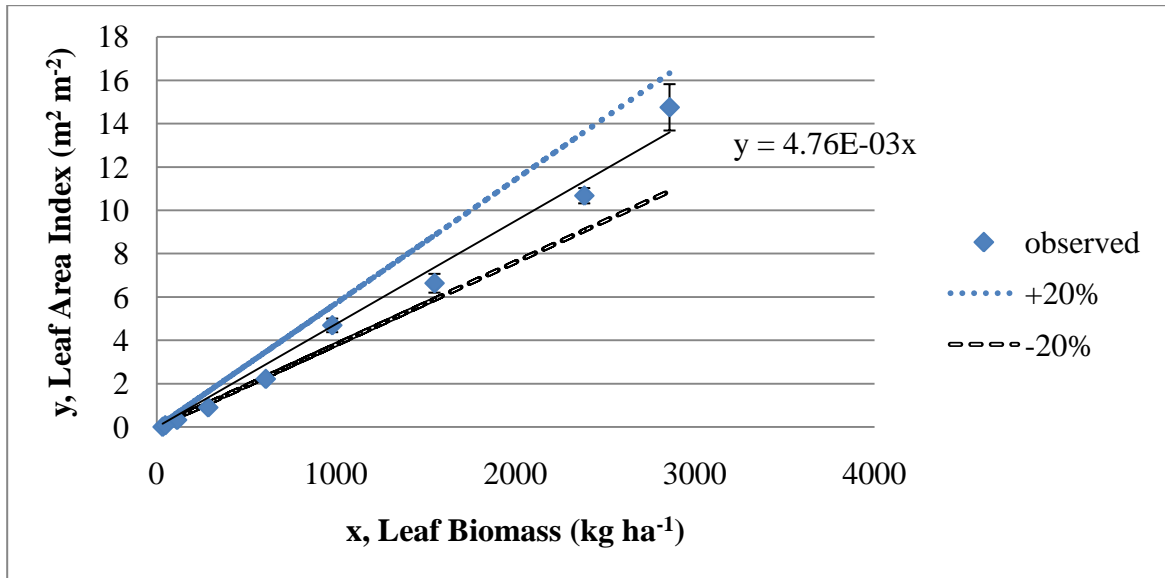
1. A, the slope of the function relating the leaf area index to the dry weight of the leaves,



2. AMX, Leaf CO<sub>2</sub> assimilation,
3. EFF, initial light conversion factor for individual leaves,
4. KDF, extinction coefficient for leaves,
5. MAINLF, hourly maintenance requirement of the leaves,
6. MAINRT, hourly maintenance requirement of the roots,
7. SCP, scattering coefficient of the leaves for PAR.

A probability distribution for each variable is assigned for the Monte-Carlo simulation. In the simulation, a new random value for the variables describing crop growth is assigned to each crop cohort. Each cohort represents a distinct grouping of plants of the same age. With scheduled harvesting, new cohorts are added every three days. The variability assigned to these cohorts is due to factors such as differences of seed batches, (whether from common batches or just the effects of older seed). Differences between cohorts could also be caused by slight environmental differences not considered in the growth model. Minor variations in temperature and moisture content of the media during germination can have significant effects on the final harvest biomass of a cohort. Changes in the nutrient solution composition over time are another source of variation, as corrections are not usually made on a daily basis.

A, the slope of the function relating leaf area index to the leaf dry weight, was derived from data collected during the spinach disease project (Albright et al., 2007). Figure 7.1 plots the observed Leaf Area Indexes corresponding to observed Leaf Biomasses. In addition, predicted LAI values as a function of Leaf Biomass are plotted, assuming the best fit slope of 4.76E-3 is both increased and decreased by 20%. A triangular distribution over this range is assumed.



**Figure 7.1** Leaf Area Index as a function of Leaf Biomass. Observed values and predicted values with the best fit line slope increased and decreased by 20%. Observed values from spinach crop harvested every two days, with an average daily light integral of  $19.2 \text{ mol m}^{-2}$ .

AMX is the maximum leaf  $\text{CO}_2$  assimilation at light saturation and in the growth model is assumed to have a value of  $40 \text{ kg CO}_2 \text{ ha}^{-1} \text{ hr}^{-1}$  per  $\text{W m}^{-2}$  at ambient  $\text{CO}_2$  concentrations. Goudriaan and van Laar (1994) suggested a value of 40 for plants with a good supply of water and nutrients and exposure to ambient  $\text{CO}_2$  concentrations. No indication was given for the range and so a mean value of 40 is chosen. AMX is assumed to have a triangular distribution with a range  $\pm 10\%$  of the mean value.

EFF is the initial light conversion factor for individual leaves and assumed to have a mean value of  $0.45 \text{ kg ha}^{-1} \text{ h}^{-1}$  at ambient  $\text{CO}_2$  concentrations (Goudriaan and van Laar, 1994). EFF is related to the concentration of  $\text{CO}_2$  and at a value of 1000 ppm  $\text{CO}_2$  becomes  $0.52 \text{ kg CO}_2 \text{ ha}^{-1} \text{ hr}^{-1}$  per  $\text{W m}^{-2}$ . 1000 ppm  $\text{CO}_2$  is 2.5 times the ambient concentration of  $\text{CO}_2$  the model assumes. EFF is assumed to have a triangular distribution with a mode of  $0.45 \text{ kg ha}^{-1} \text{ h}^{-1}$ , with a range of  $0.40$  to  $0.50 \text{ kg ha}^{-1} \text{ h}^{-1}$ .

KDF is the extinction coefficient for leaves and varies as a function of leaf area index (LAI, the area of leaf per unit area of soil surface) and the orientation of the leaves. Although the value of LAI changes as a crop grows, the assumption is several crop ages (cohorts) are present in the greenhouse at the same time. This results in an average LAI that stays relatively constant in time. In his dissertation, Both (1995) used a value of 0.72 for lettuce, however, this value was assumed over the entire crop length, 35 days and a plant with a tightly clustered head, quite unlike babyleaf spinach, where the leaves are more upright and distinct rather than clustered. For this reason, a value of KDF of 0.5 was selected. Monteith (1969) tabulated several values for KDF, which ranged from a low of 0.29 for ryegrass, up to 1.13 for cotton. KDF for this model is assumed to have a triangular distribution over the range 0.29 to 0.72, mode 0.5.

MAINLF is the maintenance requirement for leaves, expressed as a fraction of the leaf dryweight. Similarly, MAINRT is the maintenance requirement for plant roots, also expressed as a fraction of the root dryweight. These requirements are temperature dependant according to Penning de Vries and van Laar (1982). At 30 C the value is 1.41 times as large as at 25 C. Values of MAINLF and MAINRT are thus assumed to have a triangular distribution over the range  $\pm 40\%$  of their mean value; MAINLF 0.018 to 0.042, mode 0.03 and MAINRT 0.009 to 0.021, mode 0.015.

SCP is the scattering coefficient of the crop leaves for PAR. Monteith (1969) states the proportions of radiation transmitted and reflected (the definition of the SCP is the sum of these two proportions) is relatively similar for leaves of different crops. The SCP for PAR is given by Goudriaan and van Laar (1994) as 0.2 (whereas for near infra red (NIR) light it is as high as 0.9). No range for the SCP is given by Goudriaan and van Laar and so it is assumed SCP has a triangular distribution over the range 0.15 to 0.25, with mode 0.2.

Within the disease model the twelve parameters were estimated from different data sources (outlined in chapter six). Unfortunately, there are no values for the parameters of the disease model available in the literature, as the form of the model and parameters were developed as a part of this dissertation.

The parameters examined are:

1.  $\beta_{\text{conc}}$ , Concentration effect slope
2.  $\alpha_{\text{conc}}$ , Concentration effect intercept
3.  $\epsilon_{\text{mycelium}}$ , Reference amount of mycelium @ 25 zoos ml<sup>-1</sup>
4.  $M_0$ , Starting Mass of mycelium
5.  $\beta_{\text{grow}}$ , Mycelial growth rate slope
6.  $\alpha_{\text{grow}}$ , Mycelial growth rate intercept
7.  $A_{\text{mature}}$ , Maturation age of mycelium
8.  $\eta_{\text{photo}}$ , Coefficient of Photosynthetic Rate reduction
9.  $\beta_{\text{temp}}$ , Temperature effect slope
10.  $\alpha_{\text{temp}}$ , Temperature effect intercept
11.  $A_{\text{release}}$ , Zoospore release age
12.  $N_{\text{zoos}}$ , Zoospore release amount

With the crop growth variables, assigning new random values for each cohort was an obvious choice. With the disease variables, the selection of a time to assign new random values is less clear. One possibility is to assign each crop cohort new random disease variable values. However, as the disease generally takes more than one crop generation to develop, assigning new values at this frequency would likely result in an averaging of the disease variable values, and not produce the extreme events required to cause failures in some of the production systems. Another complicating factor is the temperatures which generally cause failures, usually only occur during the summer months. As a compromise it was decided to select new

random values for the disease model at the beginning of every year and after every crop failure. The annual change corresponds to a time when the nutrient solution temperatures and subsequent disease pressures are least. The change after every crop failure represents the breaking of the disease cycle through sterilization.

Seven of the parameters are a part of linear approximations for which there is an associated slope and intercept. In the case of the concentration adjustment factors ( $\beta_{\text{conc}}$  and  $\alpha_{\text{conc}}$ ) it was assumed that the values of slope and intercept are correlated, and the equation for the concentration adjustment factor (originally equation 6.15) is modified to:

$$E(C) = \beta_{\text{conc}} * (\log(Z_{\text{conc}}) - \log(25 \text{ zoos ml}^{-1})) + \alpha_{\text{conc}} \quad 7.1$$

where:  $Z_{\text{conc}}$  is the concentration of zoospores in solution ( $\text{zoos ml}^{-1}$ ).

$25 \text{ zoos ml}^{-1}$  is the baseline concentration of zoospores.

The slope ( $\beta_{\text{conc}}$ ) was assumed to have a lognormal distribution with a mean of 0.180 and a coefficient of variation (CV) of 0.5. The intercept was also assumed to vary lognormally with a mean of 1.0 and a CV of 0.5.

The mycelial growth rate parameters ( $\beta_{\text{grow}}$  and  $\alpha_{\text{grow}}$ ) were assumed to be correlated. Rather than adjust the form of the equation (as was done for the concentration parameters), triangular distributions were assumed for the minimum and maximum observed mycelial growth rates (0.010 at  $0.0 \text{ kg ha}^{-1}$  uninfected root and 0.021 at  $54.2 \text{ kg ha}^{-1}$  uninfected root respectively). A common uniform random variable was then used to generate a point on each distribution from which the slope and intercept were then calculated. The minimum growth rate was assumed to have a triangular distribution with a mean of 0.010 and limits of the mean  $\pm 0.00325$ . The maximum growth rate was assumed to have a triangular distribution with a mean of 0.021 and limits of the mean  $\pm 0.0042$ .

The Coefficient of Photosynthetic Rate reduction ( $\eta_{\text{photo}}$ ) was assumed to have a lognormal distribution with a mean of 0.79 and a CV of 0.5.

The temperature adjustment parameters ( $\beta_{\text{temp}}$  and  $\alpha_{\text{temp}}$ ) followed a similar procedure as the mycelial growth rate to account for their correlation. A triangular distribution with a mean of 0.58 and a range of  $\pm 0.034$  was assumed for the age adjustment factor at 18C. A triangular distribution with a mean of 1.0 and a range of 0.042 was assumed for the age adjustment factor at 24C. A common uniform random variable was then used to generate a point on each distribution, from which the slope and intercept were then calculated.

$\epsilon_{\text{mycelium}}$  is the reference amount of mycelium @ 25 zoos ml<sup>-1</sup>, or the amount of mycelium formed on roots when exposed to a concentration of 25 zoos ml<sup>-1</sup> in the nutrient solution. Three values of  $\epsilon_{\text{mycelium}}$  were estimated from Katzman's data (day1, day 9 and day 14 inoculations) and were then averaged for a value of 3.23 kg mycelium ha<sup>-1</sup>. It was assumed  $\epsilon_{\text{mycelium}}$  has a triangular distribution over the range of the lowest to the highest  $\epsilon_{\text{mycelium}}$  (0.71 to 6.75 kg mycelium ha<sup>-1</sup>, mode 3.23 kg mycelium ha<sup>-1</sup>)

The starting mass of mycelia ( $M_0$ ) is a means of inoculating the crop with a baseline amount of damage to simulate a naturally developing infection. It was estimated by selecting a value that produced the observed level of damage in Katzman's uninoculated control crops after 21 days. This parameter represents a major simplification of the initial infection process and so the range of values should represent uncertainty with the estimated value. For this reason a lognormal distribution was assumed with a CV of 1.0 and  $\mu_x = 1.95\text{E-}3$  kg mycelium ha<sup>-1</sup>.

The maturation age of mycelium,  $A_{\text{mature}}$ , represents the age at which the mycelial material begins to turn dark brown/black, whereas, the zoospore release age ( $A_{\text{release}}$ ) represents the age the mycelia typically begin to release zoospores. Though

the values of these variables are assumed to be the same (128 hours) the probability distributions are not. The maturation age of the mycelium was estimated from observed data, whereas, the zoospore release ages were estimated as pairs of zoospore release ages and zoospore release numbers that fit the data. The value of 128 hours for the zoospore release age was selected to be the same as the  $A_{\text{mature}}$ , because it is likely the onset of dark brown black material represents the full reproductive cycle of *Pythium*. The  $A_{\text{mature}}$  was assumed to have a triangular distribution with an estimated median value of 128 hours, a lower range of 104 hours and an upper range of 152 hours. The  $A_{\text{release}}$  was assumed to have a triangular distribution, with a mode of 128 hours, an upper range of 168 hours and a lower range of 88 hours.

The zoospore release amount ( $n_{\text{zoos}}$ ) is the number of zoospores released per kilogram of infected root material that achieved zoospore releasing age. After reaching this age, the infected material releases a constant amount of zoospores per kilogram per hour, as long as the infected material remains in solution. The estimate for  $N_{\text{zoos}}$  was developed at the same time as the  $A_{\text{release}}$ . The uncertainty of this number is quite large and it should vary lognormally with a C.V. of 50% and a  $\mu_x$  of  $1.1\text{E}6$  zoospores  $\text{kg}^{-1}$  mycelia.

The random number generator within Java was used to provide the random numbers necessary for the Monte-Carlo simulation. These random numbers were saved within a text file so common random numbers could be used for different simulated production strategies. The random number generator provided a uniformly distributed variable, whereas, the variability of some of the parameters, in the simulation were assumed to be lognormal. To transform the uniformly distributed random variables into lognormally distributed random variables the Box-Muller process was used to transform uniform variables to normal, which were then converted to lognormal.

### 7.3.3 Implementation of Simulation Model

The simulation model was implemented in Java code to use the same language as the previously coded greenhouse, growth and disease models. (Appendix A.)

## 7.4 RESULTS:

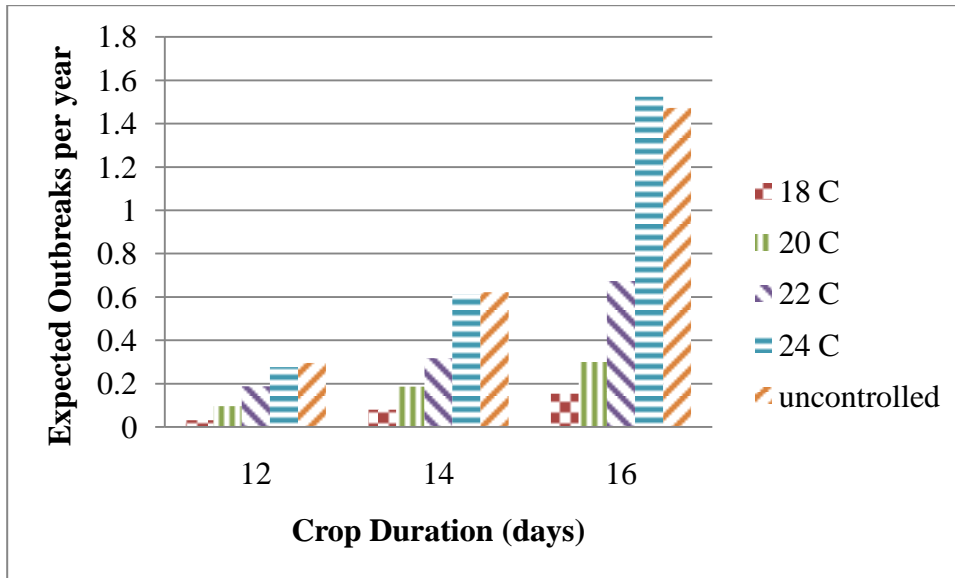
### 7.4.1 One pond system, controlled daily light integral

The results of the one pond system with light control (strict, time based harvest schedule) are presented in Table 7.1, and Figure 7.2.

**Table 7.1** Simulated mean time between *Pythium* outbreaks severe enough to require a cleanout. For a multi-cohort one pond system with nutrient solution temperatures of 18C, 20C, 22C, 24C and uncontrolled, and crop durations of 12, 14 and 16 days with a daily light integral of 17 mols m<sup>-2</sup> PAR. (Simulation run for 500 years).

Nutrient Solution Temperature (C)	Harvest Age (days)	Average Harvest dry mass (kg ha <sup>-1</sup> )	Outbreaks	Expected Frequency of Outbreaks (outbreaks year <sup>-1</sup> )	Ratio to uncontrolled
18	12	1186	16	0.032	0.11
18	14	1534	40	0.080	0.13
18	16	1885	77	0.154	0.10
20	12	1185	48	0.096	0.32
20	14	1534	93	0.186	0.30
20	16	1885	150	0.300	0.20
22	12	1185	94	0.188	0.64
22	14	1533	159	0.318	0.51
22	16	1883	337	0.674	0.46
24	12	1185	138	0.276	0.93
24	14	1532	306	0.612	0.98
24	16	1880	762	1.524	1.04
Uncontrolled	12	1090	148	0.296	
Uncontrolled	14	1409	311	0.622	
Uncontrolled	16	1721	736	1.472	

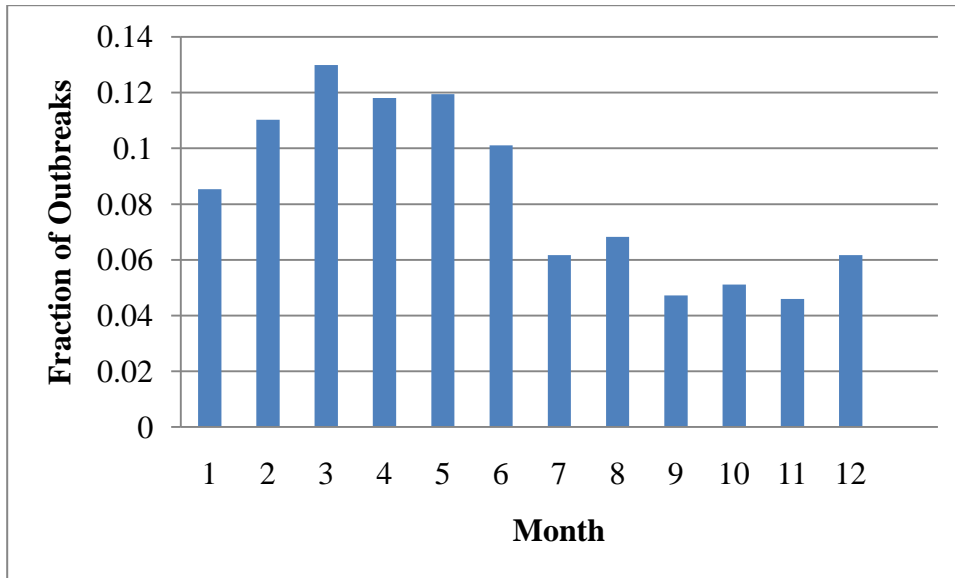




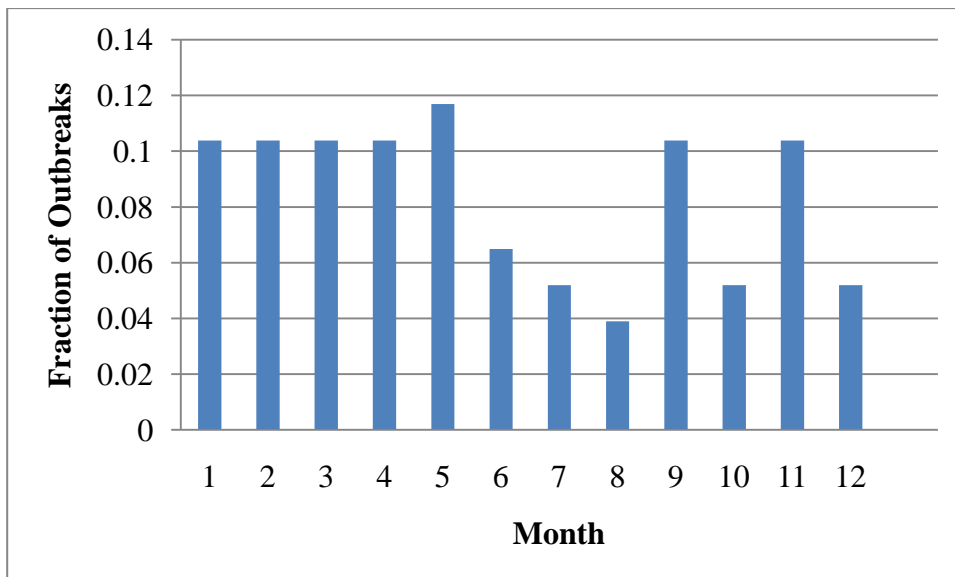
**Figure 7.2** Expected number of *Pythium* outbreaks per year, as a function of nutrient solution temperature, and crop duration. For a one pond (multi-cohort) production system with a daily light integral of 17 mols m<sup>-2</sup>.

Clearly there is a benefit to using a lower nutrient solution temperature. As would be expected, the longest crop duration examined (16 days) benefits most from a reduced nutrient solution temperature. Comparing the 18 C, 16 day condition to the uncontrolled, 16 day condition, the frequency of outbreaks was 9.5 times greater in the uncontrolled condition. The 12 and 14 day conditions also benefited considerably with improvements of 9.3 and 7.8 times, respectively. All of the temperature and crop duration conditions examined showed a decrease in the frequency of outbreaks with a reduction of temperature. The results of the uncontrolled nutrient solution temperature conditions are quite similar to those where the temperature is controlled to 24C. This is to be expected as the nutrient solution temperature tends to follow the air temperature, and the setpoint for air temperature during the day is 24C (19C night).

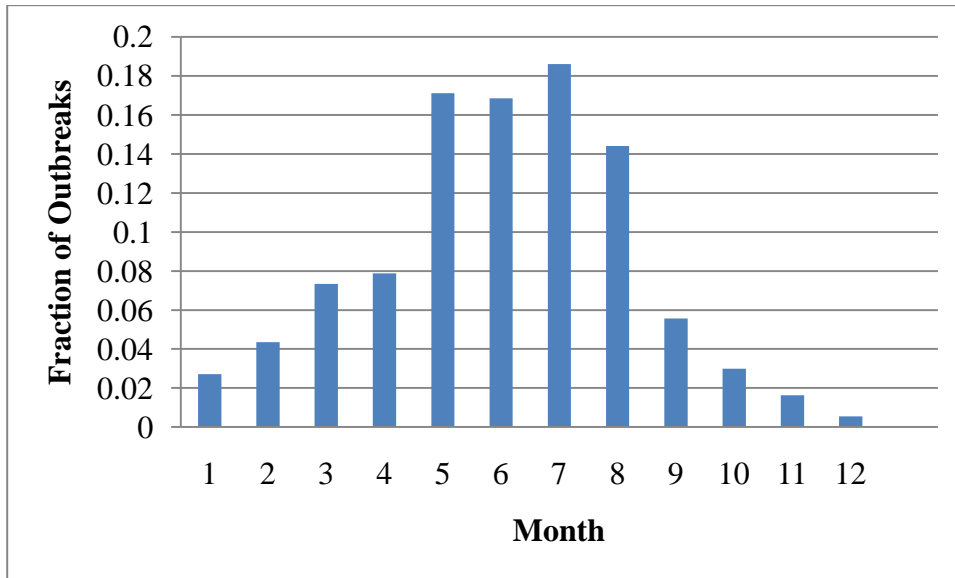
Figure 7.3, Figure 7.4 and Figure 7.5 show the number of *Pythium* occurrences by month for the 24C, 18C and uncontrolled nutrient solution temperatures, 16 day crop duration conditions respectively.



**Figure 7.3** *Pythium* outbreaks by month, for the 24C nutrient solution temperature, 16 day crop duration, and two pond condition.



**Figure 7.4** *Pythium* outbreaks by month, for the 18C nutrient solution temperature, and 16 day crop duration condition.



**Figure 7.5** *Pythium* outbreaks by month, for the uncontrolled nutrient solution temperature, and 16 day crop duration condition.

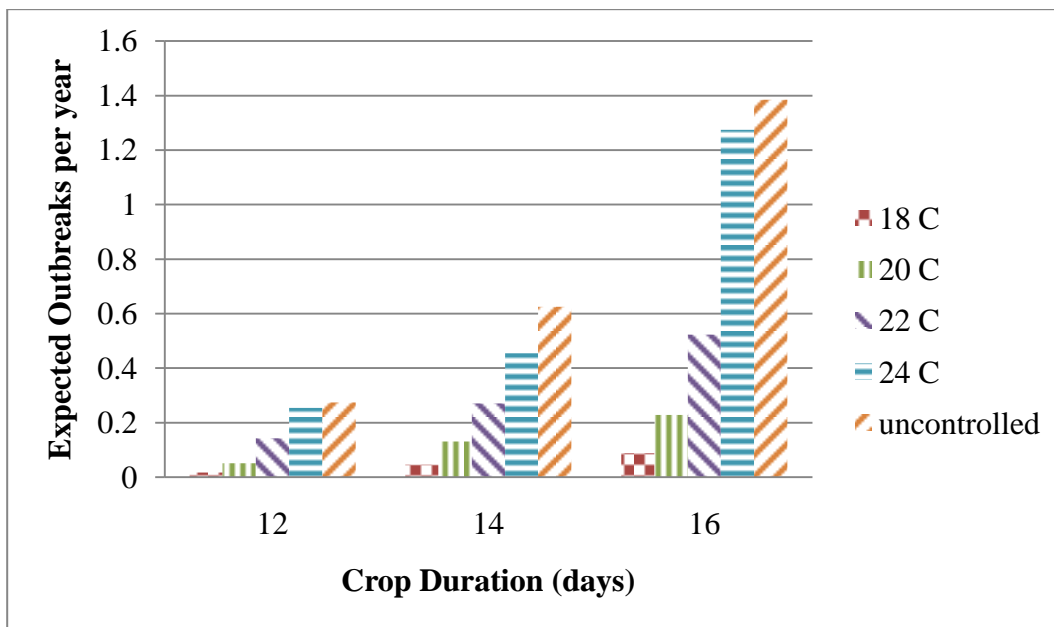
The 24 and 18C conditions show a large negative skew, with more outbreaks occurring earlier in the year than would be expected considering the nutrient temperatures are maintained the same all year long. However, this is an artifact of the simulation model where new disease parameters are assigned at the beginning of every year. For this reason, the timing of *Pythium* infections is not examined further. An investigation of the timing of *Pythium* infections throughout the year would require a more complicated method of assigning variability to the *Pythium* disease parameters. Such an analysis would be of little value as uncontrolled nutrient solution temperatures are clearly not an option for commercial production of spinach.

#### 7.4.2 Two pond system, controlled daily light integral

The results of the two pond system with light control (strict, time based harvest schedule) are presented in Table 7.2. and Figure 7.6.

**Table 7.2** Simulated mean time between *Pythium* outbreaks severe enough to require a cleanout. For a multicohort two pond system with nutrient solution temperatures of 18C, 20C, 22C, 24C and uncontrolled and crop durations of 12, 14 and 16 days with a daily light integral of 17 mols m<sup>-2</sup> PAR. Also included is the ratio of the mean time between outbreaks for the two pond system over the one pond system. (500 years of simulation).

Nutrient Solution Temperature (C)	Harvest Age (days)	Average Harvest dry mass (kg ha-1)	Outbreaks	Frequency of Outbreaks (outbreaks year <sup>-1</sup> )	Ratio to uncontrolled	2 Pond / 1 Pond
18	12	1181	8	0.016	0.06	0.50
18	14	1529	23	0.046	0.07	0.58
18	16	1883	44	0.088	0.06	0.57
20	12	1180	25	0.050	0.18	0.52
20	14	1529	65	0.130	0.21	0.70
20	16	1883	114	0.228	0.16	0.76
22	12	1180	71	0.142	0.52	0.76
22	14	1528	135	0.270	0.43	0.85
22	16	1881	261	0.522	0.38	0.77
24	12	1180	127	0.254	0.93	0.92
24	14	1527	227	0.454	0.73	0.74
24	16	1877	637	1.274	0.92	0.84
Uncontrolled	12	1179	137	0.274		0.93
Uncontrolled	14	1525	312	0.624		1.00
Uncontrolled	16	1873	692	1.384		0.94



**Figure 7.6** Expected number of *Pythium* outbreaks per year, as a function of nutrient solution temperature, and crop duration. For a two pond (multi-cohort) production system with a daily light integral of 17 mols m<sup>-2</sup>.

As in the case of the one pond system, there is a clear benefit to lower nutrient solution temperatures on the occurrence of *Pythium* outbreaks. At lower temperatures there is clearly a benefit to using a two pond system. For the case of a nutrient solution temperature of 18 C, compared to an uncontrolled, in a 12 day crop cycle, the expected frequency of *Pythium* outbreaks is 17 times smaller. Similar magnitudes of improvement are also found in the 14 and 16 day crop cycles; 13.6 times and 15.7 times, respectively. When comparing the two pond system to the one system, it is apparent that improvement in expected frequency of outbreaks is greater at lower than higher temperatures. At 18C, the frequency of outbreaks is 0.5 to 0.57 of the one pond system, whereas, with the 24C conditions, the frequency is 0.74 to 0.92 of the one pond system. A reason for this difference is at higher temperatures there is a greater chance of *Pythium* completing its reproductive cycle within the time the crop spends

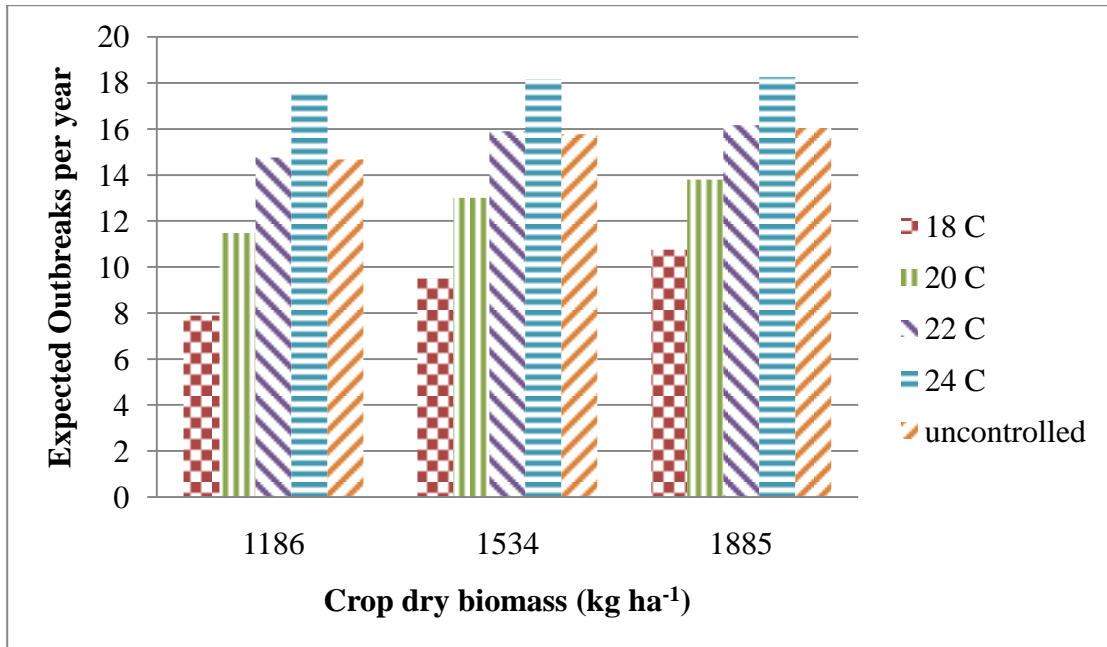
in the first pond. If the initial pond is subject to frequent outbreaks, the second finishing pond is too. At lower temperatures the release of new zoospores is more likely to be delayed until the cohorts are in the finishing pond, where the only other cohorts to be infected are older and unable to infect material in the initial pond.

#### 7.4.3 One pond system, natural daily light integral

The results of the one pond system with no light control (biomass based harvest schedule) are presented in Table 7.3 and Figure 7.7.

**Table 7.3** Simulated mean time between *Pythium* outbreaks severe enough to require a cleanout. For a multicohort one pond system with nutrient solution temperatures of 18C, 20C, 22C, 24C and uncontrolled, and target crop harvest biomasses (kg dry mass ha<sup>-1</sup>) of 1186, 1534 and 1885 with no light integral control. (500 years of simulation).

Nutrient Solution Temperature (C)	Harvest Dry mass (kg ha <sup>-1</sup> )	Average Harvest age (days)	Outbreaks	Frequency of Outbreaks (outbreaks year <sup>-1</sup> )	Ratio to uncontrolled
18	1186	24	3940	7.9	0.54
18	1534	27	4748	9.5	0.60
18	1885	29	5378	10.8	0.67
20	1186	22	5740	11.5	0.78
20	1534	25	6501	13.0	0.82
20	1885	27	6899	13.8	0.86
22	1186	21	7379	14.8	1.01
22	1534	23	7950	15.9	1.01
22	1885	25	8081	16.2	1.01
24	1186	20	8829	17.7	1.20
24	1534	21	9084	18.2	1.15
24	1885	23	9127	18.3	1.14
Uncontrolled	1186	21	7339	14.7	
Uncontrolled	1534	23	7887	15.8	
Uncontrolled	1885	25	8019	16.0	



**Figure 7.7** Expected number of *Pythium* outbreaks per year, as a function of nutrient solution temperature, and target crop harvest biomass. For a one pond (multi-cohort) production system with no supplemental light (natural daily light integral).

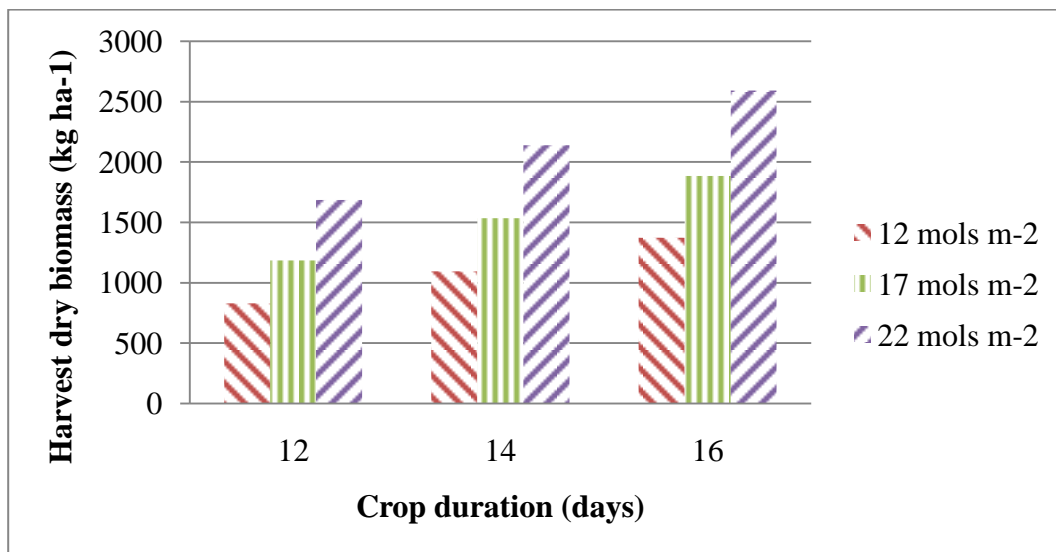
The expected frequency of *Pythium* outbreaks is quite drastically different for the conditions using harvesting triggered by biomass (with no supplemental lighting) as opposed to a strict schedule with light control. Even with a nutrient solution of 18 C and the smallest harvest biomass considered, the estimated frequency of outbreaks is 8 per year. This is nearly 250 times greater than the corresponding schedule based harvest condition. The frequency of outbreaks for the 24 C and smaller biomass condition (1186 kg ha<sup>-1</sup>) is only 2.2 times greater than the frequency for 18 C condition.

#### 7.4.4 Effect of daily light integral

The effect of varying the light integral the crops are grown under is illustrated in Table 7.4 and Figure 7.8.

**Table 7.4** Effect of daily light integrals of 12, 17 and 22 mol m<sup>-2</sup> on Pythium outbreak frequency and harvest dry biomass, for a multi-cohort one pond system with a nutrient solution temperature of 18C, and crop durations of 12, 14 and 16 days.

	Daily Light Integral (mols m <sup>-2</sup> )					
	12		17		22	
Crop duration (days)	Outbreak frequency (outbreaks year <sup>-1</sup> )	Harvest dry Biomass (kg ha <sup>-1</sup> )	Outbreak frequency (outbreaks year <sup>-1</sup> )	Harvest dry Biomass (kg ha <sup>-1</sup> )	Outbreak frequency (outbreaks year <sup>-1</sup> )	Harvest dry Biomass (kg ha <sup>-1</sup> )
12	0.03	828	0.03	1186	0.03	1685
14	0.09	1094	0.08	1534	0.07	2140
16	0.17	1373	0.15	1885	0.14	2591



**Figure 7.8** Effect of daily light integral (12, 17 and 22 mol m<sup>-2</sup>) on harvest dry biomass with crop durations of 12, 14 and 16 days, in a multi-cohort one pond system at 18C.

It is quite clear that increasing the duration of the crop increases the harvest biomass. Extending the crop cycle from 12 to 16 days (33%) increases the dry biomass by 66% with a daily light integral of 12 mol m<sup>-2</sup>, and 54% with a daily light integral of 22 mol m<sup>-2</sup>. The relationship of light to harvest biomass is also clear. Approximately doubling the daily light integral (12 to 22 mol m<sup>-2</sup>) results in approximately double the dry harvest biomass, regardless of the crop duration.



Changing the amount of light a crop receives has little effect on the frequency of disease outbreak. There is a slightly greater frequency at the low light levels. This difference is due to the fact that the factors controlling mycelium growth rate are the same for both high and low light conditions, which should result in the same mass of mycelia on each set of roots. However because the low light condition has a smaller root mass (root mass is directly proportional to shoot mass), under certain conditions a mass of mycelium on the low light roots can occupy enough of the roots to cause wilting, that might not be enough to affect the larger roots of the high light condition.

## **7.5 DISCUSSION:**

### **7.5.1 Overview of significant findings**

The results of the simulation indicate there is a substantial decrease in the frequency of *Pythium* outbreaks when lower nutrient solution temperatures are used. In the one pond system of production, every two degrees Celsius drop approximately halves the frequency. This trend holds for the three crop durations (12, 14 and 16 days in pond) examined. Similar benefits are seen in the two pond system.

The benefits of reduced nutrient solution temperature are not as apparent when the harvest schedule is controlled by the biomass of the crop, when no supplemental lighting is used. Though the frequency of outbreaks approximately doubles between the 18C and 24C conditions, these frequencies are on the order of several per year, and are much larger than those observed in the supplemental light conditions. Clearly the extended time taken to achieve target biomasses using natural light makes the crops extremely susceptible to *Pythium* outbreaks.

There is also a clear benefit to reducing the time cohorts spend growing in the pond. In the 18C nutrient solution conditions for both the one and two pond systems with supplemental lighting, the frequency of outbreaks was reduced by approximately

66% for each two day reduction in crop length. This trend is also present at the higher temperatures examined. The faster the crop can be grown, the better the chances are of avoiding an outbreak.

Harvest biomass is directly proportional to the daily light integral the crop receives. Doubling the daily light integral results in twice the harvest biomass. Increasing the crop duration also increases the harvest biomass, the cost of which is an increased risk of *Pythium* outbreak. Reduced light integral (and crop biomass) means a smaller root mass, which in turn can make a crop more vulnerable to *Pythium* root infections.

#### 7.5.2 Limitations that affect validity/generalizability

A primary limitation for using the results of this study is it was developed from data collected from experiments conducted with one pure strain of *Pythium aphanidermatum* (selected for demonstrated virulence). Though random values for the parameters describing the growth and development of *Pythium* were used to try to encompass the variability among strains, other species of *Pythium* might not be covered by that range. Specifically *Pythium dissotocum* has been identified as a *Pythium* species found in lower temperature conditions (Bates and Stanghellini, 1984), suggesting that its temperature optimum is lower than *Pythium aphanidermatum*. The 18C temperature condition effective against *Pythium aphanidermatum*, may not work as well if *Pythium dissotocum* is present. In her analysis of the sources of *Pythium* in greenhouse production, Katzman (2003) did not isolate *Pythium dissotocum* in the Cornell greenhouses. However, this does not mean it is not present, nor that it cannot be brought in. Further research is necessary to determine the values of the parameters describing growth and development of *Pythium dissotocum*.

Another limitation to the model is it was assumed there are no ill effects from physically moving a cohort from one pond to another. This assumption was made to simplify the growth model. In lettuce production, during respacing there can be a measurable impact on the growth rate, following moving of plants from one pond to another. Further research is necessary to quantify the effects of moving plants from pond to pond.

It is important to note the model assumes there is no effect of plant stress on the effects of the disease. Should the crop be stressed by too much light, too high an aerial temperature, mineral or oxygen deficiencies, they may become more vulnerable to *Pythium* infection, resulting in a faster spread of the infection than would be predicted. Disease work was conducted primarily in growth chambers with tightly regulated environmental parameters, or in the greenhouse during the winter and spring months where temperatures are under good control.

To simplify the simulation, no direct interaction between the crop growth model and the greenhouse model was assumed. This assumption was justified by having multiple cohorts in the pond at the same time, conditions remain relatively constant throughout the simulation. Having many ages of the crop present at the same time would tend to balance out the whole crop transpiration, and canopy radiation properties.

The reliability of the components of the greenhouse were not considered. Failures of equipment such as the cooling system, lights, pumps, nutrient treatment systems and control hardware/software are likely to be noticed immediately (Ferentinos 2002), or soon enough to not make a significant difference to the final results of the analysis. The pond is a large heat reservoir and not susceptible to rapid changes of temperature. An analysis of the failure modes of greenhouse components was beyond the scope of this study.

### 7.5.3 Implications for application

One clear recommendation of this study is the justification for light integral control. Harvesting on a predictable schedule is a benefit in itself, but supplemental lighting under Ithaca, NY conditions is absolutely essential to ensure the crop spends the minimum amount of time in the pond. Shading is also an important aspect of integral control. Shading provides a consistent growth rate throughout the year, and also helps protect the crop from excessive light levels and the higher temperatures and stresses associated with them.

The results of this simulation are not intended as an accurate measure of the risk involved with each production strategy, rather, the values of the results are in comparing the risks of the different systems. Further, the model itself can provide insight to a grower in developing their specific strategy for dealing with *Pythium*. Careful examination of the roots of the crop should become a frequent activity in hydroponic spinach production. Such inspections could identify a looming problem and trigger strategies to avoid or minimize the effects of an outbreak. Browning of the roots might trigger a grower to switch production to a two pond system until the system cleans out. It is possible to envision complex production systems that make use of the benefits of the systems examined in the study. The two pond system could be further expanded into a three pond system, where one third of the crop growth is spent in each pond. Or perhaps, a three pond system where the crop is moved out of the first pond after 1/3 of its growth cycle when it is still small, and unentangled. It would then go to a second pond where it would spend the remainder of its growth cycle. The second finishing ponds would alternate (sharing the first pond as a common source). If one of the secondary growth ponds showed signs of *Pythium* damage the system would change to a true three pond system where material from the

uninfected second pond would be moved to the third pond until any infection cleans up.

Implementing risk aversion strategies requires a financial investment by the grower. Nutrient solution chillers must be purchased and sized based on expected cooling loads. Supplemental lighting systems must be sized to provide an adequate light level, which is specific to the location and construction of the greenhouse. Extra labor and materials costs are required for a two pond system, as is more frequent harvesting on shorter crop cycles. All of these factors need to be taken into consideration when growers are formulating spinach production systems that suit their particular needs.

## CHAPTER 8: CONCLUSIONS

*Pythium aphanidermatum* has been identified as the primary roadblock to commercial production of hydroponic spinach. To address this issue, Katzman (2003) examined the effects of temperature, inoculum concentration and timing on the development and spread of the disease. Continuing with this work, NYSERDA commissioned a study to identify treatment techniques to remove *Pythium* from the nutrient solution (Albright et al., 2007). This information forms the basis for this dissertation, the primary objective of which is to quantitatively compare the risk of *Pythium* outbreak in different hydroponic baby spinach production systems. Such information could be used by a potential grower to select a system suitable for their risk comfort level, financial and physical conditions. An additional use of this thesis is the identification of production systems that warrant further investigation/validation.

To achieve this objective a number of tasks were undertaken. These tasks, with any associated conclusions, are as follow:

1. To investigate current and non-traditional techniques for the control of *Pythium aphanidermatum* in hydroponic nutrient solutions.

Chapter two of this dissertation describes development of a seedling bioassay to quickly determine the effectiveness of different nutrient solution treatment techniques. The bioassay allowed a quantitative comparison of the four techniques selected for examination; electrochemical, thermal, sonic and ultraviolet sterilization. All of the techniques investigated proved capable of some reduction in the concentration of zoospores in solution, with sonication providing the best performance; however, Electrochemical treatment was found to be largely ineffective.

Following this initial test of treatment technologies, ultraviolet sterilization, and filtration were selected for further hydroponic production testing. These

technologies were selected for their effectiveness in removing zoospores from solution and the ease of adapting them to in-line flow use. Chapter three of this dissertation presents the results of continuous multi-cohort production with the two active treatment techniques (UV and filtration) and a passive technique of reducing the nutrient solution temperature. The conclusions of this chapter were that:

- Inline treatment of the nutrient solution was ineffective in controlling *Pythium* zoospore damage in pond production. Filtration and UV failed to prevent the migration of zoospores from infected material. UV treatment negatively affected uninoculated plants by destroying the chelator used to keep iron soluble.
- Reducing the nutrient solution temperature to 20C was an effective means of halting the spread of disease in a 14 day crop cycle. By increasing the time it takes for *Pythium* to develop and reproduce, it is possible to harvest and remove infected material before it has a chance to release zoospores.

2. To apply the findings of the solution treatment techniques in the development of production strategies.

Temperature reduction of the nutrient solution was shown to be an effective means of controlling *Pythium aphanidermatum* in pond production. Reduced temperatures were shown by Katzman (2003) to slow the growth and reproductive rate of *Pythium a.* By increasing the reproductive cycle of *Pythium a.* to longer than the crop cycle, any infected plants were harvested/removed before having an opportunity to release zoospores and further spread the infection. One temperature (20C) and one crop duration (14 days) was investigated in chapter three. Other temperatures selected for further investigation were, 18C, 20C, 22C, 24C and uncontrolled (naturally occurring). Coupled within these temperatures, crop durations of 12, 14 and 16 days were

selected. These temperature and duration combinations represent the likely range of options that a grower would select.

A further development investigated the use of two ponds. Cohorts older than half the crop duration are placed in a second “finishing” pond. Any zoospore release in the second pond would not infect material in the initial starting pond. Thus an infection in the initial pond would hopefully die out faster than in a single pond. Once the initial pond has cleaned itself, the material moving to the second pond should also be clean.

A third aspect of production strategy investigated was the use of daily light integral control. Integral control provides a consistent amount of light to the crop through supplemental lighting and shading. This control provides a consistent amount of growth year round, and allows harvesting on a set time schedule, as opposed to natural lighting only where harvesting is performed when the crop reaches an acceptable size.

The conclusions of chapter three were that:

- because nutrient solution temperature is so important to the development of *Pythium*, temperatures of 18, 20, 22 and 24 would be investigated, along with an uncontrolled temperature condition,
- the longer a crop spends in a pond, the greater the risk of *Pythium* completing its reproductive cycle and causing a new outbreak, and so crop durations of 12, 14 and 16 days in pond were investigated, both with one and two pond systems,
- the use of supplemental lighting and shading ensures a predictable harvest schedule, and to investigate this importance from a disease perspective, the model was run without light integral control and harvests conducted when the crop reached target biomasses.



3. To develop a linked greenhouse, crop, and disease model to better understand the dynamics of a *Pythium aphanidermatum* infection and to simulate and evaluate different production strategies.

Chapters four, five and six detail development of a greenhouse simulation model, a hydroponic spinach production model, and a *Pythium* disease model. The greenhouse simulation uses the assumption of stepwise steady state heat and moisture flow within a greenhouse to estimate temperatures of layers within the greenhouse. Specifically it is capable of predicting nutrient solution and aerial temperatures based on outdoor air temperature, relative humidity, and solar radiation. The spinach growth model adapts the SUCROS framework for hourly prediction of spinach growth within a greenhouse equipped with supplemental lighting. The *Pythium* disease model was developed from observations made by Katzman (2003). Specifically it tracks the mass of mycelium present on each cohort of spinach, and predicts the timing and spread of the infection to other cohorts. Damage to the roots by the mycelium affects the growth of the spinach crop. As the spread and development of *Pythium* is highly temperature dependent, the greenhouse simulation model was necessary to predict effects of common greenhouse environmental controls on the nutrient solution and aerial temperatures, as well as level of supplemental light.

The conclusions of chapters four, five and six were that:

- though lacking direct data to validate the greenhouse simulation model, the values predicted are within reason, and are typical of observed conditions,
- the mechanistically based SUCROS spinach growth model predicts growth with reasonable accuracy and provides a good means of scaling growth based on damage to disease,

- the *Pythium* disease model developed from Katzman's (2003) data reasonably predicts the observed development and spread of *Pythium aphanidermatum* in multi-cohort pond production.
4. To simulate production strategies using the greenhouse crop disease model to develop expected risk profiles for each strategy.

To simulate different production strategies, distributions were assumed for each of the seven spinach growth, and twelve *Pythium* disease model variables. Monte-Carlo simulation was then used to model *Pythium* outbreaks. The results of the simulations were estimates of the frequency of *Pythium* outbreak. The conclusions of chapter seven were:

- The simulation suggests that every two degree drop in the nutrient solution, approximately halves the frequency of *Pythium* outbreak within the temperature range of 18C to 24C,
- every two day reduction of the amount of time the crop spends in pond, approximately halves the frequency of *Pythium* outbreaks, within the crop production range of 12 to 16 days,
- a two pond system can also approximately halve the frequency of outbreaks, but only at a temperature of 18C. Outbreak frequency increases with increasing temperature until there is no difference between a one and two pond system at 24C,
- at the highest temperature investigated, 24C, there was little difference between a one and two pond system,
- light integral control was essential to the successful production of spinach. Production strategies that relied on natural light failed rapidly, with outbreak

frequencies on the order of several per year, as opposed to fractions per year, observed in the other conditions with light integral control.

- The productivity of spinach crops is directly proportional to the amount of light the crop receives. Doubling the amount of light a crop receives over its growth cycle, doubles the harvest biomass. Similarly lengthening the growth cycle increases the harvest biomass (at the cost of increasing the risk of *Pythium* outbreak.)

## APPENDIX

The appendix contains a listing of most of the functions used in the greenhouse, spinach disease and production models. Code relating to the graphical user interface (GUI) was not included. Another omission for reasons of copyright protection is the coding of the Light and Shade System Implementation (LASSI).

```
// Functions used for the Greenhouse Simulation model
//*****
//*****

private double[][] getDataArray(int nPoints){

// Initialize data arrays
double[][] greenhouseData      = new double[nPoints+1][21];
double[] newTempsAndConds      = new double[18];
double[] tempsCondsTm1         = new double[18];
double[] ctrlStatus             = new double[6];
double[] enviroConds           = new double[4];
double[][] enviroData          = new double[9000][3];
double[] times                  = new double[2];
double[] lightData              = new double[20];
double[] cropParams             = new double[20];
double[] canopyRadProfileVals  = new double[7];
double[] aboveCanopyRadiationVals = new double[4];
double[] solarRadiationParams  = new double[12];
double[] lightingVals           = new double[6];
double[] transmissivities       = new double[2];
double[] CO2LASSIsets          = new double[3];
double[] LASSIsets              = new double[5];

// Initialize variables
double convertToMols            = 2.08135E-6; // Mol/m2 / J/m2
double ambCO2                   = 400.0; // ambient CO2 concentration ppm
double LAT                       = 42.3; // latitude of Ithaca, NY
double day                       = 0; // julian day
double time                      = 0; // hour of the day (0 to 23)
double sunriseHour;
double sunsetHour;

// Initial conditions:
double tSoilInf                  = 9.88 + 273.15;
double tSoilTm1                  = 12.54 + 273.15;
double tConcreteTm1              = 19.55 + 273.15;
double tWaterTm1                 = 20.10 + 273.15;
double tMediaTm1                  = 19.33 + 273.15;
double tSurfaceTm1                = 19.21 + 273.15;
```

```

double tCropTm1          = 18.38 + 273.15;
double tGH1AirTm1       = 19.0 + 273.15;
double tShadeTm1        = 16.23 + 273.15;
double tGH2AirTm1       = 14.20 + 273.15;
double tGlassTm1        = 12.16 + 273.15;
double tAmbient          = 5 + 273.15;
double thetaSun          = 0;
double tSky              = 5 + 273.15;
double tGHAirTm1        = tGH1AirTm1;
double humAmbient        = 0.4;
double humGH1AirTm1     = 0.4;
double humGHAirTm1      = 0.4;
double humGH2AirTm1     = 0.4;
double ventRate          = Double.parseDouble(jTextField11.getText()) * AREA;
double qHeater           = 0.0;
double infilRate         = 0.0;

```

```

tempsCondsTm1[0] = tSoilTm1;
tempsCondsTm1[1] = tConcreteTm1;
tempsCondsTm1[2] = tWaterTm1;
tempsCondsTm1[3] = tMediaTm1;
tempsCondsTm1[4] = tSurfaceTm1;
tempsCondsTm1[5] = tCropTm1;
tempsCondsTm1[6] = tGH1AirTm1;
tempsCondsTm1[7] = tShadeTm1;
tempsCondsTm1[8] = tGH2AirTm1;
tempsCondsTm1[9] = tGlassTm1;
tempsCondsTm1[10] = tSoilInf;
tempsCondsTm1[11] = tGHAirTm1;
tempsCondsTm1[12] = humGHAirTm1;
tempsCondsTm1[13] = humGH1AirTm1;
tempsCondsTm1[14] = humGH2AirTm1;
tempsCondsTm1[15] = ventRate;
tempsCondsTm1[16] = qHeater;
tempsCondsTm1[17] = infilRate;

```

```

times[0]    = day;
times[1]    = time;

```

```

lightData[0] = 0; // current light intensity (umol/s)
lightData[1] = 0; // sumTotalPPF (Mols/m2)
lightData[2] = 0; // lightsOn? (double)
lightData[3] = 0; // shadesOpen? (double)
lightData[4] = 17; // target Moles
lightData[5] = 42; // latitude
lightData[6] = 0.7; // transMVTY
lightData[7] = 0.5; // shadeFactor
lightData[8] = 180; // amount of mol PPF/time supp. lights provide
lightData[9] = 0.0036; // microMol/m^2/s PPF provided by lights
lightData[10] = 22; // offpeak start
lightData[11] = 7; // offpeak end
lightData[12] = 0.0; // sumSolarPPF (Mols/m2)
lightData[13] = 400.0; // target CO2 concentration (ppm)
lightData[14] = 0.0; // average light level past hour
lightData[15] = 0.0; // estimated outdoor temperature next hour

```

```

lightData[16] = 0.0; // sunriseHour
lightData[17] = 0.0; // sunsetHour
lightData[18] = 400.0; // ambient CO2 concentration (ppm)
lightData[19] = 0.55; // percentage of sunlight that is PAR

ctrlStatus[0] = 0; // lighting on? 1 for on, 0 for off
if (jToggleButton4.isSelected()) ctrlStatus[1] = 1; // shade closed?
else ctrlStatus[1] = 0;
if (jToggleButton2.isSelected()) ctrlStatus[2] = 1; // pad cooling
else ctrlStatus[2] = 0;
ctrlStatus[3] = 0; // heat output by the lights

enviroConds[0] = tAmbient;
enviroConds[1] = humAmbient;
enviroConds[2] = thetaSun;
enviroConds[3] = tSky;

// input the first data value (initial conditions) into the temperature array
greenhouseData[0][0] = tSoilInf;
greenhouseData[0][1] = tSoilTm1;
greenhouseData[0][2] = tConcreteTm1;
greenhouseData[0][3] = tWaterTm1;
greenhouseData[0][4] = tMediaTm1;
greenhouseData[0][5] = tSurfaceTm1;
greenhouseData[0][6] = tCropTm1;
greenhouseData[0][7] = tGH1AirTm1;
greenhouseData[0][8] = tShadeTm1;
greenhouseData[0][9] = tGH2AirTm1;
greenhouseData[0][10] = tGlassTm1;
greenhouseData[0][11] = tAmbient;
greenhouseData[0][12] = tSky;
greenhouseData[0][13] = ventRate;
greenhouseData[0][14] = qHeater;
greenhouseData[0][15] = thetaSun;
greenhouseData[0][16] = 0;
greenhouseData[0][17] = 0;
greenhouseData[0][18] = 0;
greenhouseData[0][19] = 400;
greenhouseData[0][20] = 400;

tSetpoint = 24 + 273.15;

controlOn = true;

enviroData = readDataFile("G:\\ITHACA85to97.TXT",1);

for (int i = 1; i < (nPoints + 1); i++){

    // if/else to maintain 0-24 hour loop
    if( time>=0 && time<=22) {
        time++; // increment an hour for each loop
    } // end if
    else {

```

```

    time = 0; // resets hour back to 0 (midnight) if previously 11 pm
    day++; // increments day to reflect 24 hour period
} // end else

times[0]      = day;
times[1]      = time;

sunriseHour   = Math.floor(calculateSunVals(day,42)[0]);
sunsetHour    = Math.floor(calculateSunVals(day,42)[1]);
transmissivities = calculateTransmissivity(times,sunriseHour,sunsetHour);

lightData[16] = sunriseHour;
lightData[17] = sunsetHour;

// using real data?
if (true){
    enviroConds[0] = enviroData[i][0];
    enviroConds[1] = enviroData[i][1];
    enviroConds[2] = enviroData[i][2];
    enviroConds[3] = calculateTSky(enviroConds[0]);
} // end if

// using Light/Shade Control?
if (true){

    lightData[0] = enviroConds[2]*convertToMols; // current light intensity

    // Decide whether to use regular or CO2 supplemented LASSI
    if (false) {
        // provide CO2LASSI with lightData array, time, day, current CO2 concentration, and
        //next hours outdoor temp
        CO2LASSIsets = CO2LASSI2(lightData, time, day, lightData[18], enviroData[i+1][0]-
273.15);
        lightData[2] = CO2LASSIsets[0];
        lightData[3] = CO2LASSIsets[1];
        lightData[13] = CO2LASSIsets[2];

        if (CO2LASSIsets[3] == 1){
            lightData[18] = lightData[13];
        } // end if
        else {
            lightData[18] = ( lightData[18] - ambCO2 ) * Math.exp( -CO2LASSIsets[4] ) +
ambCO2;
        } // end else

        // update the solarPAR recieved and the totalGrowth
        if (lightData[3] == 1){
            lightData[12] = lightData[12] +
            enviroConds[2]*lightData[19]*convertToMols*lightData[6]*transmissivities[0];
            lightData[1] = lightData[1] +
            (enviroConds[2]*lightData[19]*convertToMols*lightData[6]*transmissivities[0] +
            lightData[2] * lightData[8] * lightData[9])
            * ((Math.log(2.66E4) - Math.log(400)) / (Math.log(2.66E4) -
Math.log(lightData[13])));
        } // end if

```

```

else {
    lightData[12] = lightData[12] +
    enviroConds[2]*lightData[19]*convertToMols*lightData[6]*transmissivities[0];

    lightData[1] = lightData[1] +
    (enviroConds[2]*lightData[19]*convertToMols*lightData[6]*transmissivities[0]*
    lightData[7] + lightData[2] * lightData[8] * lightData[9])
    * ((Math.log(2.66E4) - Math.log(400)) / (Math.log(2.66E4) -
Math.log(lightData[13])));
} // end else

if (CO2LASSIsets[1] == 0)    ctrlStatus[1] = 1; // shade closed?
else                        ctrlStatus[1] = 0;

if (CO2LASSIsets[0] == 0)    ctrlStatus[0] = 0; // lightsON?
else                        ctrlStatus[0] = 1;

} // end if
else {
    LASSIsets                = LASSI(sunriseHour, sunsetHour, time, day, lightData);
    lightData[2]              = LASSIsets[0];
    lightData[3]              = LASSIsets[1];

    // update the solarPAR recieved and the totalGrowth
    if (lightData[3] == 1){
        lightData[12] = lightData[12] +
        enviroConds[2]*lightData[19]*convertToMols*lightData[6]*transmissivities[0];
        lightData[1] = lightData[1] +
        (enviroConds[2]*lightData[19]*convertToMols*lightData[6]*transmissivities[0] +
        lightData[2] * lightData[8] * lightData[9]);
    } // end if
    else {
        lightData[12] = lightData[12] +
        enviroConds[2]*lightData[19]*convertToMols*lightData[6]*transmissivities[0]*
        lightData[7];

        lightData[1] = lightData[1] +
        (enviroConds[2]*lightData[19]*convertToMols*lightData[6]*transmissivities[0]*
        lightData[7]
        + lightData[2] * lightData[8] * lightData[9]);
    } // end else

    if (LASSIsets[1] == 0)    ctrlStatus[1] = 1; // shade closed?
    else                        ctrlStatus[1] = 0;

    if (LASSIsets[0] == 0)    ctrlStatus[0] = 0; // lightsON?
    else                        ctrlStatus[0] = 1;

} // end if

} // end if
else{
    lightData[1] = lightData[1] +
    enviroConds[2]*lightData[19]*convertToMols*lightData[6]*transmissivities[0];
} // end else

```



```

// check for closing shades at night (Irrespective of LASSI)
if (true){
  if ((time > calculateSunVals(day, 42)[1]) || (time < calculateSunVals(day,42)[0])){
    ctrlStatus[1] = 1;
  } // end if
} // end if

// using temperature control?
if (controlOn){
  if ((i%24 >= sunriseHour) && (i%24 <= sunsetHour)){
    tSetpoint = daysetpoint + 273.15;
  } // end if
  else{
    tSetpoint = nightsetpoint + 273.15;
  } // end else
  newTempsAndConds = determineHeatingVenting(tempsCondsTm1, ctrlStatus,
enviroConds);
} // end if
else{
  tempsCondsTm1[15] = 0.0;
  tempsCondsTm1[16] = 0.0;
  newTempsAndConds = calculateNewTemps(tempsCondsTm1, ctrlStatus, enviroConds);
} // end else

greenhouseData[i][0] = newTempsAndConds[10];
greenhouseData[i][1] = newTempsAndConds[0];
greenhouseData[i][2] = newTempsAndConds[1];
greenhouseData[i][3] = newTempsAndConds[2];
greenhouseData[i][4] = newTempsAndConds[3];
greenhouseData[i][5] = newTempsAndConds[4];
greenhouseData[i][6] = newTempsAndConds[5];
greenhouseData[i][7] = newTempsAndConds[6];
greenhouseData[i][8] = newTempsAndConds[7];
greenhouseData[i][9] = newTempsAndConds[8];
greenhouseData[i][10] = newTempsAndConds[9];
greenhouseData[i][11] = enviroConds[0];
greenhouseData[i][12] = enviroConds[3];
greenhouseData[i][13] = newTempsAndConds[15];
greenhouseData[i][14] = newTempsAndConds[16];
greenhouseData[i][15] = enviroConds[2];
greenhouseData[i][16] = lightData[3];
greenhouseData[i][17] = lightData[2];
greenhouseData[i][18] = lightData[1];
greenhouseData[i][19] = lightData[13];
greenhouseData[i][20] = lightData[18];

tempsCondsTm1 = newTempsAndConds;

// reset the daily light integral (total and solar)
if (time == Math.floor(sunriseHour)){
  lightData[1] = 0;
  lightData[12] = 0;
} // end if

```



```
tAirLow = newTempsAndConds[6];
tempsCondsTm1[16] = hiRate;
newTempsAndConds = calculateNewTemps(tempsCondsTm1, ctrlStatus, enviroConds);
tAirHigh = newTempsAndConds[6];
avgRate = (hiRate + lowRate)/2;
tempsCondsTm1[16] = avgRate;
newTempsAndConds = calculateNewTemps(tempsCondsTm1, ctrlStatus, enviroConds);
tAirAvg = newTempsAndConds[6];

if (tAirAvg < tSetpoint){
    lowRate = avgRate;
}
else {
    hiRate = avgRate;
}

} while ( Math.abs(tAirLow - tAirHigh) > tTolerance);

return newTempsAndConds;

} // end searchgHeating

private double[] searchVenting(double hiRate, double lowRate, double[] tempsCondsTm1, double[]
ctrlStatus, double[] enviroConds) {
    // search for optimal venting rate between two specified levels using bisection
    double[] newTempsAndConds = tempsCondsTm1;
    double avgRate = 0.0;
    double tAirLow = 0.0;
    double tAirHigh = 0.0;
    double tAirAvg = 0.0;

    do {
        tempsCondsTm1[15] = lowRate;
        newTempsAndConds = calculateNewTemps(tempsCondsTm1, ctrlStatus, enviroConds);
        tAirLow = newTempsAndConds[6];
        tempsCondsTm1[15] = hiRate;
        newTempsAndConds = calculateNewTemps(tempsCondsTm1, ctrlStatus, enviroConds);
        tAirHigh = newTempsAndConds[6];
        avgRate = (hiRate + lowRate)/2;
        tempsCondsTm1[15] = avgRate;
        newTempsAndConds = calculateNewTemps(tempsCondsTm1, ctrlStatus, enviroConds);
        tAirAvg = newTempsAndConds[6];

        if (tAirAvg < tSetpoint){
            hiRate = avgRate;
        }
        else {
            lowRate = avgRate;
        }
    } while ( Math.abs(tAirLow - tAirHigh) > tTolerance);

    return newTempsAndConds;
} // end searchVenting
```

////////////////////////////////////

```

private double[] determineHeatingVenting(double[] tempsCondsTm1, double[] ctrlStatus, double[]
enviroConds){
    double[] newTempsAndConds = new double[17];
    double tMaxVent          = 0.0;
    double tMaxHeat          = 0.0;
    double tNoHeatVent       = 0.0;

    //check to determine if either the heating system or venting system needs to be maxed out
    tempsCondsTm1[15] = MAXVENTRATE;
    tempsCondsTm1[16] = 0.0;
    newTempsAndConds = calculateNewTemps(tempsCondsTm1, ctrlStatus, enviroConds);
    tMaxVent          = newTempsAndConds[6];
    if (tMaxVent > tSetpoint + tTolerance){
        return newTempsAndConds;
    }
    tempsCondsTm1[15] = 0.0;
    tempsCondsTm1[16] = QHEATERMAX;
    newTempsAndConds = calculateNewTemps(tempsCondsTm1, ctrlStatus, enviroConds);
    tMaxHeat          = newTempsAndConds[6];
    if (tMaxHeat < tSetpoint - tTolerance){
        return newTempsAndConds;
    }

    // At this point of the code must be within a range that can be affected by a control Action
    // determine what happens without either heating or cooling
    tempsCondsTm1[15] = 0.0;
    tempsCondsTm1[16] = 0.0;
    newTempsAndConds = calculateNewTemps(tempsCondsTm1, ctrlStatus, enviroConds);
    tNoHeatVent       = newTempsAndConds[6];

    if (tNoHeatVent >= tSetpoint + tTolerance){
        // Need to search for venting rate
        newTempsAndConds = searchVenting(MAXVENTRATE, 0.0, tempsCondsTm1, ctrlStatus,
enviroConds);
    }
    else {
        // Need to search for heating rate
        newTempsAndConds = searchHeating(QHEATERMAX, 0.0, tempsCondsTm1, ctrlStatus,
enviroConds);
    }

    return newTempsAndConds;
} // end determineHeatingVenting

```

////////////////////////////////////

```

private double[] calculateNewTemps(double[] previousTempsAndConds, double[] greenhouseConds,
double[] ambientConds){

    // the array of temperatures humidities, and heat/vent rates to return
    double newTempsAndConds[] = new double[18];

    tSoilTm1           = previousTempsAndConds[0];
    tConcreteTm1       = previousTempsAndConds[1];
    tWaterTm1          = previousTempsAndConds[2];

```

```

tMediaTm1      = previousTempsAndConds[3];
tSurfaceTm1   = previousTempsAndConds[4];
tCropTm1      = previousTempsAndConds[5];
tGH1AirTm1    = previousTempsAndConds[6];
tShadeTm1     = previousTempsAndConds[7];
tGH2AirTm1    = previousTempsAndConds[8];
tGlassTm1     = previousTempsAndConds[9];
tSoilInf       = previousTempsAndConds[10];
tGHAirTm1     = previousTempsAndConds[11];
humGHAirTm1   = previousTempsAndConds[12];
humGH1AirTm1  = previousTempsAndConds[13];
humGH2AirTm1  = previousTempsAndConds[14];
ventRate      = previousTempsAndConds[15];
qHeater       = previousTempsAndConds[16];
infilRate     = previousTempsAndConds[17];

if (greenhouseConds[0] == 1) lightsOn = true;
else                          lightsOn = false;
if (greenhouseConds[1] == 1) shadesClosed = true;
else                          shadesClosed = false;
if (greenhouseConds[2] == 1) padsOn = true;
else                          padsOn = false;

tAmbient      = ambientConds[0];
humAmbient    = ambientConds[1];
thetaSun      = ambientConds[2];
tSky          = ambientConds[3];

psychroAmb    = calculatePsychroVals(tAmbient, humAmbient, Patm);
WAmb         = psychroAmb[5];
VsaAmb       = psychroAmb[6];
enthalpyAmb  = psychroAmb[7];
rhoAmb       = psychroAmb[8];

// set conditions of the incoming air from vent if the evaporative cooling pads are on
if (padsOn){
    psychroVent    = calculateStateOfAirFromPad(psychroAmb, 0.75);
    tVentAir      = psychroVent[0];
    WVent        = psychroVent[5];
    VsaVent      = psychroVent[6];
}
else {
    psychroVent    = psychroAmb;
    tVentAir      = psychroAmb[0];
    WVent        = psychroAmb[5];
    VsaVent      = psychroAmb[6];
}

// Based on whether the shades are open or not solve for temps using the shade model or not
if (!shadesClosed){

    // arrays to hold linear equations
    Matrix a      = new Matrix(new double[8][8]);
    Matrix b      = new Matrix(new double[8][1]);
    Matrix X      = new Matrix(new double[8][1]);

```

```

psychroGHTm1 = calculatePsychroVals(tGHAirTm1, humGHAirTm1, Patm);
WGHTm1       = psychroGHTm1[5];
VsaGHTm1     = psychroGHTm1[6];
enthalpyGHTm1 = psychroGHTm1[7];
rhoGHTm1     = psychroGHTm1[8];

VGH = AREA * ZGHAIR;

thetaGlass    = thetaSun*glassTrans*AREA; // Sunlight level after passing through glass
enviroVals[0] = thetaGlass + thetaL;      // Net all wave Radiation reaching canopy W/m2
enviroVals[1] = (1-CANOPYREFL)*(thetaGlass+thetaL)*Math.exp(-
1*K*LAI)*ALPHAM;
enviroVals[2] = 0.12;                    // air speed at height of 2m, m/s

evapoTranspiration = calculateEvapoTranspiration(psychroGHTm1, enviroVals, tCropTm1,
AREA);

WGH = (((WAmb/VsaAmb)*infilRate + (WVent/VsaVent)*ventRate + evaporation[0] +
evapoTranspiration[0] + (WGHTm1/VsaGHTm1)*VGH)/(VGH + ventRate +
infilRate))*VsaGHTm1;

tSurfCropAvgCubed = Math.pow(((tSurfaceTm1 + tCropTm1) / 2),3);
tSurfGlassAvgCubed = Math.pow(((tSurfaceTm1 + tGlassTm1) / 2),3);
tGlassCropAvgCubed = Math.pow(((tGlassTm1 + tCropTm1) / 2),3);
tGlassSkyAvgCubed = Math.pow(((tGlassTm1 + tSky) / 2),3);

// energy balance soil layer
a00 = -1 * (1/(ZSOIL/KASOIL + ZCONCRETE/KACONCRETE) + KASOIL/(ZSOIL/2) +
TMASSSOIL);
a01 = 1/(ZSOIL/KASOIL + ZCONCRETE/KACONCRETE);
b0 = -1 * ((KASOIL/(ZSOIL/2))*tSoilInf + TMASSSOIL*tSoilTm1);
a.set(0,0,a00);
a.set(0,1,a01);
b.set(0,0,b0);

// energy balance concrete layer
a10 = 1/(ZSOIL/KASOIL + ZCONCRETE/KACONCRETE);
a11 = -1 * (HAWATER + (1/(ZSOIL/KASOIL + ZCONCRETE/KACONCRETE)) +
TMASSCONCRETE);
a12 = HAWATER;
b1 = -1 * (TMASSCONCRETE * tConcreteTm1);
a.set(1,0,a10);
a.set(1,1,a11);
a.set(1,2,a12);
b.set(1,0,b1);

// energy balance water layer
a21 = HAWATER;
a22 = -1 * (2*HAWATER + TMASSWATER);
a23 = HAWATER;
b2 = -1 * (TMASSWATER * tWaterTm1 + PUMPHEAT);
a.set(2,1,a21);
a.set(2,2,a22);
a.set(2,3,a23);

```

```

b.set(2,0,b2);

// energy balance media layer
a32 = HAWATER;
a33 = -1 * (KAMEDIA/(ZMEDIA/2) + HAWATER + TMASSMEDIA);
a34 = KAMEDIA/(ZMEDIA/2);
b3 = -1 * (TMASSMEDIA * tMediaTm1);
a.set(3,2,a32);
a.set(3,3,a33);
a.set(3,4,a34);
b.set(3,0,b3);

// energy balance media surface layer
a43 = KAMEDIA/(ZMEDIA/2);
a44 = -1*(hASurface + RADCNSTSM*LAI*tSurfCropAvgCubed + KAMEDIA/(ZMEDIA/2)+
RADCNSTSM*(1-LAI)*tSurfGlassAvgCubed);
a45 = RADCNSTSM*LAI*tSurfCropAvgCubed;
a46 = hASurface;
a47 = RADCNSTSM*(1-LAI)*tSurfGlassAvgCubed;
b4 = -1*((1-CANOPYREFL)*(thetaGlass+thetaL)*Math.exp(-1*K*LAI)*ALPHAM);
a.set(4,3,a43);
a.set(4,4,a44);
a.set(4,5,a45);
a.set(4,6,a46);
a.set(4,7,a47);
b.set(4,0,b4);

// energy balance canopy layer
a54 = RADCNSTSC*tSurfCropAvgCubed;
a55 = -1*(hACrop + RADCNSTSC*tGlassCropAvgCubed +
RADCNSTSC*tSurfCropAvgCubed);
a56 = hACrop;
a57 = RADCNSTSC*tGlassCropAvgCubed;
b5 = -1*((1-CANOPYREFL)*(thetaGlass+thetaL)*(1-Math.exp(-1*K*LAI)) -
evapoTranspiration[1]);
a.set(5,4,a54);
a.set(5,5,a55);
a.set(5,6,a56);
a.set(5,7,a57);
b.set(5,0,b5);

// energy balance air
a64 = (hASurface );
a65 = (hACrop );
a66 = -1*(hAGlass + hACrop + hASurface + (1006 + WGH*1805)*(VGH + ventRate +
infilRate)/VsaGHTm1);
a67 = hAGlass;
b6 = -1*(qHeater + qLamps);
b6 = b6 - (1006 *(tAmbient -273.15) + WAmb*(2501000 + 1805*(tAmbient - 273.15))) *
(infilRate/VsaAmb);
b6 = b6 - (1006 *(tVentAir -273.15) + WVent*(2501000 + 1805*(tVentAir - 273.15))) *
(ventRate/VsaVent);
b6 = b6 - ((1006*(tGHAirTm1 - 273.15) + WGHTm1*(2501000 + 1805*(tGHAirTm1 -
273.15)))*VGH/VsaGHTm1);

```

```

        b6 = b6 + (1006*(-273.15) + WGH*(2501000 + 1805*(-273.15)))*(VGH + ventRate +
infilRate)/VsaGHTm1;
        a.set(6,4,a64);
        a.set(6,5,a65);
        a.set(6,6,a66);
        a.set(6,7,a67);
        b.set(6,0,b6);

        // energy balance glass
        a74 = RADCNSTSM*(1-LAI)*tSurfGlassAvgCubed;
        a75 = RADCNSTSC*LAI*tGlassCropAvgCubed;
        a76 = hAGlass;
        a77 = -1*(hAGlassAmb + RADCNSTSG*tGlassSkyAvgCubed + hAGlass +
RADCNSTSC*LAI*tGlassCropAvgCubed + RADCNSTSM*(1-LAI)*tSurfGlassAvgCubed);
        b7 = -1 * (hAGlassAmb*tAmbient + (1-glassRefl)*thetaSun*(1-glassTrans) +
RADCNSTSG*tGlassSkyAvgCubed*tSky);
        a.set(7,4,a74);
        a.set(7,5,a75);
        a.set(7,6,a76);
        a.set(7,7,a77);
        b.set(7,0,b7);

        // solve the linear equations using matrix tools
        X = a.solve(b);

        // Assign the solutions to the previous timestep values
        newTempsAndConds[0] = X.get(0,0); // tSoilTm1
        newTempsAndConds[1] = X.get(1,0); // tConcreteTm1
        newTempsAndConds[2] = X.get(2,0); // tWaterTm1
        newTempsAndConds[3] = X.get(3,0); // tMediaTm1
        newTempsAndConds[4] = X.get(4,0); // tSurfaceTm1
        newTempsAndConds[5] = X.get(5,0); // tCropTm1
        newTempsAndConds[6] = X.get(6,0); // tGHAirTm1 (GH1AirTm1)
        newTempsAndConds[7] = X.get(6,0); // tGHAirTm1 (set withdrawnshade temp to air
temp)
temp)
        newTempsAndConds[8] = X.get(6,0); // tGHAirTm1 (set GH2Air = greenhouse air
temp)
        newTempsAndConds[9] = X.get(7,0); // tGlassTm1
        newTempsAndConds[10] = tSoilInf; // tSoilInf
        newTempsAndConds[11] = X.get(6,0); // tGHAirTm1
        newTempsAndConds[12] = calculateRelativeHumidity(X.get(6,0), Patm, WGH);
        newTempsAndConds[13] = humGHAirTm1; // (humGH1)
        newTempsAndConds[14] = humGHAirTm1; // (humGH2)
        newTempsAndConds[15] = ventRate; // Venting rate
        newTempsAndConds[16] = qHeater; // Heating rate
        newTempsAndConds[17] = infilRate; // infiltration rate
    } // End if for shades
else {

        // arrays to hold linear equations
        Matrix A = new Matrix(new double[10][10]);
        Matrix B = new Matrix(new double[10][1]);
        Matrix x = new Matrix(new double[10][1]);

        psychroGH1Tm1 = calculatePsychroVals(tGH1AirTm1, humGH1AirTm1, Patm);

```



```

    WGH1Tm1      = psychroGH1Tm1[5];
    VsaGH1Tm1    = psychroGH1Tm1[6];
    enthalpyGH1Tm1 = psychroGH1Tm1[7];
    rhoGH1Tm1    = psychroGH1Tm1[8];
    psychroGH2Tm1 = calculatePsychroVals(tGH2AirTm1, humGH2AirTm1, Patm);
    WGH2Tm1      = psychroGH2Tm1[5];
    VsaGH2Tm1    = psychroGH2Tm1[6];
    enthalpyGH2Tm1 = psychroGH2Tm1[7];
    rhoGH2Tm1    = psychroGH2Tm1[8];

    VGH1          = AREA * ZGH1AIR;
    VGH2          = AREA * ZGH2AIR;

    thetaGlass    = thetaSun*glassTrans;    // Sunlight level after passing through glass
    thetaShade     = thetaGlass*shadeTrans;  // Sunlight left after passing through shade
    enviroVals[0]  = thetaShade + thetaL;     // Net all wave Radiation reaching canopy W/m2
    enviroVals[1]  = (1-CANOPYREFL)*(thetaShade+thetaL)*Math.exp(-
1*K*LAI)*ALPHAM;
    enviroVals[2]  = 0.12;                   // air speed at height of 2m, m/s

    evapoTranspiration = calculateEvapoTranspiration(psychroGH1Tm1, enviroVals, tCropTm1,
AREA);

    WGH1 = WAmb*infilRate1/VsaAmb + WVent*ventRate/VsaVent +
WGH1Tm1*VGH1/VsaGH1Tm1;
    WGH1 = WGH1 + (WAmb*infilRate2/VsaAmb +
WGH2Tm1*VGH2/VsaGH2Tm1)*(GH1to2ExRate)/(VGH2 + infilRate2 + GH1to2ExRate);
    WGH1 = (WGH1*VsaGH1Tm1)/(infilRate1 + ventRate + GH1to2ExRate + VGH1 -
Math.pow(GH1to2ExRate,2)*(VGH2 + infilRate2 + GH1to2ExRate));

    WGH2 = WAmb*infilRate2/VsaAmb + WGH1*GH1to2ExRate/VsaGH1Tm1 +
WGH2Tm1*VGH2/VsaGH2Tm1;
    WGH2 = WGH2 * VsaGH2Tm1 / (VGH2 + infilRate2 + GH1to2ExRate);

    tSurfCropAvgCubed = Math.pow((( tSurfaceTm1 + tCropTm1 ) / 2),3);
    tSurfShadeAvgCubed = Math.pow((( tSurfaceTm1 + tShadeTm1 ) / 2),3);
    tShadeCropAvgCubed = Math.pow((( tShadeTm1 + tCropTm1 ) / 2),3);
    tShadeGlassAvgCubed = Math.pow((( tShadeTm1 + tGlassTm1 ) / 2),3);
    tGlassSkyAvgCubed = Math.pow((( tGlassTm1 + tSky ) / 2),3);

    // energy balance soil layer
    a00 = -1 * (1/(ZSOIL/KASOIL + ZCONCRETE/KACONCRETE) + KASOIL/(ZSOIL/2) +
TMASSSOIL);
    a01 = 1/(ZSOIL/KASOIL + ZCONCRETE/KACONCRETE);
    b0 = -1 * ((KASOIL/(ZSOIL/2))*tSoilInf + TMASSSOIL*tSoilTm1);
    A.set(0,0,a00);
    A.set(0,1,a01);
    B.set(0,0,b0);

    // energy balance concrete layer
    a10 = 1/(ZSOIL/KASOIL + ZCONCRETE/KACONCRETE);
    a11 = -1 * (HAWATER + (1/(ZSOIL/KASOIL + ZCONCRETE/KACONCRETE)) +
TMASSCONCRETE);
    a12 = HAWATER;
    b1 = -1 * (TMASSCONCRETE * tConcreteTm1);

```

```

A.set(1,0,a10);
A.set(1,1,a11);
A.set(1,2,a12);
B.set(1,0,b1);

// energy balance water layer
a21 = HAWATER;
a22 = -1 * (2*HAWATER + TMASSWATER);
a23 = HAWATER;
b2 = -1 * (TMASSWATER * tWaterTm1 + PUMPHEAT);
A.set(2,1,a21);
A.set(2,2,a22);
A.set(2,3,a23);
B.set(2,0,b2);

// energy balance media layer
a32 = HAWATER;
a33 = -1 * (KAMEDIA/(ZMEDIA/2) + HAWATER + TMASSMEDIA);
a34 = KAMEDIA/(ZMEDIA/2);
b3 = -1 * (TMASSMEDIA * tMediaTm1);
A.set(3,2,a32);
A.set(3,3,a33);
A.set(3,4,a34);
B.set(3,0,b3);

// energy balance media surface layer
a43 = KAMEDIA/(ZMEDIA/2);
a44 = -1*(hASurface + RADCNSTSM*LAI*tSurfCropAvgCubed + KAMEDIA/(ZMEDIA/2)
+ RADCNSTSM*(1-LAI)*tSurfShadeAvgCubed );
a45 = RADCNSTSM*LAI*tSurfCropAvgCubed;
a46 = hASurface;
a47 = RADCNSTSM*(1-LAI)*tSurfShadeAvgCubed;
b4 = -1*(1-CANOPYREFL)*(thetaShade+thetaL)*Math.exp(-1*K*LAI)*ALPHAM;
A.set(4,3,a43);
A.set(4,4,a44);
A.set(4,5,a45);
A.set(4,6,a46);
A.set(4,7,a47);
B.set(4,0,b4);

// energy balance canopy layer
a54 = RADCNSTSC*tSurfCropAvgCubed;
a55 = -1*(hACrop + RADCNSTSC*tShadeCropAvgCubed +
RADCNSTSC*tSurfCropAvgCubed);
a56 = hACrop;
a57 = RADCNSTSC*tShadeCropAvgCubed;
b5 = -1*((1-CANOPYREFL)*(thetaShade+thetaL)*(1-Math.exp(-1*K*LAI))-
evapoTranspiration[1]);
A.set(5,4,a54);
A.set(5,5,a55);
A.set(5,6,a56);
A.set(5,7,a57);
B.set(5,0,b5);

```

```

// energy balance air TG1
a64 = hASurface;
a65 = hACrop;
a66 = -1 * (hAShade1 + hACrop + hASurface + (1006 + WGH1*1805)*(infilRate1 + VGH1 +
ventRate + GH1to2ExRate)/(VsaGH1Tm1));
a67 = hAShade1;
a68 = (1006 + WGH2*1805) * (GH1to2ExRate)/(VsaGH2Tm1);
b6 = -1 * (qHeater + qLamps);
b6 = b6 - (1006*(tVentAir-273.15) + WVent*(2501000 + 1805*(tVentAir-
273.15)))*(ventRate/VsaVent);
b6 = b6 - (1006*(tAmbient-273.15) + WAmb *(2501000 + 1805*(tAmbient-
273.15)))*(infilRate1/VsaAmb);
b6 = b6 + (1006*(-273.15) + WGH1*(2501000 + 1805*(-273.15)))*(infilRate1 + VGH1 +
ventRate + GH1to2ExRate)/(VsaGH1Tm1);
b6 = b6 - (1006*(-273.15) + WGH2*(2501000 + 1805*(-
273.15)))*(GH1to2ExRate/VsaGH2Tm1);
b6 = b6 - (1006*(tGH1AirTm1 - 273.15) + WGH1Tm1*(2501000 + 1805*(tGH1AirTm1 -
273.15)))*(VGH1/VsaGH1Tm1) - evapoTranspiration[1];

A.set(6,4,a64);
A.set(6,5,a65);
A.set(6,6,a66);
A.set(6,7,a67);
A.set(6,8,a68);
B.set(6,0,b6);

// energy balance shade
a74 = RADCNSTSM*(1-LAI)*tSurfShadeAvgCubed;
a75 = RADCNSTSC*LAI*tShadeCropAvgCubed;
a76 = hAShade1;
a77 = -1*(hAShade2 + RADCNSTSG*tShadeGlassAvgCubed + hAShade1 +
RADCNSTSC*LAI*tShadeCropAvgCubed + RADCNSTSM*(1-LAI)*tSurfShadeAvgCubed);
a78 = hAShade2;
a79 = RADCNSTSG*tShadeGlassAvgCubed;
b7 = -1 * ((1-shadeRefl)*thetaGlass*(1-shadeTrans));
A.set(7,4,a74);
A.set(7,5,a75);
A.set(7,6,a76);
A.set(7,7,a77);
A.set(7,8,a78);
A.set(7,9,a79);
B.set(7,0,b7);

// energy balance air TG2
a86 = (1006 + WGH1*1805) * (GH1to2ExRate)/(VsaGH1Tm1);
a87 = hAShade2;
a88 = -1*(hAGlass + hAShade2 + (1006 + WGH2*1805) * (infilRate2 + GH1to2ExRate +
VGH2)/(VsaGH2Tm1));
a89 = hAGlass;
b8 = -1 * (1006 * (tAmbient - 273.15) + WAmb * (2501000 + 1805 * (tAmbient - 273.15))) *
(infilRate2/VsaAmb);
b8 = b8 - (1006 * (tGH2AirTm1 - 273.15) + WGH2Tm1 * (2501000 + 1805 * (tGH2AirTm1 -
273.15))) * (VGH2/VsaGH2Tm1);
b8 = b8 - (1006 * (-273.15) + WGH1 * (2501000 + 1805 * (-273.15))) *
(GH1to2ExRate/VsaGH1Tm1);

```

```

    b8 = b8 + (1006 * (-273.15) + WGH2 * (2501000 + 1805 * (-273.15))) * (infilRate2 +
GH1to2ExRate + VGH2)/(VsaGH2Tm1);
    A.set(8,6,a86);
    A.set(8,7,a87);
    A.set(8,8,a88);
    A.set(8,9,a89);
    B.set(8,0,b8);

    // energy balance glass
    a97 = RADCNSTSG*tShadeGlassAvgCubed;
    a98 = hAGlass;
    a99 = -1 * (hAGlassAmb + RADCNSTSG*tGlassSkyAvgCubed + hAGlass +
RADCNSTSG*tShadeGlassAvgCubed);
    b9 = -1 * (hAGlassAmb*tAmbient + (1-glassRefl)*thetaSun*(1-glassTrans) +
RADCNSTSG*tGlassSkyAvgCubed*tSky);
    A.set(9,7,a97);
    A.set(9,8,a98);
    A.set(9,9,a99);
    B.set(9,0,b9);

    // solve the linear equations using matrix tools
    x = A.solve(B);

    // Assign the solutions to the previous timestep values
    newTempsAndConds[0] = x.get(0,0); // tSoilTm1
    newTempsAndConds[1] = x.get(1,0); // tConcreteTm1
    newTempsAndConds[2] = x.get(2,0); // tWaterTm1
    newTempsAndConds[3] = x.get(3,0); // tMediaTm1
    newTempsAndConds[4] = x.get(4,0); // tSurfaceTm1
    newTempsAndConds[5] = x.get(5,0); // tCropTm1
    newTempsAndConds[6] = x.get(6,0); // tGH1AirTm1
    newTempsAndConds[7] = x.get(7,0); // tShadeTm1
    newTempsAndConds[8] = x.get(8,0); // tGH2AirTm1
    newTempsAndConds[9] = x.get(9,0); // tGlassTm1
    newTempsAndConds[10] = tSoilInf; // tSoilInf
    newTempsAndConds[11] = calculateAdiabaticMixing(x.get(6,0), WGH1, x.get(8,0),
WGH2, VGH1, VGH2, Patm)[0];// tGHAirTm1
    newTempsAndConds[12] = calculateAdiabaticMixing(x.get(6,0), WGH1, x.get(8,0),
WGH2, VGH1, VGH2, Patm)[1];// humGHTm1
    newTempsAndConds[13] = calculateRelativeHumidity(x.get(6,0), Patm, WGH1); //
(humGH1)
    newTempsAndConds[14] = calculateRelativeHumidity(x.get(8,0), Patm, WGH2); //
(humGH2)
    newTempsAndConds[15] = ventRate; // Venting rate
    newTempsAndConds[16] = qHeater; // Heating rate
    newTempsAndConds[17] = infilRate; // infiltration rate
}
return newTempsAndConds;

} // end calculateNewTemps

////////////////////////////////////

```

```

private double calculateTSky(double tAir){
    double tSky = 0.0552 * Math.pow(tAir,1.5);
    return tSky;
} // end calculateTSky

////////////////////////////////////////////////////////////////////////////////

private double[] calculateStateOfAirFromPad(double[] psychroVals, double efficy){
    double[] newPsychroVals = new double[13];
    double newAirTdb      = 0.0;
    double Wnew           = 0.0;
    double RHnew          = 0.0;
    double Tdb            = psychroVals[0];
    double Twb            = psychroVals[10];
    double PsatWb         = calculateSatVapPress(Twb);
    double Wsat           = calculateHumidityRatio(PsatWb,Patm);
    newAirTdb              = Tdb - (Tdb - Twb) * efficy;

    // Calculate new PsychroVals based on new Tdb and original Twb
    Wnew = ((2501.0 - 2.381 * (Twb - 273.15))*Wsat - (newAirTdb-273.15) + (Twb -
273.15))/(2501.0 + 1.805*(newAirTdb-273.15) - 4.186*(Twb-273.15));
    if (Wnew < 0.0){
        Wnew = 0.00000001;
    }

    RHnew = ((psychroVals[2] * Wnew)/calculateSatVapPress(newAirTdb))/(Wnew + 0.62198);
    newPsychroVals = calculatePsychroVals(newAirTdb, RHnew, Patm);

    return newPsychroVals;
} // end calculateStateOfAirFromPad

////////////////////////////////////////////////////////////////////////////////

private double[] calculateAdiabaticMixing(double Tdb1, double W1, double Tdb2, double W2,
double volume1, double volume2, double Patm)
    // function to determine the properties of a combination of two air volumes

    double RH1 = ((Patm * W1)/calculateSatVapPress(Tdb1))/(W1 + 0.62198);
    double RH2 = ((Patm * W2)/calculateSatVapPress(Tdb2))/(W2 + 0.62198);

    double[] psychroVals1 = calculatePsychroVals(Tdb1,RH1,Patm);
    double[] psychroVals2 = calculatePsychroVals(Tdb2,RH2,Patm);

    double hnew = (psychroVals1[7]*volume1/psychroVals1[6] +
psychroVals2[7]*volume2/psychroVals2[6])/(volume1/psychroVals1[6] + volume2/psychroVals2[6]);
    double Wnew = (psychroVals1[5]*volume1/psychroVals1[6] +
psychroVals2[5]*volume2/psychroVals2[6])/(volume1/psychroVals1[6] + volume2/psychroVals2[6]);

    double Tdbnew = (hnew - 2501*Wnew)/(1.006 + 1.805*Wnew) + 273.15;
    double RHnew = ((Patm * Wnew)/calculateSatVapPress(Tdbnew))/(Wnew + 0.62198);

    return calculatePsychroVals(Tdbnew, RHnew, Patm);
} // end calculateAdiabaticMixing
////////////////////////////////////////////////////////////////////////////////

```

```

private double calculateHumidityRatio(double P, double Patm){
    double W = 0.62198* (P/(Patm + P));
    return W;
} // end calculateHumidityRatio

////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////

private double calculateRelativeHumidity(double Tair, double Patm, double W){
    double RH = ((Patm * W)/(0.62198 + W)) / calculateSatVapPress(Tair);
    return RH;
} // end calculateRelativeHumidity

////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////

private double calculateEnthalpy(double temp, double W){
    double h = 1.006 * (temp - 273.15) + W * (2501 + 1.805 * (temp - 273.15));
    return h;
} // end calculateEnthalpy

////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////

private double calculateTwb(double Tdb, double Tdp, double Patm){
    double Twb = 0.0;
    double t = Tdb - 273.15; // drybulb C
    double dp = Tdp - 273.15; // dewpoint C
    double p = Patm*0.01; // Atmospheric Pressure mbar

    double tmin = Math.min(t,dp);
    double tmax = Math.max(t,dp);
    double tcur = 0.0;
    double e = 0.0;
    double tcvp = 0.0;
    double peq = 0.0;
    double diff = 0.0;

    e = calculateSatVapPress(dp+273.15)*0.01; // multiplied by 0.01 to convert to mbar
    tcur=(tmax+tmin)/2;
    tcvp=calculateSatVapPress(tcur+273.15)*0.01;
    peq=0.000660*(1+0.00155*tcur)*p*(t-tcur);
    diff=peq-tcvp+e;

    while (Math.abs(diff) > 0.01)
    {
        if (diff < 0) tmax=tcur;
        else tmin = tcur;

        tcur=(tmax+tmin)/2;
        tcvp=calculateSatVapPress(tcur+273.15)*0.01;
        peq=0.000660*(1+0.00155*tcur)*p*(t-tcur);
        diff=peq-tcvp+e;
    }
}

```



```
private double calculateDewPoint(double temp, double Pv){
    double dewPoint = 0.0;
    double p = 0.0; //

    p = Math.log(Pv);
    if (temp <= 273.15) {
        dewPoint = -60.45 + 7.0322*p + 0.37*Math.pow(p,2);
    }
    else {
        dewPoint = -35.957 - 1.8726*p + 1.1689*Math.pow(p,2);
    }
    return dewPoint+273.15;
} // end calculateDewPoint

////////////////////////////////////////////////////////////////////////////////////////////////////

private double[] calculatePsychroVals(double Tdb, double RH, double Patm){
    // Tdb      is drybulb temperature in Kelvin
    // RH       is the decimal relative humidity
    // Patm     is atmospheric pressure in Pa
    // Pvs     is the Saturation Vapour Pressure, Pa
    // Pv      is the Vapor Pressure, Pa
    // W       is the Humidity Ratio, kg H2O/ kg dry air
    // Vsa     is the air specific volume, m3/kg dry air
    // h       is the enthalpy of air-vapour mixture, J/kg
    // rho     is the moist air density, kg/m3
    // Tdp     is the dew point temperature K
    // delta   is the slope of the saturation vapor pressure line at temperature KPa/K
    // Twb     is the wet bulb temperature K
    // gamma   is the psychrometric constant kPa/K
    // hfg     is the heat of vaporization of water, kJ/kg

    double RA = 287.055; // Gas constant for air, J/kgK
    double[] psychroVals = new double[14]; //Array to hold psychrometric values for T and RH
    double Pvs = calculateSatVapPress(Tdb);
    double Pv = Pvs*RH;
    double W = calculateHumidityRatio(Pv,Patm);
    double Vsa = RA * Tdb * (1 + 1.6078 * W) / (Patm * (1 + W));
    double h = calculateEnthalpy(Tdb, W);
    double rho = ((Patm / 1000) * (1 + W)) / (0.28703 * Tdb * (1 + 1.16078 * W));
    double Tdp = calculateDewPoint(Tdb,Pv);
    double delta = Pvs/1000 * (17.269/(237.3+(Tdb-273.15)))*(1-(Tdb-273.15)/(237.3+Tdb-273.15));
    double Twb = calculateTWetBulb(Tdb, h, Patm, W);
    double gamma = 0.00066 * (1 + 0.00115*Twb) * Patm/1000;
    double hfg = 2501 - 2.42*(Tdb - 273.15);
    // Psychrometric constant (g) in kPa oC-1 for liquid water as a function of barometric pressure (P) in kPa and wet-bulb temperature (Tw) in oC was given by Fritschen and Gay (1979) as
    // Fritschen, L.J. & Gay, L.W. 1979. Environmental Instrumentation. New York, NY: Springer-Verlag.

    psychroVals[0] = Tdb; // Tdb      is the dry bulb temperature (K)
    psychroVals[1] = RH; // RH      is the relative humidity (decimal)
    psychroVals[2] = Patm; // Patm    is the Atmospheric Pressure (Pa)
```



```

psychroVals[3] = Pvs; // Pvs      is the Saturation Vapour Pressure, Pa
psychroVals[4] = Pv;  // Pv       is the Vapor Pressure, Pa
psychroVals[5] = W;   // W        is the Humidity Ratio, kg H2O/ kg dry air
psychroVals[6] = Vsa; // Vsa     is the air specific volume, m3/kg dry air
psychroVals[7] = h;   // h        is the enthalpy of air-vapour mixture, J/kg
psychroVals[8] = rho; // rho     is the moist air density, kg/m3
psychroVals[9] = Tdp; // Tdp    is the dew point temperature K
psychroVals[10] = Twb; // Twb   is the wet bulb temperature K
psychroVals[11] = delta;
psychroVals[12] = gamma;
psychroVals[13] = hfg;
// psychroVals[11] = lambda;

return psychroVals;
}

////////////////////////////////////////////////////////////////////////////////////////////////////

private double calculateSatVapPress(double temp){
//Psychrometric constants for calculating Saturation vapour pressure from Temperature
// 1993 ASHRAE Handbook: Fundamentals
double c1 = 0.0;
double c2 = 0.0;
double c3 = 0.0;
double c4 = 0.0;
double c5 = 0.0;
double c6 = 0.0;
double c7 = 0.0;

if ((temp-273.15) > 0) {
    c1 = -5.800221e+3;
    c2 = -5.516256e+0;
    c3 = -4.864024e-2;
    c4 = +4.176477e-5;
    c5 = -1.445209e-8;
    c6 = 0.0;
    c7 = +6.545967;
}
else {
    c1 = -5.674536e+3;
    c2 = -5.152306e-1;
    c3 = -9.677843e-3;
    c4 = +6.221570e-7;
    c5 = +2.074782e-9;
    c6 = -9.484024e-13;
    c7 = +4.163502;
}
double sum = c1/temp + c2 + c3*temp + c4*Math.pow(temp,2) + c5*Math.pow(temp,3) +
c6*Math.pow(temp,4) + c7*Math.log(temp);
double satVapPress = Math.exp(sum); // Saturation Vapour Pressure
return satVapPress*1000; // kludge fix calculates kPa returns Pa?
}

////////////////////////////////////////////////////////////////////////////////////////////////////

```

```

private double[] calculateEvapoTranspiration(double[] psychroVals, double[] enviroVals, double
tCanopy, double AREA){
    // Stanghellini 115, ISHS 1981 Acta hort p693

    double[] evapoTrans = new double[2]; // Array to hold evapotranspiration (mass and energy)
    double Tdb      = psychroVals[0]; // Dry bulb temperature K
    double Patm     = psychroVals[2]; // Atmospheric Pressure
    double Pvs      = psychroVals[3]; // Saturated Vapor pressure Pa
    double Pv       = psychroVals[4]; // Vapor pressure Pa
    double rho      = psychroVals[9]; // Air density kg/m3
    double delta    = psychroVals[11]; // Slope of saturation vapor pressure curve at temp of air
kPa/K
    double gamma    = psychroVals[12]; // Psychrometric constant kPa/K
    double hfg      = psychroVals[13]; // heat of vaporization of water kJ/kg
    double Rn       = enviroVals[0]; // Net all wave Radiation W/m2
    double G        = enviroVals[1]; // Radiation that penetrates to the ground W/m2
    double mu       = enviroVals[2]; // air speed at height of 2m, m/s
    double Eo       = 0.0; // Evapotranspiration mm/hr
    double ET       = 0.0; // Evapotranspiration J/m2 hr

    Eo = (0.408*delta*(Rn-G) + gamma*(37/Tdb)*mu*((Pvs-Pv)/1000)) / (delta +
gamma*(1+0.34*mu)); // Evapotranspiration in mm/hr

    ET = Eo * 1000; // J/m2 hr

    evapoTrans[0] = (3.0/1000) * AREA /1000;
    evapoTrans[1] = evapoTrans[0] * calculateHeatOfVaporization(tCanopy);

    return evapoTrans;
}

```

```

////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////

```

```

private double calculateHeatOfVaporization(double T){
    return 2501000 - 2.42 * (T - 273.15);
}

```

```

////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////

```

```

private double calculateVPD(double Patm, double Pvs, double Pv, double temp){
    // Calculat the vapor pressure deficit
    // equation source http://wahiduddin.net/calc/density\_altitude.htm, equation 4
    // temp = air dry bulb temperature, K
    // Patm, Atmospheric Pressure, Pa
    // Pvs, Saturated vapor pressure, Pa
    // Pv, Vapor pressure, Pa
    double VPD = 0.0; // Vapor pressure deficit, kg/m3
    double Pds = 0.0; // Pressure of dry air when saturatued with water Pa
    double Pd = 0.0; // Pressure of dry air when air not fully saturated Pa
    double RD = 287.055; // Ideal gas constant for dry air J/kgK
    double RV = 461.495; // Ideal gas constant for water vapor J/kgK
    double Dvsat = 0.0; // Saturated air density
    double Dv = 0.0; // Air density
}

```





```

} // end else
transmissivities[1] = transAdjust2;

return transmissivities;

} // end calculateTransmissivity
////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////

// Functions used for the spinach growth model
//*****
//*****

private double linearExtrap(String variableToExtrap, double havTemp){

    double extrapedValue = 0;

    if ( variableToExtrap == "DVRVT" ) {
        extrapedValue = havTemp * (0.027 / 24.0) / 30.0;
        // 0.027 from goudrian sucros87 divided by 24 to go from daily to hourly
    } // end if "DVRVT"

    if ( variableToExtrap == "AMTMP" ) {

        if ( havTemp <= 0 ) extrapedValue = 0;
        else if ( havTemp > 0 && havTemp <=10 ) extrapedValue = havTemp/10.0;
        else if ( havTemp > 10 && havTemp < 25) extrapedValue = 1.0;
        else if ( havTemp > 25 && havTemp <= 35) extrapedValue = (10.0 - (havTemp - 25.0))/10.0;
        else extrapedValue = 0;
    } // end if "AMTMP"

    if ( variableToExtrap == "FRT" ) {
        //extrapedValue = (10.0 + (16.0 - havTemp)/16.0 * 30.0)/100.0; //starting at 40% day 0 to 10%
day 16
        extrapedValue = 0.15;
    } // end if "FLV"

    if ( variableToExtrap == "EFF" ) {
        extrapedValue = 0.55 + 0.2 * (havTemp/20); // starts at 0.45 increases to 0.55 as LAI increases
LAI varies from 0.1 to 20
        //extrapedValue = 0.45;
    } // end if "FLV"

    return extrapedValue;
}

////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////

```

```

private double[] SUCROSE87(double HAVTMP, double hour, double[] cropParams, double[]
canopyRadProfileVals, double[]aboveCanopyRadiationVals){

    // Function to return the growth of the stage of crop:

    // Receive values from calling function for each stage of the crop growth and return various
components of growth..
    // Also receives environmental conditions for the hour
    // Also receives the level of Pythium damage and innoculum

    // Conditions recieved from the input
double NPL = cropParams[0];    // plant density (plants/m2)
double LA0 = cropParams[1];    // initial leaf area (ha/ha)
double WLW = cropParams[2];    // dry weight of leaves (kg/ha)
double WRT = cropParams[3];    // dry weight of roots (kg/ha)
double LAI = cropParams[4];    // Leaf area index (ha/ha)
double DVS = cropParams[5];    // age of crop in days (days)

double photoSynRateAdjDis = cropParams[12];

    // Leaf CO2 Assimilation
double AMX = cropParams[23]; // 40 kg CO2/ha/h
double AMAX = AMX * linearExtrap("AMTMP",HAVTMP);
cropParams[6] = AMAX;

    // Daily gross CO2 assimilation
// double EFF = linearExtrap("EFF",LAI); // 0.45;
// EFF = 0.45; //
// cropParams[7] = EFF;

double KDF = cropParams[8]; // 0.72;
double SCP = cropParams[9]; // 0.2;
//cropParams[8] = 0.83;

    // for function TOTASS
double HTGA = 0; // hourly total gross assimilation kg CO2/ha/h

    if (aboveCanopyRadiationVals[1] > 0)
        HTGA = Cal_hourly_assimilation( aboveCanopyRadiationVals, canopyRadProfileVals,
cropParams); // hourly total gross assimilation kg CO2/ha/h
    else
        HTGA = 0;

    // Carbohydrate Production
double GPHOT = HTGA * 30.0/44.0;

    // disease adjustment to carbohydrate production
GPHOT = GPHOT * photoSynRateAdjDis;

    // Maintenance
double MAINLV = cropParams[21]/24.0; // adjust rate from daily to hourly (kg/ha/d)
double MAINRT = cropParams[22]/24.0; // adjust rate from daily to hourly (kg/ha/d)

double MAINTS = MAINLV * WLW + MAINRT * WRT;

```









```

double convertToMols = 0.0036; // factor to convert light from HPS from umol/m2/s to mol/m2/h
double pcPAR = 0.55; // use a constant amount for the percentage of PAR regardless of cloudcover

// initialize PAR variables
double solarPAR = 0;
double HPSPAR = 0;
double PAR = 0; // Photosynthetically active radiation above crop canopy

// calculate the amount of PAR added by supplemental lights
if (lightsOn == 1) HPSPAR = supLightRate * convertToMols * 66.76; // 66.76 converts to J/m2/s
from mol/m2 hr
else HPSPAR = 0;

// calculation only works for daily, not hourly values
solarPAR = HTR/3600 * pcPAR * tau;
// J/m2/s (HTR hourly total radiation dividec by 3600 to get J/m2/s

if (shadesClosed == 1) solarPAR = solarPAR * shadeTrans;
// if the shades closed reduce the PAR by the shade factor

double FRDF = 0; // fraction of diffuse PAR

PAR = solarPAR;

double Io = solarRadiationParams[6];
double DOY = solarRadiationParams[8];
double LAT = solarRadiationParams[7] ;
double Sdec = solarRadiationParams[0] ;
double EOT = solarRadiationParams[11];

double solarTime = hour + (4*(75 - LAT) + EOT)*(1/60);

double solarHourAngle = Math.toRadians(15*(12-solarTime));

double SunsetHourAngle = Math.acos(-1*Math.tan(Math.toRadians(LAT))*Math.tan(Sdec));

double Hdioa = (24/Math.PI)*Io*(1 +
Math.cos(2*Math.PI*DOY/365))*Math.cos(Math.toRadians(LAT))*Math.cos(Sdec)*Math.cos(Sunset
HourAngle);

double LATradians = Math.toRadians(LAT);

// add in the 0.5 to calculate sinb between hours
double SINB = Math.max(0.000001,SINLD + COSLD * Math.cos(2.0 * Math.PI * (hour + 12.0 +
0.5) / 24.0 ));

double sunrise = solarRadiationParams[9];
double sunset = solarRadiationParams[10];

double Hhioa = 0;
double Kt = 0;
double fdif = 0;

double hourScale = 0;

```

```

if ((hour-1 >= Math.floor(sunrise)) && (hour < (Math.floor(sunset) + 1))) {

    hourScale = (Math.cos((hour-sunrise)/(sunset-sunrise)*Math.PI) - Math.cos((hour+1-
sunrise)/(sunset-sunrise)*Math.PI))/2;

    Hhioa = Hdioa * hourScale;

    Kt = HTR / Hhioa;

    fdif = 1.39 - 4.03*Kt + 5.53*Math.pow(Kt,2) - 3.11*Math.pow(Kt,3);

    if ( fdif > 1 ) fdif = 1;

    if ( fdif < 0 ) fdif = 0;
    fdif = 0.5;

} // end if
else {
    Hhioa = 0;
    fdif = 0;
} // end else

FRDF = fdif;

double PARDF = PAR * FRDF; // amount of diffuse PAR
double PARDR = PAR - PARDF; // amount of direct PAR

// check to ensure that the calculation of FRDF doesn't screw up the calculation
if (PAR <= 0){
    PARDF = 0;
    PARDR = 0;
} // end if

// add in the PAR from the supplemental lights (all assumed to be diffuse
PARDF = PARDF + HPSPAR;

aboveCanopyRadiationVals[0] = SINB;
aboveCanopyRadiationVals[1] = PARDF;
aboveCanopyRadiationVals[2] = PARDR;
aboveCanopyRadiationVals[3] = PAR;

return aboveCanopyRadiationVals;
} // end Cal_above_canopy_radiation

////////////////////////////////////////////////////////////////////////////////////////////////////////////////

private double[] Cal_canopy_radiation_profile(double[] aboveCanopyRadiationVals, double[]
cropParams){

    double[] canopyRadProfileVals = new double[4];

    double SCP = cropParams[9];
    double KDF = cropParams[8];
    double LAIC = cropParams[10];

```



```

double PARDR = aboveCanopyRadiationVals[2];

double VISDF = 0;
double VIST = 0;
double VISD = 0;
double VISSHD = 0;
double FGRSH = 0;
double VISPP = 0;
double FGRSUN = 0;
double VISSUN = 0;
double FGRS = 0;
double FSLLA = 0;
double FGL = 0;

// constants needed for the five point Gaussian integration
double[] GSDST = new double[5];
GSDST[0] = 0.0469101;
GSDST[1] = 0.2307653;
GSDST[2] = 0.5;
GSDST[3] = 0.7692347;
GSDST[4] = 0.9530899;
double[] GSWT = new double[5];
GSWT[0] = 0.1184634;
GSWT[1] = 0.2393143;
GSWT[2] = 0.2844444;
GSWT[3] = 0.2393143;
GSWT[4] = 0.1184634;

double SQV = Math.sqrt(1.0 - SCP);
double REFH = (1.0 - Math.sqrt(1.0 - SQV))/(1.0 + Math.sqrt(1.0 + SQV));
double REFS = REFH * 2.0 / (1.0 + 2.0 * SINB);

double CLUSTF = KDF / (0.8 * SQV);
double KBL = (0.5 / SINB) * CLUSTF;
double KDRT = KBL * SQV;

double FGROS = 0.0;
for (int k = 0; k <5; k++){
    LAIC = LAI * GSDST[k];

    VISDF = (1.0 - REFH) * PARDF * KDF * Math.exp(-KDF * LAIC);
    VIST = (1.0 - REFS) * PARDR * KDRT * Math.exp(-KDRT * LAIC);
    VISD = (1.0 - SCP) * PARDR * KBL * Math.exp(-KBL * LAIC);

    VISSHD = VISDF + VIST - VISD;
    if (AMAX > 0) FGRSH = AMAX * (1.0 - Math.exp(-VISSHD * EFF / AMAX));
    else FGRSH = 0;

    VISPP = (1.0 - SCP) * PARDR / SINB;
    FGRSUN = 0;
    for (int m = 0; m <5; m++){

        VISSUN = VISSHD + VISPP * GSDST[m];
        if (AMAX > 0) FGRS = AMAX * (1.0 - Math.exp(-VISSUN * EFF / AMAX));
        else FGRS = 0;
    }
}

```



```

FGROS = 0;

for (int j = 0; j <5; j++){

    // Selection of depth into canopy
    LAIC = LAI * GSDST[j];

    cropParams[10] = LAIC;
    // Call procedure Cal_canopy_radiation_profile
    canopyRadProfileVals = Cal_canopy_radiation_profile(aboveCanopyRadiationVals,
cropParams);

    PARLSH = canopyRadProfileVals[0];
    PARLSL = canopyRadProfileVals[1];
    PARLPP = canopyRadProfileVals[2];
    FSLLA = canopyRadProfileVals[3];

    ASSSH = AMAX * (1 - Math.exp(-EFF * PARLSH/AMAX));
    ASSSL = 0;

    for (int k = 1; k <5; k++){

        PARLSL = PARLSH + PARLPP * GSDST[k];
        ASSSL = ASSSL + AMAX * (1 - Math.exp(-PARLSL * EFF/AMAX))*GSWT[k];

    } // end k

    FGROS = FGROS + ((1 - FSLLA) * ASSSH + FSLLA * ASSSL) * LAI * GSWT[j];

} // end j

HTGA = FGROS;

return HTGA;
} // end Cal_daily_assimilation
////////////////////////////////////////////////////////////////////////////////

```

```

// Functions used for the spinach disease and spinach production model
//*****
//*****

```

```

private void generateRandomNumbersFile(){

    PrintWriter outputStream = null;
    double[] us = new double[2];
    double[] ns = new double[2];

    if (false){

        try {
            outputStream = new PrintWriter(new FileOutputStream("g:\\randomCropValues3.txt"));
        } // end try
    }
}

```

```

catch(FileNotFoundException e) {
    System.out.println("error in opening file out.txt");
    System.exit(0);
} // end catch

// for loop for generating random values for the growth model
for (int z = 0; z < 1000000; z++){

    // random value for A
    outputStream.println(getTriangular(Math.random(), 3.81E-3, 5.72E-3, 4.76E-3));
    // random value for AMAX
    outputStream.println(getTriangular(Math.random(), 36, 44, 40));
    // random value for EFF
    outputStream.println(getTriangular(Math.random(), 0.4, 0.5, 0.45));
    // random value for KDF
    outputStream.println(getTriangular(Math.random(), 0.29, 0.72, 0.5));
    // random value for MAINLF
    outputStream.println(getTriangular(Math.random(), 0.018, 0.042, 0.03));
    // random value for MAINRT
    outputStream.println(getTriangular(Math.random(), 0.009, 0.021, 0.015));
    // random value for SCP
    outputStream.println(getTriangular(Math.random(), 0.15, 0.25, 0.2));

    if (z%1000 == 0) System.out.println(z);

} // end for
} // end if
else {

    try {
        outputStream = new PrintWriter(new FileOutputStream("g:\randomPythiumValues2.txt"));
    } // end try
    catch(FileNotFoundException e) {
        System.out.println("error in opening file out.txt");
        System.exit(0);
    } // end catch

    for (int t = 0; t < 700000; t++){

        us[0] = Math.random();
        us[1] = Math.random();
        ns = boxMuller(us);
        // random value for CADJA
        outputStream.println(getlogNormal(ns[0], 0.180, 0.018));
        // random value for CADJB
        outputStream.println(getlogNormal(ns[1], 0.761, 0.114));
        // random value for IFCTPCT
        outputStream.println(getTriangular(Math.random(), 0.71, 6.75, 3.23));
        us[0] = Math.random();
        us[1] = Math.random();
        ns = boxMuller(us);
        // random value for INITMYC
        outputStream.println(getlogNormal(ns[0], 1.95E-3, 3.9E-4));
        // random value for MGRA
        outputStream.println(getlogNormal(ns[1], 0.000195, 3.9E-5));
    }
}

```





```

private double getlogNormal(double n, double ux, double sx){

    double logN = 0;

    double sl = Math.sqrt(Math.log(1 + Math.pow(sx,2)/Math.pow(ux,2) ));
    double ul = Math.log(ux) - 0.5*Math.pow(sl,2);

    logN = Math.exp(ul + sl*n);

    return logN;
} // end getlogNormal

////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////

private double getTriangular(double u, double a, double b, double c){

    double r = 0;

    if (u <= ((c-a)/(b-a))){
        r = a + Math.sqrt(u*(b-a)*(c-a));
    } // end if
    else {
        r = b - Math.sqrt((1-u)*(b-a)*(b-c));
    } // end else

    return r;
} // end getTriangular

////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////////

private void growthModel(){
    double[][] enviroData      = new double[113880][5];
    double[] cohortValues      = new double[1000000];
    double[] diseaseValues     = new double[100000];

    enviroData      = readDataFile("g:\greenhouseOutNaturalTnosuplight12years.txt", 2);
    cohortValues    = readDataFile3("g:\randomCropValues3.txt",1000000);
    diseaseValues   = readDataFile3("g:\randomPythiumValues3.txt",3600000);

    int nPoints = enviroData.length;

    double[] cropParams        = new double[25];
    double[] solarRadiationParams  = new double[12];
    double[] lightingVals       = new double[6];
    double[] aboveCanopyRadiationVals = new double[4];
    double[] canopyRadProfileVals  = new double[7];
    double[][] mycelialAgesMass    = new double[1][3];
    double[] diseaseParams        = new double[21];
    double[] pondVals             = new double[3]; // array of pond specific information

    Vector pondVector              = new Vector();
    // Vector that contains the disease params, crop Params, mycelial information for the various cohorts
    Vector harvests                = new Vector();
    // Vector that contains harvest information (size of crop, quality, etc)

```

```

Vector culls          = new Vector();
// Vector that contains culling information (size of crop, etc.)

int day      = 0;
int time     = 0;

double LAT    = 42.3;
double transMVTY = 0.7;
double shadeFactor = 0.5;
double supPPF = 400; // umol/m2/s (set artificially high to ensure that the target mols of 19 can
be reached in winter.

// Initial crop conditions
double cropDensity      = 1200; // plant density (plants/m2)
double leafAreaInitial  = 0.1; // initial leaf area (ha leaf/ha ground)
double leafDryWt        = 33.0; // initial dry weight of leaves (kg/ha)
double rootDryWt        = 5.0; // initial dry weight of roots (kg/ha)
double LAI              = 0.1; // initial Leaf area index
double cropAge          = 0; // age of crop in hours
double AMAX             = 45; // AMAX = 40 // temperature adjusted assimilation rate
double EFF              = 0.50; // EFF = 0.45;
double KDF              = 0.50; // KDF = 0.72;
double SCP              = 0.2; // SCP = 0.2;
double LAIC             = 1; // LAIC
double shootQuality     = 1; // ranking of quality
double photoSynRateAdjDis = 1; // adjustment to the growth of the crop due to root
damage
double harvestAge      = 336; // age at which crop cohort is harvested
double cropArea        = 1; // Area of crop cohort (ha)
double pondSwitchAge   = 168; // age at which the crop is switched to a different pond (can
be modified mid cycle to reflect more than 1 pond)
double pondNumber      = 0; // which pond number the cohort is located in (starts with pond
0)
double timeOfHarvest   = 0; // simulation time at which the crop was harvested (hr)
double floatTime       = 336; // hour at which the crop is floated in the pond (doesn't
necessarily correspond to harvest
double cohortID        = 0; // the unique identification number for each cohort
(incremented by one for each cohort added to ponds)
double A               = 0.00496; // the slope of the line relating leaf biomass to LAI
double MAINLV          = 0.03; // daily maintenance rate of leaves (kg/ha/d)
double MAINRT          = 0.015; // daily maintenance rate of roots (kg/ha/d)
double AMX             = 45; // Maximum assimilation rate

// Put values in cropParams array
cropParams[0] = cropDensity;
cropParams[1] = leafAreaInitial;
cropParams[2] = leafDryWt;
cropParams[3] = rootDryWt;
cropParams[4] = LAI;
cropParams[5] = cropAge;
cropParams[6] = AMAX;
cropParams[7] = EFF;
cropParams[8] = KDF;
cropParams[9] = SCP;
cropParams[10] = LAIC;

```

```

cropParams[11] = shootQuality;
cropParams[12] = photoSynRateAdjDis;
cropParams[13] = harvestAge;
cropParams[14] = cropArea;
cropParams[15] = pondSwitchAge;
cropParams[16] = pondNumber;
cropParams[17] = timeOfHarvest;
cropParams[18] = floatTime;
cropParams[19] = cohortID;
cropParams[20] = A;
cropParams[21] = MAINLV;
cropParams[22] = MAINRT;
cropParams[23] = AMX;

// initialize disease Parameters array
// Variables within disease params
double mycelialMass      = 0.0; // mycelial mass kg/ha
double mycelialGrowth    = 0.0; // Amount of mass of mycelia in first hour as a function of
zoospore conc
double equivAgeMycelia   = 1.0; // equivalent age of the mycelia (function of temperature)
(hr)
double zoosReleased      = 0.0; // zoos released (absolute number) (7.5E10 = 25 z/ml 1 ha
0.3 m deep)
double zoosConc          = 0.0; // zoospore concentration in pond (zoos/ml)
double massRootDBBL     = 0.0; // percentage of the root that is dark brown/black
// constants within disease params
double TADJA             = 0.07; // multiplier of temperature adjustment equation
double TADJB             = -6.12E-01; // exponent of temperature adjustment equation
double CADJA             = 0.180; // slope of concentration adjustment equation
double CADJB             = 0.761; // intercept of concentration adjustment equation
double MGRA              = 0.000195; // slope of mycelial growth rate adjustment
equation
double MGRB              = 0.010; // intercept of mycelial growth rate adjustment
equation
double IFCTPCT           = 3.23; // reference amount of mycelial infection at 25
zoos/ml kg/ha basis
double myceliaMatureAge  = 128; // Age at which mycelium matures to dark brown/black
(hours)
double PRADJA            = 0.79; // photosynthetic rate adjustment due to root
damage
mycelialMass            = 1.5e-3; // starting mass of mycelium
meanZooReleaseAge       = 360;
zoosPerKgRoot           = 8.6e7;
meanZooReleaseAge       = 128;
zoosPerKgRoot           = 1.1e6;
cropParams[15]          = 192; //pondSwitchAge;
cropParams[13]          = 384; //harvestAge;
cropParams[18]          = 384;

int infctime             = -216; // time at which to apply the zoospores to the pond
double initInfectLevel   = 25; // concentration of inoculation
double TA = 0;
int tempadjusttime = -216;

```

```

double newTempadjust = -6;

// Put values in diseaseParams array
diseaseParams[0] = mycelialMass;
diseaseParams[1] = mycelialGrowth;
diseaseParams[2] = equivAgeMycelia;
diseaseParams[3] = zoosReleased;
diseaseParams[4] = zoosConc;
diseaseParams[5] = massRootDBBL;
diseaseParams[6] = TADJA;
diseaseParams[7] = TADJB;
diseaseParams[8] = CADJA;
diseaseParams[9] = CADJB;
diseaseParams[10] = MGRA;
diseaseParams[11] = MGRB;
diseaseParams[12] = IFCTPCT;
diseaseParams[13] = myceliaMatureAge;
diseaseParams[14] = PRADJA;
diseaseParams[15] = zoosPerKgRoot;
diseaseParams[16] = meanZooReleaseAge;
diseaseParams[17] = 0;
diseaseParams[18] = 0;
diseaseParams[19] = 0;
diseaseParams[20] = 0;           // disease cohort ID

// initialize pondVals array
double areaPerCohort = 10000;
double noCohortsInPond = 1;
double PondArea = areaPerCohort * noCohortsInPond; // area of pond in square meters
double VolPondInMIPerHa = PondArea * ZWATER * 1000 * 1000;

pondVals[0] = tWaterTm1 - 273.15;
pondVals[1] = VolPondInMIPerHa;
pondVals[2] = 0; // relative age of the nutrient solution in the pond. (temperature adjusted)

double[] newDiseaseParams = diseaseParams.clone();
double[] newCropParams = cropParams.clone();
double[][] newMycelialAgesMass = mycelialAgesMass.clone();

double lightsOn;
double shadesClosed;
double airTemp;
double solutionTemp;
double humidity;
double HTR;

// information on the cohorts to populate the pond (for more than one cohort
// per pond. First index is the age of the cohort, second index contains
// the area that it occupies (ha)
double[] cohorts = new double[10];

cohorts[0] = 72; // age of first cohort in hours
cohorts[1] = 144;
cohorts[2] = 216;
cohorts[3] = 288;

```

```

int numberOfPonds = 1; // Variable to hold the number of ponds

// array to hold the number of zoospores released in the current hour
double[] newzoosReleased = new double[numberOfPonds];

// array to hold the concentration of zoospores released in the previous hour
double[] pondZoosConc = new double[numberOfPonds];
double[] oldPondZoosConc = new double[numberOfPonds];

// output stream for writing harvest data
PrintWriter harvestOutputStream = null;

try {
    harvestOutputStream = new PrintWriter(new FileOutputStream("g:\\harvests.txt"));
} // end try
catch(FileNotFoundException e) {
    System.out.println("error in opening file out.txt");
    System.exit(0);
} // end catch

// output stream for writing harvest data
PrintWriter cullOutputStream = null;

try {
    cullOutputStream = new PrintWriter(new FileOutputStream("g:\\culls.txt"));
} // end try
catch(FileNotFoundException e) {
    System.out.println("error in opening file out.txt");
    System.exit(0);
} // end catch

NumberFormat formatter = new DecimalFormat();
formatter = new DecimalFormat("0.###E0");

boolean randomizeValues = true;
boolean cropCull = false;

// Disease and pond simulation for loop
for (int i = 0; i < 365*24*500; i++){

    if (i%8760 == 0 ){
        System.out.println(i/8760);
    } // end if

    // establish the hour of the day and the day of the year based on the
    // iteration of the loop
    // if/else to maintain 0-24 hour loop
    if( time>=0 && time<=22) {
        time++; // increment an hour for each loop
    } // end if
    else {
        time = 0; // resets hour back to 0 (midnight) if previously 11 pm
    }
}

```

```

    day++; // increments day to reflect 24 hour period
} // end else

// reset the day of the year
if (day > 365 ){
    day = 0;
}

if (randomizeValues){
    // assign new baseline yearly Pythium values
    if ( ((day == 1)&&(time == 0)) || (cropCull == true) ){

        // reset the cropCull variable
        cropCull = false;

        // increment the disease cohhort id number
        newDiseaseParams[20] = newDiseaseParams[20] + 1;

        // randomized values of crop Parameters

        newDiseaseParams[8] = diseaseValues[(int)newDiseaseParams[20] * 12];
        newDiseaseParams[9] = diseaseValues[(int)newDiseaseParams[20] * 12 + 1];
        newDiseaseParams[12] = diseaseValues[(int)newDiseaseParams[20] * 12 + 2];
        newDiseaseParams[0] = diseaseValues[(int)newDiseaseParams[20] * 12 + 3];
        newDiseaseParams[10] = diseaseValues[(int)newDiseaseParams[20] * 12 + 4];
        newDiseaseParams[11] = diseaseValues[(int)newDiseaseParams[20] * 12 + 5];
        newDiseaseParams[13] = diseaseValues[(int)newDiseaseParams[20] * 12 + 6];
        newDiseaseParams[14] = diseaseValues[(int)newDiseaseParams[20] * 12 + 7];
        newDiseaseParams[6] = diseaseValues[(int)newDiseaseParams[20] * 12 + 8];
        newDiseaseParams[7] = diseaseValues[(int)newDiseaseParams[20] * 12 + 9];
        newDiseaseParams[16] = diseaseValues[(int)newDiseaseParams[20] * 12 + 10];
        newDiseaseParams[15] = diseaseValues[(int)newDiseaseParams[20] * 12 + 11];

    } // end if
} // end if

if (i == tempadjusttime) TA = newTempadjust;

// retrieve the environmental parameters for the current hour
airTemp      = enviroData[i%113880][0];
solutionTemp = enviroData[i%113880][1] + TA;
lightsOn     = enviroData[i%113880][2];
shadesClosed = enviroData[i%113880][3];
HTR          = enviroData[i%113880][4];

// spinach growth model portion

// calculate the radiation parameters for the day
solarRadiationParams = Cal_daily_solar_radiation_parameters(day, LAT);

// adjust the lightingVals array to reflect the current values for the hour
lightingVals[0] = HTR; // current solar light level (J/m2/hr)
lightingVals[1] = transMVTY; // greenhouse transmittance
lightingVals[2] = shadesClosed; // whether shades closed (1/0)

```

```

lightingVals[3] = shadeFactor;    // shade transmittance
lightingVals[4] = lightsOn;      // whether supplemental lights on (1/0)
lightingVals[5] = supPPF;        // PPF from supplemental light (umol/m2/s)

// calculate the amount and quality of the above canopy radiation for the hour
aboveCanopyRadiationVals = Cal_above_canopy_radiation (lightingVals, time,
solarRadiationParams);

// update the pond temperature in the pondVals array
pondVals[0] = solutionTemp;

// loop for strictly harvesting/culling
for(int m = 0; m < pondVector.size(); m = m + 3){
    // retrieve the necessary data arrays for this particular cohort
    diseaseParams    = ((double[])pondVector.elementAt(m));
    cropParams       = ((double[])pondVector.elementAt(m+1));
    mycelialAgesMass = ((double[][])pondVector.elementAt(m+2));

    // determine whether the cohort should be harvested
    if (cropParams[5] == cropParams[13]){
//        if (cropParams[2] >= 1221){
//        if (cropParams[2] >= 1583){
//        if (cropParams[2] >= 1958){

        // set the time the crop was harvested
        cropParams[17] = (double)i;

        // add the values in the cohorts cropParams to the harvests

        harvestOutputStream.println(
            formatter.format(cropParams[2])  + "\t" +
            formatter.format(cropParams[3])  + "\t" +
            formatter.format(cropParams[5])  + "\t" +
            formatter.format(cropParams[19]) + "\t" +
            formatter.format(diseaseParams[0]) + "\t" +
            formatter.format(diseaseParams[5]) + "\t" +
            formatter.format(diseaseParams[20]) + "\t" + i);

        // remove the cohorts from the pond vector
        pondVector.remove(m);
        pondVector.remove(m);
        pondVector.remove(m);

    } // end if for harvesting

    // determine if the cohort should be moved
    if ((cropParams[5] == cropParams[15]) && (numberOfPonds > 1)){
        cropParams[16] = cropParams[16] + 1; // can only move one pond at a time for now
    } // end if

    // determine if the cohort should be culled and ponds cleaned out
    if ( diseaseParams[0]/cropParams[3] > 0.10 ){

        cullOutputStream.println(

```



```

        formatter.format(cropParams[2]) + "\t" +
        formatter.format(cropParams[3]) + "\t" +
        formatter.format(cropParams[5]) + "\t" +
        formatter.format(cropParams[19]) + "\t" +
        formatter.format(diseaseParams[0]) + "\t" +
        formatter.format(diseaseParams[5]) + "\t" +
        formatter.format(diseaseParams[20]) + "\t" + i);

// remove all of the cohorts in the ponds to start over.
while(pondVector.size() > 0){

    //culls.add(pondVector.elementAt(0));
    pondVector.remove(0);

} // end while

// reset the counter for the number of zoos released per pond
for (int q = 0; q < numberOfPonds; q++){
    oldPondZoosConc[q] = 0;
    newzoosReleased[q] = 0;
} // end for q

// call for a resetting of the pythium random variables
cropCull = true;

} // end culling if

} // loop through the cohorts in the ponds to harvest

// determine if time to float a new cohort
// check out the float time from the oldest cohort

if (( ( i % 72) == 0 ) && (pondVector.size() < 15 )) {
    // need a regen time for recovering after a cull. Add every three days as long as
    //the pondvector size is below a certain size (depends on the crop length.)
    // pond vector is 15 for 5 cohorts, 12 for 4

    // retrieve the original crop parameter values
    cropParams = newCropParams.clone();

    // increment the crop cohhort id number
    cropParams[19] = cropParams[19] + 1;
    // reset the crop id number of the template
    newCropParams[19] = cropParams[19];

    if (randomizeValues){
        // randomized values of crop Parameters
        cropParams[20] = cohortValues[(int)cropParams[19] * 7];
        cropParams[6] = cohortValues[(int)cropParams[19] * 7 + 1];
        cropParams[7] = cohortValues[(int)cropParams[19] * 7 + 2];
        cropParams[8] = cohortValues[(int)cropParams[19] * 7 + 3];
        cropParams[21] = cohortValues[(int)cropParams[19] * 7 + 4];
        cropParams[22] = cohortValues[(int)cropParams[19] * 7 + 5];
        cropParams[9] = cohortValues[(int)cropParams[19] * 7 + 6];
    }
}

```

```

} // end if

pondVector.add(newDiseaseParams.clone());
//pondVector.add(newCropParams.clone());
pondVector.add(cropParams);

pondVector.add(newMycelialAgesMass.clone());
} // end if for adding a cohort

// reset the counter for the number of zoos released per pond
for (int q = 0; q < numberOfPonds; q++){
    oldPondZoosConc[q] = pondZoosConc[q];
    newzoosReleased[q] = 0;
} // end for q

// for loop for the various cohorts of the crop
for (int t = 0; t < pondVector.size(); t = t + 3){

    // retrieve the necessary data arrays for the particular cohort
    diseaseParams = (double[])pondVector.elementAt(t);
    cropParams = (double[])pondVector.elementAt(t+1);
    mycelialAgesMass = (double[][])pondVector.elementAt(t+2);

    // insert the corresponding zoospore Concentration from the previous hours
    // released zoospores (cropParams[16] holds the pond number
    diseaseParams[4] = pondZoosConc[(int)cropParams[16]];

    // Calculate the parameters affecting the rate of sporangia growth
    diseaseParams = Cal_disease_Parameters( diseaseParams, cropParams, mycelialAgesMass,
pondVals );

    // increment the amount of zoospores released from each cohort
    newzoosReleased[(int)cropParams[16]] = newzoosReleased[(int)cropParams[16]] +
diseaseParams[3];

    // calculate the growth of the sporangia during this current hour
    mycelialAgesMass = Cal_mycelial_growth(diseaseParams, mycelialAgesMass);

    // calculate the mass of mycelium that has matured to dark brown/black
    diseaseParams = Cal_percent_root_DBBL(diseaseParams, mycelialAgesMass);

    // Modify crop Parameters to take into account disease
    cropParams = Adj_crop_Parameters ( cropParams, diseaseParams);

    // calculate growth of crop cohort
    cropParams = SUCROSE87(airTemp, time, cropParams, canopyRadProfileVals,
aboveCanopyRadiationVals);
    cropParams[5] = cropParams[5] + 1.0; // increment cohort age by 1 hour

    // store cohort array data back in the overall crop Vector
    pondVector.set(t,diseaseParams.clone());
    pondVector.set(t+1,cropParams.clone());
    pondVector.set(t+2,mycelialAgesMass.clone());

} // end for t loop through cohorts within specific pond

```





```

try{
    freader    = new FileReader(fileName);
    input      = new StreamTokenizer(freader);
    int hour   = 0;
    input.nextToken();
    input.nextToken();
    input.nextToken();
    input.nextToken();
    input.nextToken();

    while( input.ttype != StreamTokenizer.TT_EOF){
        if( input.ttype == StreamTokenizer.TT_NUMBER){
            data1[hour][0] = ((input.nval) - 32.0) * (5.0/9.0) + 273.15;    // F to K
        }
        input.nextToken();
        if( input.ttype == StreamTokenizer.TT_NUMBER){
            data1[hour][1] = (input.nval)/100.0;    // decimal
        }
        input.nextToken();
        if( input.ttype == StreamTokenizer.TT_NUMBER){
            data1[hour][2] = (input.nval)*1055.05585/0.09290304 ; // BTU/ft2/hr to J/m2/hr
        }
        input.nextToken();
        input.nextToken();
        input.nextToken();
        input.nextToken();
        input.nextToken();
        hour++;
    } // end while
    freader.close();
} // end try
catch( FileNotFoundException ex){
    System.out.println("file not found");
}
catch( IOException ex ){
    System.out.println(ex.getMessage());
}
finally {
    try{
        if (freader != null) freader.close();
    }
    catch(IOException ex){
        System.out.println(ex.getMessage());
        System.exit(0);
    } // end try catch

} // end finally
} // end sourceType 1 if

if (sourceType == 2) {

    FileReader freader    = null;
    StreamTokenizer input  = null;

```



```

// adjust photosynthesis rate
// growth adjusted by reducing the amount of photosynthate available for distribution
// return cropParams

// calculate percentage reduction in photosynthate production
double PRADJA      = diseaseParams[14];
double massRootDBBL  = diseaseParams[5];
double totalRootMass = cropParams[3];
double fractRootInf  = diseaseParams[0]/cropParams[3];

// bound the fraction of root that is dark brown black
if (fractRootInf > 0.95 ) fractRootInf = 0.95;
if (fractRootInf < 0   ) fractRootInf = 0;

double photoSynRateAdjDis = 1 - PRADJA * fractRootInf;

// bound the photosynthetic rate adjustment
if (photoSynRateAdjDis < 0.1) photoSynRateAdjDis = 0.1;
if (photoSynRateAdjDis > 1) photoSynRateAdjDis = 1;

cropParams[12] = photoSynRateAdjDis;

return cropParams;
} // end Adj_crop_Parameters

//////////////////////////////////////////////////////////////////////

private double[] Cal_disease_Parameters(double[] diseaseParams, double[] cropParams,
double[][]mycelialAgesMass, double[] pondVals ){
// this function is run each hour for each cohort of spinach present in the pond system.
// returns the diseaseParams array
// Calculate the parameters affecting the rate of sporangia growth
// gets diseaseParams, cropParams, solution temperature
// determine the sporangial growthrate adjusted for temperature
// determine the equivalent age (function of temperature)
// determine the initial infection mass of roots (function of zoos conc)
// determine total mass in current hour
// return diseaseParams

double solutionTemp = pondVals[0]; // nutrient solution temperature

double totalRootMass = cropParams[3];

// variables within disease params
double mycelialMass      = diseaseParams[0];
double mycelialGrowth    = diseaseParams[1];
double equivAgeMycelia  = diseaseParams[2];
double zoosReleased     = diseaseParams[3];
double zoosConc         = diseaseParams[4];
double massRootDBBL     = diseaseParams[5];
// constants within disease params
double TADJA            = diseaseParams[6];
double TADJB           = diseaseParams[7];
double CADJA           = diseaseParams[8];
double CADJB           = diseaseParams[9];

```

```

double MGRA          = diseaseParams[10];
double MGRB          = diseaseParams[11];
double IFCTPCT       = diseaseParams[12];
double zoosPerKgRoot = diseaseParams[15];
double meanZooReleaseAge = diseaseParams[16];
double stdevZooReleaseAge = diseaseParams[17];
double rootDryWt     = cropParams[3];

// calculate the Temperature effects on growth and aging of the mycelium
double tempAdjust = TADJA * Math.exp(TADJB * solutionTemp); // adjustment to sporangial
growthrate as a function of temperature

//linear adjustment
tempAdjust = TADJA * solutionTemp + TADJB;

equivAgeMycelia = tempAdjust; // Set the Mycelial age adjustment (same as temperature growth
rate adjustment)

// calculate the mass of new mycelial material this hour (growth of existing plus new infection)
// mass of roots available to infect is the difference of the total root mass and the total mass of
mycelia infected root
// kg / ha

double healthyRootMass = totalRootMass - mycelialMass; // mass of healthy root available
for infection

// the Mycelial growth rate is a function of the percentage of roots that are healthy
// kg sporangia per kg infected root per hour

double mycelialGrowthRate = (MGRA * healthyRootMass + MGRB) * tempAdjust;
if (mycelialGrowthRate > (0.021 * tempAdjust) ) mycelialGrowthRate = 0.021 * tempAdjust;
if (mycelialGrowthRate < 0 ) mycelialGrowthRate = 0;

if (mycelialGrowthRate > 0.021 ) mycelialGrowthRate = 0.021;
if (mycelialGrowthRate < 0 ) mycelialGrowthRate = 0;

// calculate the adjustment to the concentration of zoospores
double logconc = 0;
double concAdjust = 0;

if (zoosConc > 0) {
    logconc = Math.log10(zoosConc);
    concAdjust = CADJA * logconc + CADJB; // adjustment to the infection mass as a function of
initial concentration
    if (concAdjust < 0) concAdjust = 0;
} // end if
else {
    concAdjust = 0;
} // end else

// calculate the amount of mycelial mass due to the concentration of zoospores in solution
// infectionAMT is a function of the amount of healthy root present

```







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