Modelling and Remote Sensing of Canopy Light Interception and Plant Stress in Greenhouses

Vaia Sarlikioti

Thesis committee

Thesis supervisors

Prof. dr. ir. L.F.M Marcelis
Professor of Crop Production in Low-energy Greenhouses, Wageningen Univeristy

Prof. dr. O. Van Kooten
Professor of Horticultural Supply Chains, Wageningen Univeristy

Thesis co-supervisor

Dr. P.H.B. de Visser
Researcher, Wageningen UR Greenhouse Horticulture

Other members

Prof. dr. ir. E.J. van Henten, Wageningen University
Dr. ir. J. Vos, Wageningen University
Prof. dr. C. Kittas, University of Thessaly, Greece
Prof. dr. H. Stützel, Leibniz University Hannover, Germany

This research was conducted under the auspices of the C.T. de Wit Graduate School for Production Ecology and Resource Conservation.

Modelling and Remote Sensing of Canopy Light Interception and Plant Stress in Greenhouses

Vaia Sarlikioti

Thesis
submitted in fulfilment of the requirements for the degree of doctor
at Wageningen University
by the authority of the Rector Magnificus
Prof. dr. M.J. Kropff,
in the presence of the
Thesis Committee appointed by the Academic Board
to be defended in public
on Monday, 10 October 2011
at 4.00 p.m. in the Aula.

Vaia Sarlikioti
Modelling and Remote Sensing of Canopy Light Interception and Plant Stress in Greenhouses

Thesis, Wageningen University, Wageningen, NL (2011)
With references, with summaries in English, Dutch and Greek

ISBN 978-94-6173-005-3

Table of contents

Chapter 1

General Introduction

Greenhouse horticulture: development towards full control

A greenhouse crop can be approached as an open system that can be affected by a number of parameters such as light, climate or nutrient supply. The last decades efforts have been made to understand the function of this system and the interaction of the different parameters. The intensive nature of greenhouse cultivation as such gives an impetus for the development of decision support systems (DSS) to help the growers in managing their farm efficiently. The foundations of DSS are plant models. (Marcelis *et al.,* 2000). Although many models have been proposed during the past decades, their lack in accuracy makes it useless to integrate them yet in day- by- day decision making for the farm management. Accuracy in models can be achieved either by improving modelling assumptions as such, like incorporating the explicit plant structure instead of using a layer approach when dealing with plant canopy or by improving the online feed of information to the model by coupling models with remote sensor data that are taken in real time. Since remote sensing is not yet widely used in the greenhouse, a way to incorporate remote sensing in the greenhouse environment should first be investigated. In the following subchapters these two approaches are explicitly described.

Plant modelling

Physiological plant models have become an integrated tool of plant science research. These models describe, at different degrees of complexity, plant physiological processes that set light to our understanding of plant functioning and help us develop new cultivation strategies (Fourcaud *et al.*, 2008). A number of physiological plant models have been proposed during the years (e.g. Gary *et al.*, 1995; Marcelis *et al.*, 1998; Heuvelink, 1999; Boote and Scholberg, 2006). These models offer an accurate description of plant growth and its interactions with the environment providing a useful tool in our understanding of plant functioning. Although they make a distinction between different plant organs, they do not consider the plant structure in space. Especially functions like light interception, environmental plant adaptation, competition within and between species for light or nutrients, and assimilate allocations cannot be easily explained if plant structure in space is not taken into account.

Modelling light interception and photosynthesis

In most process based models leaf area is one of the most important crop properties which have a strong physiological impact on plant functioning through light interception and thus photosynthesis. In current models leaf area index (LAI) is input as a single value for the total of the crop canopy. This approach, although it gives a good general estimation of light absorption and photosynthesis, fails to capture the effect of the vertical light distribution in the canopy. This light distribution is affected by canopy architecture and its inclusion in the models gives us the necessary accuracy for the day to day management of the crop.

Functional-structural plant models (FSPM) or virtual plant models (Hanan,1997; Sievänen *et al.* 2000; Godin and Sinoquet, 2005; Vos *et al.* 2007) are terms used to refer to models explicitly describing the development in time of the 3D architecture or structure of plants as driven by physiological processes. These physiological processes are the result of environmental factors. Functional-structural plant models were proposed in the last decade as a means to investigate the function of plant structure in plant development combining traditional plant modelling with a 3D structure (Vos *et al.*, 2007).

Modelling light interception and photosynthesis in a tomato crop

The tomato crop is of high economic importance and one of the most important horticultural crops. Because of the intensity of greenhouse cultivation in the Netherlands optimum cultivation practices as well as genotypes with specific characteristics for this type of cultivation are used. Since experimentation is quite time consuming and cost money, 3D models can become a central tool in searching the ideal crop type and management support when it comes to light interception and photosynthesis. Tomato cultivation is using a high-wire system were the plants are planted in double rows. Such a system is intended to optimize the light distribution in the canopy. However, a high heterogeneity in the plant canopy still occurs and that can have an impact on the local light distribution inside the canopy. A 3D model can accurately calculate the vertical as well as the horizontal light and photosynthetic distribution inside the canopy. Also a 3D model can be used to provide answers to important questions, as to which plant architectural characteristics such as leaf size and shape, leaf elevation angle, leaf orientation, internode length or leaf spatial distribution would give optimal results when it comes to light interception and canopy photosynthesis. The same answers can be given for cultivation practices such as row spacing.

Online monitoring with sensors

When light reaches a surface a part is absorbed, a part is transmitted and a part is reflected depending on the optical properties of the surface. The fraction of light reflected by a surface can be detected with sensors. In plants, reflection can be related to the light emitted during different physiological processes or to morphological parameters.

In the past, optical remote sensing has been developed and used for the online monitoring of parameters such as chlorophyll and nitrogen content, plant stress etc. in field grown crops for example in wheat (Gitelson *et al.*, 2002) potato (Jongschaap, 2006), sugar beets (Clevers, 1997), miscanthous (Vargas et. al, 2002) and rice (Tian *et al.*, 2005). Remote sensing is defined as, 'the small or large-scale acquisition of information of an object or phenomenon by the use of either recording or real-time sensing devices that is not in physical or intimate contact with the object (Lintz & Simonett 1976). Remote sensing can be either at close range where the observations are taken either by sensors positioned on the ground close to the object, or long range where information are gathered by planes or satellites.

Attempts to measure leaf area index (LAI) and Photosynthetically Active Radiation (PAR) interception, with the use of remote sensing, have been successful in field crops. Different approaches have been used for the data collection, with sensors either on short range (ground) or far range (low flights or satellites) (Bouman *et al.*, 1992; Clevers, 1997). Efforts in the field to use reflectance measurements for the estimation of the canopy nitrogen content, biomass and photosynthesis in wheat (Gitelson *et al.*, 2002) potato (Jongschaap, 2006), sugar beets (Clevers, 1997), miscanthus (Vargas et. al, 2002) and rice (Tian *et al.*, 2005), resulted in positive correlations.

Many studies aimed at determining combinations of reflectance of different wavelengths for correcting the effect of disturbing factors (such as old leaves and soil background) on the relationship between crop reflectance and crop characteristics such as LAI. These combinations of the reflectance at different wavelength bands are known as vegetation indices (VI's). Vegetation indices are quantitative measures that are used in an attempt to measure vegetation abundance or vigor (Wang *et al.*, 2005).

Some of the vegetation indices proposed during the years are the Weighted Difference Vegetation Index (WDVI) (Bouman *et al.*, 1992), the infrared/ red (IR/R) ratio and the red edge leaf effect, with the most commonly used the simple ratio (SR) and the Normal Difference Vegetation Index (NVDI) (Wang *et al.*, 2005). SR and NDVI are based on ratios of red (R) to near-infrared (NIR) reflectance where SR= NIR/R and NDVI= (NIR−R)/ (NIR+R). R and NIR ratio-based indices are strongly rooted in the contrasting response of R and NIR reflectance to increases in LAI (Chen *et al.*, 2002).

The core characteristics of remote sensing, i.e. non destructive measurements and real time plant monitoring, indicate it as a highly desirable application for the greenhouse industry. Though remote sensing has been applied quite successfully in the open field, it has hardly been tested in protected cultivations. Open-field methods cannot be directly transported to greenhouses due to complicating conditions, such as existence of greenhouse structure and ground covering with white plastic. In this thesis the conditions, under which remote sensing can be applied for plant monitoring inside a greenhouse, will be investigated.

New technological advancements in the accuracy of reflectance detection have led to linking specific wavelengths with specific plant physiological functions. In that respect one of the interesting applications of crop remote sensing is the monitoring of photosynthetic stress. The so-called photochemical reflectance index (PRI = $(R_{531} - R_{570})/(R_{531} + R_{570})$, where R_{531} and R_{570} are reflectance signals at 531 and 570 nm, respectively, has been used to monitor dynamic changes in photosynthesis under different stress factors (Evain *et al.*, 2004). PRI provides a quick and non-destructive assessment on photosynthesis-related physiological properties of the leaf and canopy (Peñuelas *et al.*, 1994; Méthy *et al.*, 1999; Evain *et al.*, 2004; Weng *et al.*, 2006) for a wide range of species (Gamon *et al.*, 1997; Guo & Trotter, 2004). One of the processes that induce photosynthetic stress is the limited water uptake from the soil. Water stress affects the sufficient opening of stomata, and consequently photosynthetic rate starts to decrease as well as the quantum yield of photosystem II (Chaves *et al.*, 2002). As a result of a decreased rate of CO_2 assimilation, light energy absorbed by the leaf cannot be used to drive photosynthetic electron transport and a part of this energy is dissipated as heat increasing the non-photochemical quenching (Krause and Weis, 1991; Baker and Rosenqvist, 2004). Therefore water stress can be defined as the loss of plant photosynthetic activity as a result of water deprivation.

PRI can be used as an easy and effective way of online monitoring photosynthetic stress. Nowadays that the production of tomato fruits in greenhouses is a highly intensive agricultural industry, where the cost profit limit is marginal and efficient use of resources is vital, such a timely prediction of plant stress is especially important and such a solution worth investigating.

Key objectives

The general objective of this thesis is to develop an accurate 3D model for light interception and photosynthesis simulations and to develop methods for an online monitor of these physiological properties by means of remote sensing.

The individual aims as they are presented in this thesis are:

- Development of a functional structural model simulating the light distribution in a row crop for diffuse and direct light.
- Understanding the role of different plant architectural components in vertical light distribution and define an optimum plant canopy.
- Developing a procedure to estimate the leaf area index and light interception for greenhouse grown row crops based on reflectance measurements.
- Developing a procedure to estimate photosynthetic stress in crops based on reflectance changes at 531nm.

Thesis outline

In **CHAPTER 1** of this thesis a general description of the problem as well as the general objective are presented.

CHAPTER 2.1 is presenting a first description of the 3D model. The effect of row spacing on light interception is also investigated.

CHAPTER 2.2 presents in detail the 3D tomato model. In the chapter the development of the model is described and it is used for the exploration of the spatial light distribution and photosynthesis. In this chapter the effect using an explicit 3D model versus the common modelling approaches for light interception and photosynthesis is addressed.

CHAPTER 2.3 the effect of plant architectural parameters on light interception and photosynthesis is explored. Sensitivity analysis is performed for leaf azimuth angle (phyllotaxis), leaf elevation angle, leaflet angle, leaf length, leaf shape, leaf thickness as well as internode length for the impact of their change to both light interception and photosynthesis. Two different architectural ideotypes are proposed based on the results of the analysis.

In **CHAPTER 3.1** a way of online monitoring of light interception and Leaf Area Index is explored. Reflectance in a number of different wavelength bands as well as NDVI is tested and the best predictor is picked for tomato and sweet pepper crops.

In **CHAPTER 3.2** the use of photochemical reflectance index (PRI) as an indicator of early water stress is presented. In that chapter water stress is described as a function of photosynthetic reduction and the capability of PRI to act as an early water stress indicator is debated.

In **CHAPTER 4** the results of this thesis are summarized and put into a more general context. The shortcomings of the methods used as well as the further steps in this research are discussed.

Chapter 2 Modelling PAR interception and photosynthesis

Chapter 2.1

Towards a 3D structural tomato model for calculating light interception

Published as:

Sarlikioti V., de Visser P.H.B., Marcelis L.F.M. 2011. Towards a 3D structural tomato model for calculating light interception. Acta Horticulturae 893:721-728.

Abstract

A number of physiological tomato models have been proposed the last decades, their main challenge being the correct simulation of fruit yield. For this, an accurate simulation of light interception, and thus photosynthesis, is of primary importance. Light interception is highly dependent of the canopy structure which is affected amongst others by distance between plant rows, distance of plants within the row, leaf pruning and crop variety. In order to simulate these processes, a functional structural tomato model for the simulation of light interception on an individual leaf basis is proposed. The 3D model was constructed using L-systems formalism. For the architectural part of the model, manual measurements of leaf length, width, angle of the leaf main stem to plant stem and leaf orientation were conducted. Diurnal pattern of leaf orientation was also tested. The architectural model was coupled with a nested radiosity model for light calculation. Area per individual leaflet served as input of the light module for calculation of reflection, absorption and transmission of light. The model was used to test different crop planting scenarios on their effect on light interception. Results were then compared with light simulation for a totally homogeneous canopy.

Introduction

A number of tomato models have been proposed during the years (e.g. Gary *et al.*, 1995; Marcelis *et al.*, 1998; Heuvelink, 1999; Boote and Scholberg, 2006). These models offer an accurate description of plant growth and its interactions with the environment providing a useful tool in our understanding of plant functioning. Although they make a distinction between different plant organs, they do not consider the plant structure in space. Especially functions like light interception, environmental plant adaptation, competition within and between species for light or nutrients, and assimilate allocations can not be easily explained if plant structure in space is not taken into account.

Functional-structural plant models (FSPM) or virtual plant models (Hanan,1997; Sievänen *et al.*, 2000; Godin and Sinoquet, 2005; Vos *et al.*, 2007) are terms used to refer to models explicitly describing the development in time of the 3D architecture or structure of plants as driven by physiological processes. These physiological processes are the result of environmental factors. Functional-structural plant models were proposed the last decade as a mean to investigate the function of plant structure in plant development combining traditional plant modeling with a 3D structure (Evers *et al.*, 2005, Buck-Sorlin *et al.*, 2011). For light extinction in particular the knowledge of how the plant develops in space is essential. So the use of such a model for light calculations would probably improve our knowledge of light distribution inside the crop canopy.

Light extinction inside a plant canopy can reach up to 60% while for a crop canopy the light extinction can be up to 90% (Valladares 2003). This variation in incident light availability inside the crop canopy induces extensive structural and physiological modifications. Light variation has also a big effect in plant photosynthetic capacity (A_{max}) which typically, decreases two- to four times from top to the bottom of the canopy (Meir *et al.*, 2002).

The aim of this study was to explore the structural variations inside the tomato plant canopy and use this to develop a structural plant model for tomato. Furthermore, with the use of a structural plant model different plant spacing scenarios and their effect on light distribution inside the canopy were tested.

Materials and Methods

Plant measurements

Experimental set-up.

The experiment was carried out in a high-wired tomato cv. Aranca crop in a glasshouse of Improvement Center (52˚N, Bleiswijk, the Netherlands). Plants were planted in the greenhouse at the end of January. The rows in the greenhouse were northeast- southwest oriented. Two rows of plants were planted in each gully. The distance between the two rows of one gully was 50 cm, the path width was 110 cm and the distance between 2 plants on the same row was 0.53 m. Plant height was 2.60 m. The plants were grown on rockwool (Grodan, type Expert). The greenhouse air was enriched with pure CO_2 at 400 ppm during day time. The lowest leaves were removed every week and plants were lowered such that plant height remained the same throughout the experiment.

Structural plant measurements.

For a complete picture of plant development, detailed measurements of the 3D stem and leaf curvature were manually performed on 3 plants every other week. Measurements were performed for six weeks from 2 of July to 13 of August 2008. A protractor and a ruler were used for the measurements. The measurements included leaf elevation angle, leaf length, leaf width and leaf orientation. In our experiment we defined leaf elevation angle as the angle of leaf petiole to the horizontal where it is attached to the stem (Boonen *et al.*, 2002). The rosette at the top of the plants with leaves smaller than 2 cm was considered as first node. Digitization of the plants was attempted with the use of a Fastrak 3D digitizer (Polhemus Inc, Colchester, VT, USA) but was not always possible. This was probably due to a powerful disturbance of the magnetic field of the digitizer due to metallic parts (heating pipes) positioned inside the canopy.

The plant was divided in three zones (upper, middle and lower zone) and the above mentioned structural plant characteristics were measured. Every zone had a length of 90 cm except the lowest one that had a length of 80 cm. Measurements were carried out weekly on 3 randomly picked leaves per zone in 13 replicate plants.

Tomato composite leaf consists of a large terminal leaflet and up to 8 lateral leaflets, which can also be compounded. Many smaller leaflets or folioles may be interspersed between the larger leaflets depending on the cultivar (Atherton and

Rudich, 1986). In our cultivar the leaf was composed of a big terminal leaflet, 3 pairs of larger leaflets and two pairs of smaller leaflets alternately placed. Measurements of angles of all leaflets for 10 randomly selected leaves in each plant level were taken, in order to determine a relationship between leaflet angle and leaflet position in the leaf. Leaflet angle was defined as the angle between the leaflet petiole and the leaf petiole.

Given that the angle of leaves to the horizontal directly affects the flux of solar radiation per unit area (Falster and Westoby, 2003), measurements to establish the diurnal pattern of the leaf elevation angles were also made. Leaf elevation angles of 15 plants were measured early in the morning (09:00 hours) and in the afternoon (15:00 hours).

Light measurements.

Incident light was measured with the use of Sunscan (Delta-T, UK). The sensor was positioned perpendicular to the plant row. Measurements were taken every 25cm from the top of the canopy, in 8 different spots in the greenhouse.

Model description.

The model consisted of two modules:

- Structural module. In this module the spatial development of the plant was described in terms of symbols according to L-systems formalism (Lindenmayer and Prusinkiewicz, 1990). The plant was structured as a number of phytomers in deferent developmental stages. A phytomer is defined as the basic structural unit which for our model consisted of an internode and a composite leaf. Relationships of leaf elevation angle to node number, leaf length to node number and leaf width to node number as well as leaflet area to leaf area and leaflet angle to petiole node number were determined by regression analysis. These relationships where used as input for the development of the structural part of the model. Leaflets were represented with rectangular shapes in the model in an approximation of their real shape. Plant and row spacing also served as an input. The basic simulation unit of the model included 20 plants (5 plants per row, 4 rows).
- Light module. A nested radiosity model (CARIBU) developed by Chelle and Andrieu (1998) was used for light calculations. The module

calculates light absorbance on the leaflet level. Multiple light scattering on leafs surface was taken into account for light calculations. The model assumed an infinite plant canopy for the calculations (infinite canopy was achieved by multiplying the basic simulation unit into space). Reflectance and transmittance parameters for the upper and lower side of the leaf were input in the model. Output values of light interception were given in leaflet as well as leaf level. Model calculation assumed diffuse light conditions. Forty three virtual light sources were symmetrically arranged around the crop in order to achieve these conditions.

A more detailed description of the model is provided in chapter 2.2.

Lambert- Beer.

In many crop models Lambert- Beer's law (Monsi and Saeki, 1953) is used for the simulation of PAR interception. According to the law in a uniform infinite randomly distributed canopy of absorbing leaves, it can be shown that the amount of photosynthetically active radiation intercepted (I) by a crop can be given by the following equation:

$I= (1-\rho)*I_0*(1-e^{-k*L})*100$,

where ρ stands for canopy reflection coefficient, I_0 is the radiation level at zero canopy depth, L the leaf area index of the canopy, and k is the light extinction coefficient. Extinction coefficient was set to 0.65 as reported by Papadopoulos and Pararajasingham, 1997) and confirmed by our own measurements (data not shown).

Statistical analysis.

Statistical analysis was performed with Genstat 11 software (VSN International Ltd., Herts, UK). Differences of leaf elevation angle, length and width between three plant heights were tested with Linear Mixed Model (REML). Leaf elevation angles were compared between morning and afternoon with General Linear Models, repeated measurement analysis. Curves for leaflet angle to leaf petiole and for light interception to LAI were fitted with Regression Analysis. Goodness of fit was estimated by coefficient of determination (R^2). Statistical differences between curves was tested, by testing the statistical differences between the coefficients of the regression curves. P was 0.05.

Results and Discussion

Structural characteristics of tomato leaves were monitored at three different depths in the canopy (Fig. 2.1.1). In the upper part of the plant, leaves had on average a slightly positive angle to the horizontal, while leaves in the middle and lower part of the canopy showed a negative angle to the horizontal. The negative value of -25^{o} found in our experiment correspond to the mean value used in the tomato model of Higashide (2009). Length and width were smallest in the top part of the canopy, as these leaves were not yet full-grown. There was no statistically significant difference in leaf size between middle and lower part of the canopy.

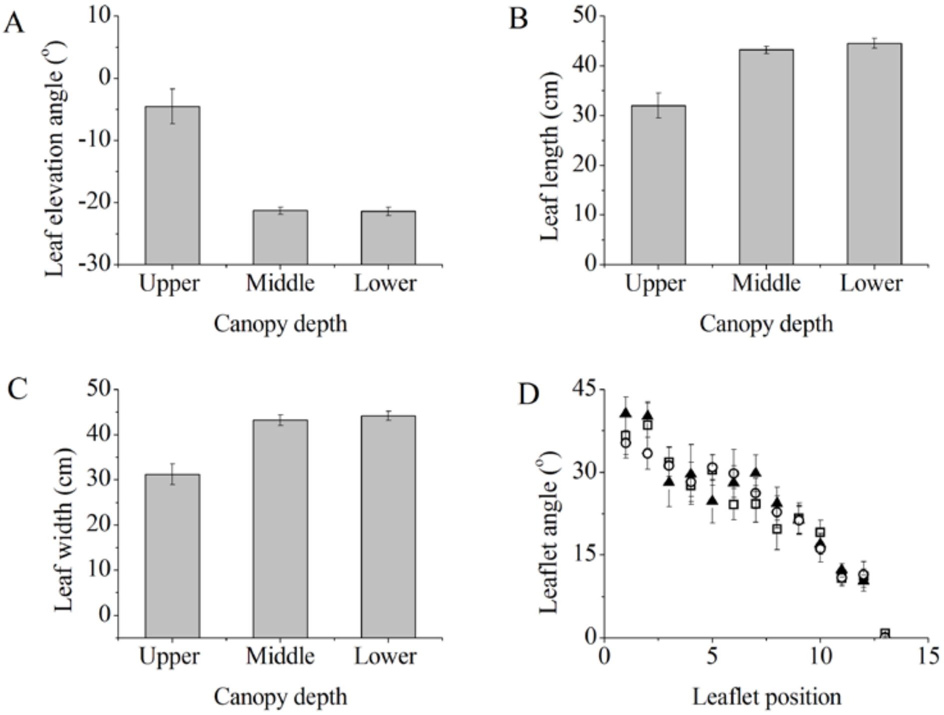

Fig. 2.1.1. Structural measurements for three different depths in a tomato canopy. Mean values of leaf elevation angle to the horizontal (A), leaf length (B) and leaf width (C) are presented for the three different canopy depths as well as the relation between leaflet angle to leaf petiole and leaflet position (1 is most proximal leaflet to the stem) for top (□), middle (○) and lower leaves (▲) (D). Error bars represent s.e of the mean. Data are averages of 5 weeks, 13 plants per week. Data for graph D are averages of 10 leafs per canopy level.

Leaflet angles diminished from a maximum of 40° for the most distal leaflet to 0° for the most proximal leaflet (Fig. 2.1.1D). Leaflet angle showed no statistically significant differences between the three different canopy levels implying that the leaflets' angle is in fact an internal characteristic of the plant and dependent only on the position of the leaflet on the leaf.

Leaf elevation angle differed significantly between morning and afternoon only for the upper leaves (average leaf elevation angles were -3,1° and -7,6° for morning and afternoon subsequently) (Fig. 2.1.2). A diurnal pattern was not observed at the middle and lower leaves. Forseth (1990) observed no diurnal changes in leaf elevation angles to the horizontal of a number of species (for example cotton, beans, Solanum), but he did find significant changes of the plant angle to the azimuth. He also linked this azimuth movement to an increase in light interception and a proportional increase of productivity. Although Forseth does not differentiate leaf elevation angle behavior between different canopy depths, his work is an indication that measurements of only leaf elevation angle to the horizontal is not adequate to conclude about a diurnal pattern on tomato leafs. Leaf elevation angle changes can also be linked to turgor loss of plant cells due to daily transpiration. So further research is needed as for the causes of this behavior on upper tomato leafs.

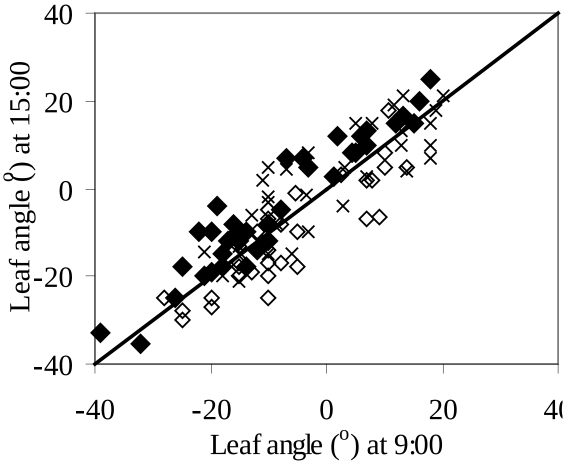

Fig 2.1.2. Comparison of leaf elevation angle (angle to the horizontal) in the morning (09:00hours) and afternoon (15:00hours) at three different heights of the canopy. Symbols ♦, ◊, and x refer to upper, middle and lower part of the plant consequently. The line is 1:1.

The above described structural measurements were used to develop a structural tomato model. The accuracy of the model was tested against measured values. In general the difference of the measured values versus the calculated values was of 4% with a maximum difference of 10% at the lowest point. The model was used to test three different crop planting scenarios and compare them to results of Lambert- Beer equation which is currently used in most crop modeling for light simulation. The three different planting scenarios consisted of: i. Normal culture. Planting distances were the same as the ones found in the experiment (50cm between the two rows of one gully and 110 cm of path ii. Big path. The distance between the rows in the same gully was reduced to 15 cm and the path width increased to 145cm width. iii. Even-distance rows. The plants were equally distributed in space (distance between plants at the same gully as well as the pathway was equal to 80cm). In order to test the accuracy of the tomato's model light calculations, the light interception of a completely homogeneous crop was compared to Lambert- Beer calculations (Fig 2.1.3). Plant density was 4.1 plants per m^2, with 1 stem per plant. It was assumed that plant size and structure was the same in all scenarios.

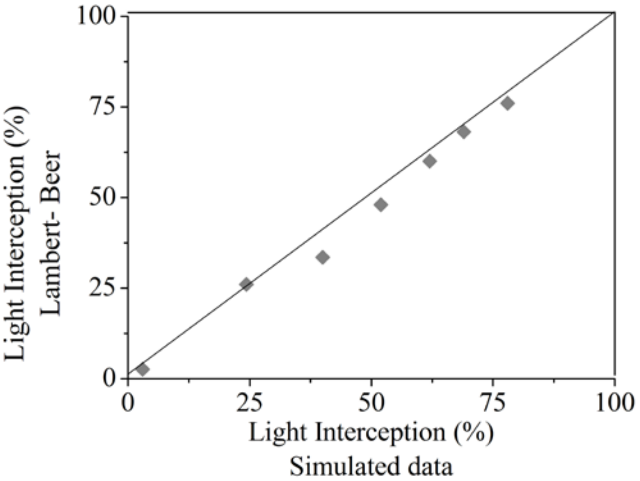

Fig 2.1.3. Comparison of light interception (%) between simulation data for a totally homogeneous canopy and Lambert- Beer law calculations for the same canopy. Data points are from the top (0 interception) to the bottom of the canopy (100).

In general row structures led to higher light interception in the upper part of the plants (LAI≤1.5) and lower interception in the lower part compared to homogeneous crop simulation. Light interception reaching at the deeper part of the crop increased with path width and resulted in 30% light reaching soil level in our "big path" treatment. In accordance Papadopoulos and Pararajasingham (1997) reported that light penetration increased and therefore light interception decreased with the increase of plant spacing in a crop. Similar effects have also been reported in other crops such as corn (Stewart *et al.*, 2003). Cournède *et al.* (2007), in their structural model concluded that deviations in calculations of light interception from Lambert- Beer law were mainly because the latter assumes a even distribution of the leaf elevation angles. The assumption of an even leaf distribution nevertheless has been challenge in the past. Maddonni *et al.*, (2002) reported that in case of row structure the leaf orientation is changed and it is biased in being positioned perpendicular to the row. Also Toler et al (1999) found that k factor is dependant to the row distances and is decreasing with the increase of the row spacing. It would be argued that changes in k factor in relation to plant spacing are because of morphological changes in plants and mainly orientation and leaf elevation angle. The calculation of k factor from our model simulation shows that even if the structural characteristics of the plant stay the same, average k factor decreases with the increase of plant spacing (Fig. 2.1.4B). Inside the canopy k-factor is also decreasing from the top to the bottom. This decrease is directly linked with the decrease of leaf elevation angle. In practice leaf elevation angles are not constant but are dependant on the planting strategy. Extinction coefficient calculated for even-distance rows showed no differences to the one of homogeneous canopy. Nevertheless further increase of the path between the rows led to a decrease of the k factor, a decrease that reached 50% in case of “big path” treatment.

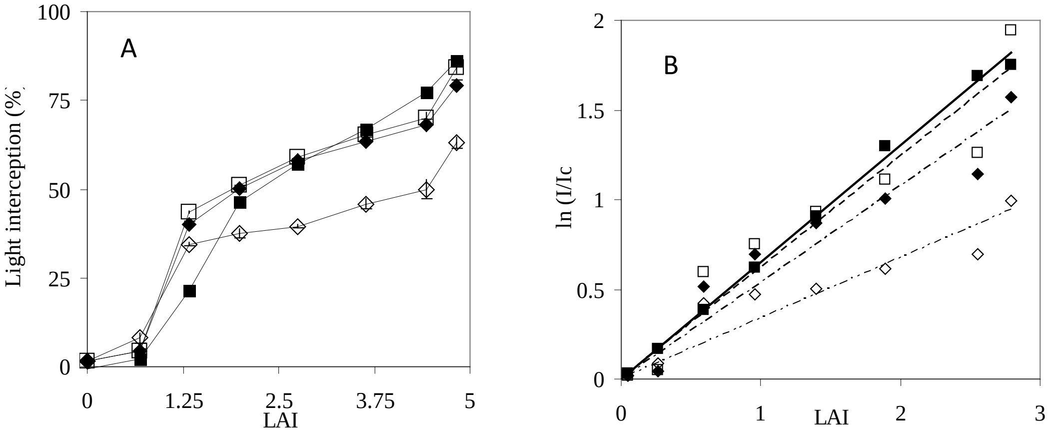

Fig. 2.1.4. A. Calculated light interception for different canopy structures. Light interception was calculated by structural model for plants grown in rows with path width of 110 (♦), 80 (□) and 45cm (◊). The ■ symbols correspond to calculation

from Lambert- Beer B. Average extinction coefficients k for the different treatments. LAI refers to cumulative leaf area from top to bottom of plant canopy.

CONCLUSIONS

In tomato plants structural differences in leaf can mainly be observed in the upper 90cm of the plant, in the still developing zone. Changes of structural plant characteristics affect directly light interception by the crop canopy. Nevertheless even if plant structure stays the same light penetration can be manipulated easily by changing row spacing in the crop, thus affecting light interception and potentially plant production.

Chapter 2.2

Exploring the spatial distribution of canopy light interception and leaf photosynthesis by means of a functional structural model

Published as:

Sarlikioti V., de Visser P.H.B., Marcelis L.F.M. 2011. Exploring the spatial distribution of light interception and photosynthesis of canopies by means of a functional-structural plant model. Annals of Botany, 107:875-883.

Abstract

At present most process-based models and the majority of three dimensional models, include simplifications of plant architecture that can compromise the accuracy of light interception simulations and, accordingly, canopy photosynthesis. The aim of this paper is to analyze canopy heterogeneity of an explicitly described tomato canopy in relation to temporal dynamics of horizontal and vertical light distribution and photosynthesis under direct and diffuse light conditions. Detailed measurements of canopy architecture, light interception and leaf photosynthesis were carried out on a tomato crop. These data were used for the development and calibration of a functional-structural tomato model. The model consisted of an architectural static virtual plant coupled with a nested radiosity model for light calculations and a leaf photosynthesis module. Different scenarios of horizontal and vertical distribution of light interception, incident light and photosynthesis were investigated under diffuse and direct light conditions. Simulated light interception showed a good correspondence to the measured values. Explicitly described leaf elevation angles resulted in higher light interception in the middle of the plant canopy compared to fixed and ellipsoidal leaf elevation angle distribution models, although the total light interception remained the same. Fraction of light intercepted at a north-south orientation of rows differed from east-west orientation by 10% on winter and 23% on summer days. The horizontal distribution of photosynthesis differed significantly between the top, middle and lower canopy layer. Taking into account the vertical variation of leaf photosynthetic parameters in the canopy, led to ca. 8% increase on simulated canopy photosynthesis. Leaf elevation angles of heterogeneous canopies should be explicitly described as they have a big impact both on light distribution and photosynthesis.

Introduction

Physiological plant models have become an integral tool of plant science research. These models describe, at varying degrees of complexity, plant physiological processes that improve our understanding of plant functioning and help us develop new cultivation strategies (Fourcaud *et al.*, 2008). Physiological models, or process-based models (PBMs), usually focus on plant production and development, by describing biophysical processes as rates using ordinary differential equations (ODE' s) or stochastic processes (Marcelis *et al.*, 1998; Heuvelink, 1999, Gayler *et al.*, 2006). Light interception is one of the most important functions, as it drives photosynthesis and, therefore growth. Although highly dependent on canopy structure (Vos *et al.*, 2010), light interception is usually computed in PBMs as a function of leaf area index (LAI) and extinction coefficient (Lai *et al.*, 2000; Baldocchi *et al.*, 2000). In most models the extinction coefficient is determined by fitting a Lambert–Beer law relation to experimental data or is estimated as a function of a certain leaf elevation angle distribution. Although these approaches give a good estimation of total light interception of a crop, they fail to capture the effect of plant and canopy heterogeneity on light interception and, therefore, on photosynthesis (Vos *et al.*, 2010). Since plant architecture is influenced by a number of processes (such as genotype, water availability, cultivation practices or diseases), models that explicitly describe the impact of these processes on plant architecture can be a useful tool in our understanding of such phenomena, for example the effect of wilting on light capture.

In recent years, techniques have become available for developing functional–structural plant models (FSPMs), which are also called 'virtual plants' that combine the modelling of physiological processes with the 3D architecture of the plant. This combination boosts the capability of models to simulate the interaction between plants and their environment (Hanan, 1997; Sievänen *et al.*, 2000; Godin and Sinoquet, 2005; Vos *et al.*, 2007). The 3D plant structure is especially important for the description of light interception and, therefore, the photosynthetic capacity of plants. Three-dimensional models require a detailed quantification of plant structure in space (Vos *et al.*, 2007). Plants are considered as the sum of distinct units called phytomers that are formed repeatedly based on a hierarchical system (Barthélémy and Caraglio, 2007). Static 3D plants coupled with radiation models have proven to be valuable tools in investigating the effect of single-plant architecture as well as crop structure on light interception and canopy photosynthesis (Vos *et al.*, 2010). Zheng *et al.*, (2008) showed that certain plant types with steeper leaf elevation angles exhibited a higher light penetration of the canopy when sun elevation was high. Therefore, even simple static virtual plants have a great potential for crop breeding research.

Despite the advantage of virtual plant models over PBMs in their explicit description of plant architecture, still in such models we still often need to approximate 3D structure. Leaf elevation angle is assumed either as constant (Najla *et al.*, 2009) or to follow a spherical or ellipsoidal distribution (Rakocevic *et al.*, 2000; Farque *et al.*, 2001). This approach is mainly due to the tediousness and the time-consuming nature of the measurements involved (Fourcaud *et al.*, 2008). Such an approach may give robust results in the case of crops that show a particularly regular and coordinated development, such as wheat and rice (Evers *et al.*, 2005; Drouet and Pagès, 2007; Zheng *et al.*, 2008). However, in order to fully understand the light distribution in the plant canopy and explore the full impact of crop architecture on light interception and photosynthesis of row crops with a high canopy (such as tomato), functional–structural models should incorporate a detailed description of all architectural parameters in general and leaf elevation angles in particular.

The aim of this research was to analyse the canopy heterogeneity of an explicitly described tomato canopy on horizontal and vertical light distribution and photosynthesis under direct and diffuse light conditions at different times of the year. In order to do so, a static functional structural tomato model was developed and then used as a tool for analysing the impact of canopy heterogeneity.

Materials and Methods

Experiment

A tomato (*Lycopersicum esculentum* var. Aranca) crop was planted in December 2006 in a commercial greenhouse in Bleiswijk, the Netherlands (52^o). Measurements were performed in July and August of 2007 when the plants were 1.75 m tall. During this period average temperature in the greenhouse was 17.5^{o}C, average daytime CO_2 concentration was 371 μmol mol^{-1} and relative air humidity was set at 73%. Daily outside global radiation was 40 MJ during the time of the experiment. Plants were grown in double rows, with rows oriented from north to south. The distance between the double rows was 1.2 m (path), the distance between each row of the double row (within the row distance) was 0.4 m and the distance between plants within the row was 0.3 m, resulting in a plant density of 4.1 stems/m^2.

Measurements of architectural development

Each week for six weeks, angle, length, width, internode length and azimuth orientation of all leaves of five plants were manually measured weekly with a ruler and a protractor. Measurements were made during the morning hours (09:00-13:00). Leaf elevation angle was determined as the angle of the leaf petiole with the horizontal at the leaf insertion point on the stem. The first leaf longer than 2 cm was defined as leaf number 1. Azimuth angle was determined as the leaf horizontal angle measured clockwise from a constant point defined as “north”. North (or 0 degrees) was defined as the point perpendicular to the plant rows when facing towards the inner side of the double row.

The tomato plant has composite leaves with 10 to 13 leaflets. Leaflet angle was measured on 10 leaves at different canopy heights on 6 plants in total. The angle of the leaflet to the horizontal at the point that it connects to the petiole was defined as leaflet angle.

The crop leaf area was estimated non-destructively through leaf length and leaf width measurements at the widest point. The relationship between the area of a leaf and its length and width was estimated by taking photographs against a white background of 25 randomly chosen leaves from various canopy depths with a digital camera (Canon, IXUS 800 IS) positioned perpendicular to the leaf. A ruler was set next to the leaf for calibration of the image scale during image processing. ImageJ (National Institutes of Health, Bethesda, MD) was used for image analysis. A relationship was established between leaf length and width, and leaf area. This relationship was used to calculate the leaf area index from length and width measurements on all dates. Leaflet length and leaflet area were also measured on these 25 leaves in order to establish a relationship between the leaflet length and leaflet area.

Light interception measurements

Photosynthetically active radiation (PAR) interception was measured with a 0.8m light rod in the crop and a reference sensor above the crop (Sunscan, Delta-T, UK) under diffuse light conditions (overcast sky). The light rod was positioned perpendicular to the row and light interception was measured from the top to the bottom of the plant at 0.25m height intervals. The measurements were repeated at eight selected spots in the crop, once a week for seven weeks. Measurements were also taken in the middle between the double row and in the middle of the path. For these measurements the sensor was positioned parallel to the crop at three different plant heights (0.5 m, 1 m and at the base of the plant).

Photosynthesis measurements

Photosynthesis light response curves were measured with the use of a portable open gas exchange measurement device (LCpro$^+$, ADC, UK). PAR levels were set to 0, 100, 250, 500, 700 and 1400 µmol m^{-2} s^{-1}. CO_2 concentration and relative humidity were set to ambient greenhouse values (360 µmol mol^{-1} CO_2 and 73% RH, respectively). On three dates during the experiment, measurements were done on a leaf at two different canopy heights (upper and middle, respectively) on six plants and at three different dates during the experiment. Upper, middle and bottom canopy height layers were defined as intervals of 0.5 m from the top to the bottom of the canopy.

Model Description

The functional–structural model presented here consists of three different modules (Fig.2.2.1):

- The architectural module. This is a static model that describes the plant structure in space and the topology of the various organs, using the L-systems formalism (e.g. Prusinkiewicz, 1999).
- A nested-radiosity module. The input of this module is the 3D plant architecture and the position and the intensity of the light sources, using the model of Chelle and Andrieu (1998). The light emitted by the light sources is traced through the canopy and the light absorbed by each leaflet is given as an output.
- The photosynthesis module. This module calculates gross photosynthesis based on the biochemical model of Farquhar (1980).

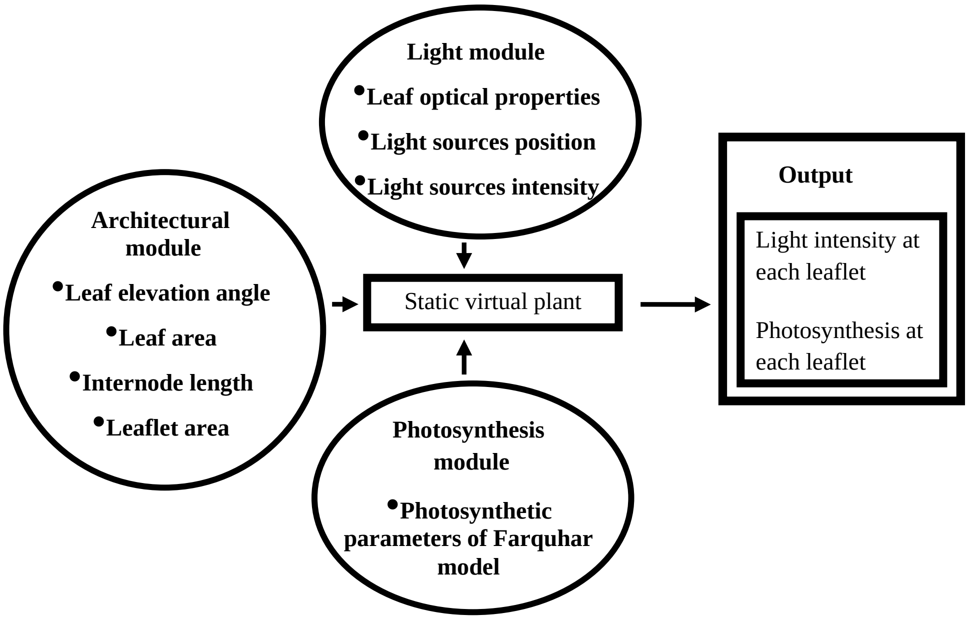

Fig. 2.2 1. Model flow chart.

Architectural module

The basic structural unit of the module is the phytomer. A phytomer consists of an internode, a composite leaf and a bud containing an apex. The basic unit is repeated 27 times in order to form a complete plant. Every three leaves a generative shoot forms a flower truss. Trusses are not represented in the model. In order to account for the light interception from the trusses a fake truss was inserted every 3 leaves. This fake truss was represented as a small leaf with the same length as the length of the truss and the same number of leaves as the number of fruits. Relationships of the change of the leaf elevation angle and length to the node number were established for each date. An average internode length of 7.5 cm was used for all plants.

The tomato plant has composite leaves that vary in size. Typical leaves consist of a large terminal leaflet and up to eight large lateral leaflets. Many smaller lateral leaflets may alternate with the large leaflets. The leaflets are usually petiolate and irregularly lobed, depending on the genotype (Atheron and Rudich, 1986). A representative leaf structure of the particular genotype in terms of leaflet number was chosen for the construction of the model and was measured in detail. The composite tomato leaf was modelled as a branch structure in which each leaflet is represented as a discrete lamina based on equations of leaflet angle (^{o}) and leaflet area (cm^2).

Relationships of the leaflets angle to the leaf petiole, as well as the leaflet length to the leaf length as determined in the experiment were incorporated in the model.

The above mentioned relationships were derived from experimental measurements as described in the '*Measurements of architectural development'* section. The visual output of the architectural model is presented in Fig. 2.2.2.

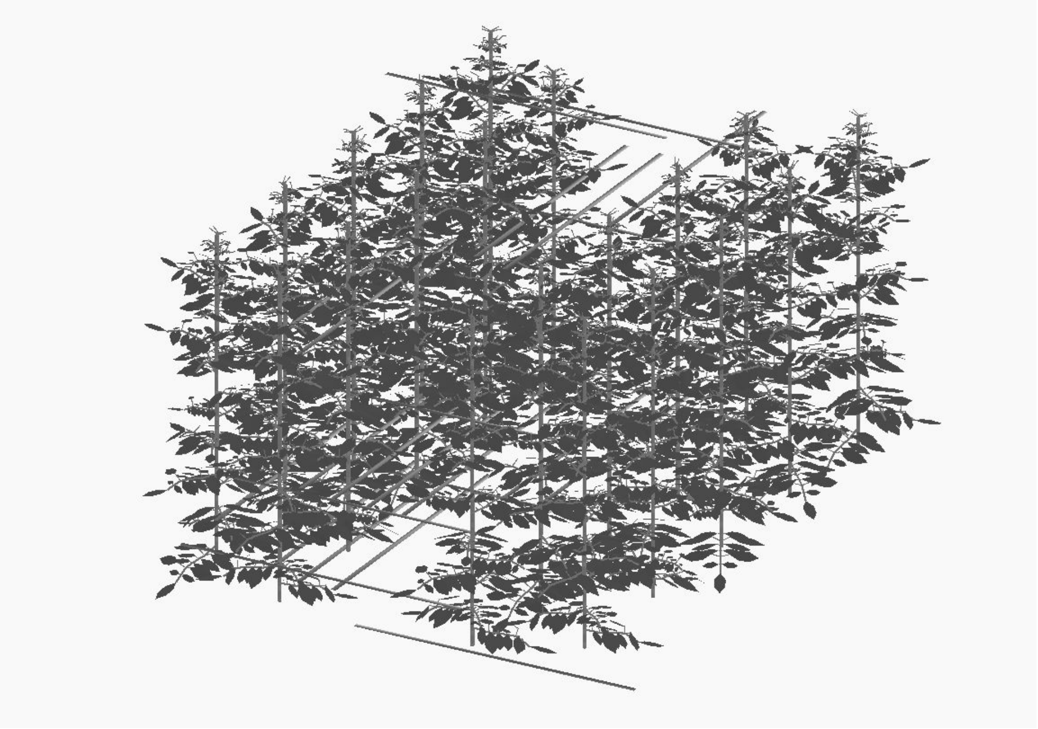

Fig 2.2.2. Example of the visual output of the 3D tomato model. The basic unit of the model is two plant rows of five plants each. Lines along the path and the plant canopy represent the visual sensors used for the model calibration

Radiosity module

PAR reaching the crop consists of a direct and a diffuse light component (Spitters, 1989). For the simulation of diffuse light conditions, 48 directional light sources were positioned uniformly in a hemisphere around the canopy, simulating a uniformly overcast sky. The light intensity of diffuse light conditions was 460 μmol PAR m^{-2} s^{-1}. For the simulation of direct sunlight, a bright sky was simulated with light sources that were given x,y,z coordinates similar to the sun's trajectory on two distinct dates (21 December and 21 June). For direct light conditions, the intensity of the light sources at a half hour time step, was derived from the 10-yearly average of light incidence on these dates under Dutch conditions (daily radiation was equal to 7 MJ d^{-1}

in winter and equal to 50 MJ d^{-1} in summer). The nested radiosity module calculates the light absorbed by every leaflet, by using a radiosity approach for a basic crop unit and subsequent nesting of the unit to account for the surrounding canopy (Chelle and Andrieu, 1998). Multiple scattering was calculated on a canopy of 20 plants (as calculated by the architectural module). These 20 plants formed the basic model unit. In the nested radiosity module the basic unit is multiplied infinitely in space in order to preclude phenomena associated with border effects (e.g., too high levels of light incidence from the sides). Reflectance and transmittance of the full spectrum of the upper and lower sides of the tomato leaves were measured with the use of an InstaSpec IV CCD spectrometer (Oriel, Stradford, CT, USA**)** and a LiCor 1800-12 integrating sphere (LICOR Inc. Lincoln, NE, USA). Twelve leaves from different canopy heights were measured in total and the average values were inputted into the model(upper side reflectance= 0.17, upper side transmittance= 0.06, lower side reflectance= 0.12, lower side transmittance= 0.03).

In order to validate the model with the measured data, we introduced to the model ‘virtual’ sensors that were situated at the measurement spots. These sensors were represented as surfaces with the same dimensions and optical properties as the Sunscan sensor and were positioned inside the canopy at the same heights with measured values.

In order to investigate the effect of leaf elevation angle distribution on light interception and photosynthesis, we made comparisons between our model (EXPL), which explicitly describes leaf elevation angles, a 3D model with a fixed leaf elevation angle (-20^o for all leaves in the canopy) (CONST) and a 3D model with an ellipsoidal leaf elevation angle distribution (ELLIP, χ=2.7 where χ is the ratio of the horizontal semiaxis length to the vertical semiaxis length of an ellipsoid) were made.

Photosynthesis module

Photosynthesis is calculated according to the biochemical model of Farquhar *et al.* (1980) on the basis of absorbed light. The module calculates photosynthetic rate at leaflet level according to the equation:

$$A_v = J * \frac{p_i - \Gamma}{4 * (p_i + 2 * \Gamma)} - RD \qquad \text{(eqn 2.2.2)}$$

where p_i is the intercellular partial pressure of CO_2 in Pa, Γ is the CO_2 compensation point in Pa, RD the dark respiration in μmol CO_2 m^{-2} s^{-1} and J is the rate of electron transport rate per unit leaf area and is calculated from the following equation:

$$J = \frac{(\alpha * p + J\max - \sqrt{\alpha * p * J\max^2 - 4 * \theta * \alpha * p * J\max}}{2 * \theta} \qquad \text{(eqn. 2.2.1)}$$

where α and θ are coefficients from the data fitting, Jmax the potential electron transport rate (μmol electrons μmol^{-1} photons) and p the light absorbed by the leaflet surface ($\mu mol\ m^{-2}\ s^{-1}$). Based on the measurements as described in '*Photosynthesis measurements*' coefficients of the photosynthesis equations were differed between the upper and the middle layer of the canopy. For the lower canopy, we assumed the same coefficients as in the middle canopy. For the upper canopy, the coefficient values were α= 0.1, θ= 0.69 and Jmax= 124.4 and for the middle and lower canopy layer α= 0.1, θ= 0.65 and Jmax= 75.18).

Model calculations for photosynthesis calibration showed a good correlation with the measured data at two different canopy heights (R^2= 0.93; data not shown).

Total canopy photosynthetic rate was compared between the model that explicitly describes leaf elevation angles (EXPL), a 3D model with a constant leaf elevation angle (CONST) and a 3D model with an ellipsoidal leaf elevation angle distribution (ELLIP). For the EXPL and ELLIP models, two different scenarios were investigated: in one, scenario the photosynthesis parameters of the top of the plant were used for the whole canopy, while in the other scenario we attributed different photosynthetic parameters for the upper (0-0.75 m) layer and the middle and the bottom canopy layers (0.75 m-1.8 m). For all canopies, LAI was kept constant at 3.1.

Statistical analysis

Statistical analysis was performed with GenStat 12th Edition. Regression analysis was applied to derive the various architectural and biochemical relationships implemented in the model, except for the parameters of the light response curves of photosynthesis, which were derived from a mixed linear model.

Results

Developing and calibrating the crop architecture module

Dynamics of structural properties of the crop remained more or less constant during the two months of the experiment. The upper leaves showed a positive leaf elevation angle with respect to the horizontal, while the lower leaves showed a negative leaf elevation angle. Below the tenth youngest phytomer, leaf elevation angle did not vary with phytomer angle (Fig. 2.2.3A). Leaf length rapidly increased from the top to the 7th phytomer from 2 to 30 cm and then remained almost constant in the lower leaves (Fig. 2.2.3B). However, leaf area increased continuously from the top to the bottom of the canopy (Fig. 2.2.3C). Most leaves were positioned perpendicular to the plant row, towards the path and the middle of the plant row (Fig 2.2.3D).

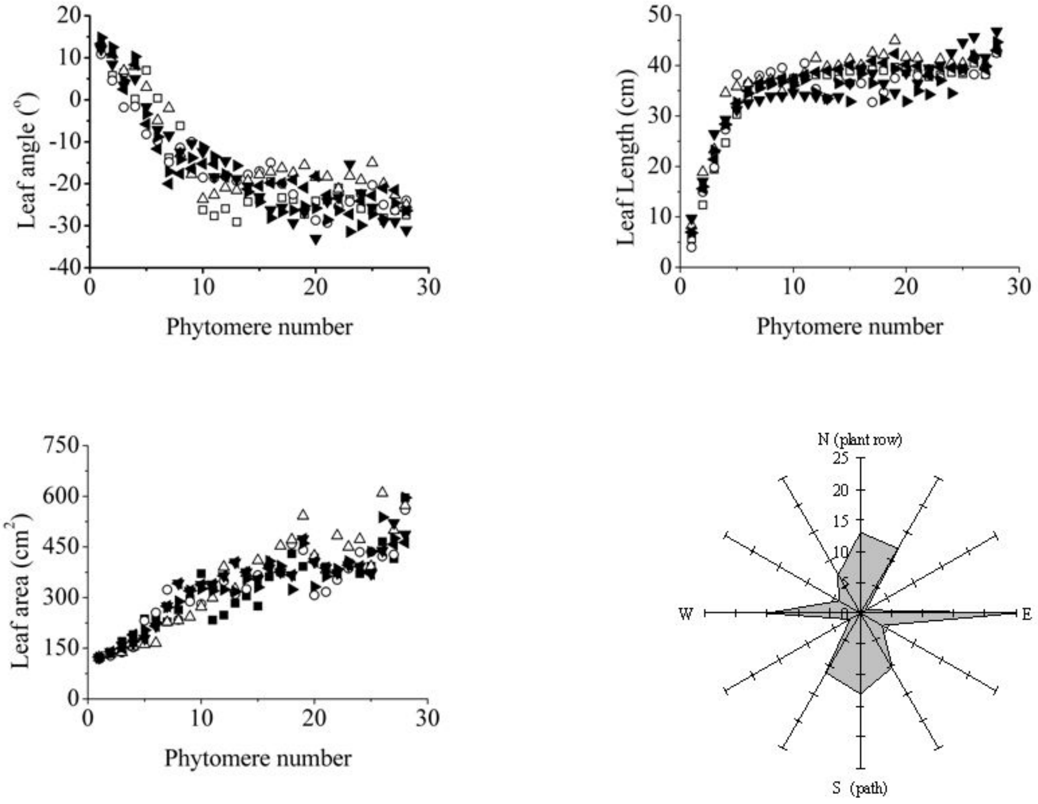

Fig 2.2.3. Relationship between A) leaf elevation angle to the horizontal plane, B) leaf length, C) leaf area in relation to phytomere number starting from the top of the plant and D) leaf azimuth angle distribution. Each symbol represents a specific week in fig.3 A-B-C. Vertical bars in Fig. 3D represent the number of leaves per leaf orientation class.

Leaflet angles depended on leaflet position on the leaf petiole. The terminal leaflet had an angle of zero and the leaflets tended to be more erect towards the plant stem. Leaflets in tomato leaves occur in pairs opposite to each other. In Fig. 2.2.4A every two leaflets represent one pair (i.e. leaflets 1 and 2 are one pair etc.). There was no significant difference between the leaflet angles of the two leaflets of the same pair (Fig 4A). A positive linear (0.43+0.52*leaflet length, r^2= 0.88, p<0.001) relationship was observed between leaflet length and leaflet area (Fig 2.2.4B).

The above relationships were used to derive the parameters and equations of the architectural model.

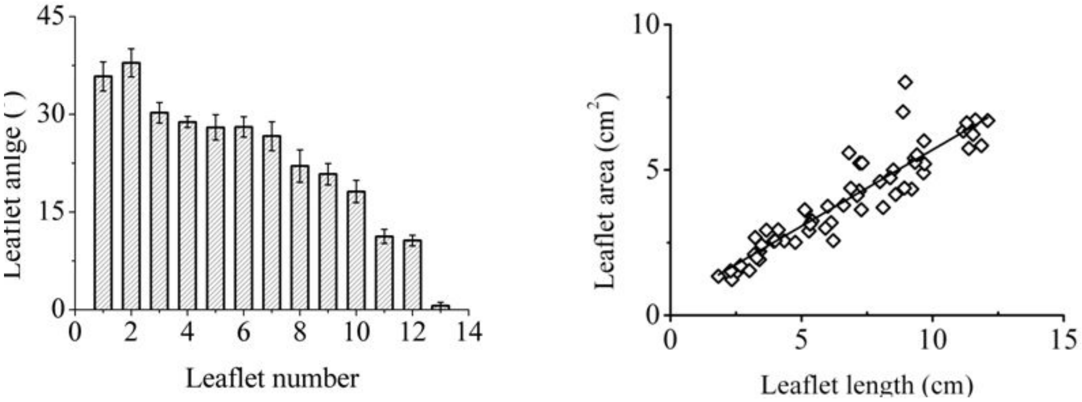

Fig 2.2.4. A) Leaflet angle with respect to the horizontal in relation to the leaflet position on the leaf petiole. The position counting starts from the leaflets nearer to the stem and ends with the terminal leaflet. Every two leaflets form a pair positioned opposite to each other on the leaf petiole. Each column represents the average of 10 leaves ±.SE mean. B) Relationship of leaflet area to leaflet length (y= 0.15x+0.25, R^2=0.81). N = 10 leaves.

Light interception

Light interception was measured and simulated for six dates. For each simulation date, crop structure was based on leaf area and leaf elevation angles as measured on dates corresponding to the light measurements. Simulated light levels corresponded well to the measured data (Fig 2.2.5). An underestimation of light interception was observed for simulated values at the top of the crop.

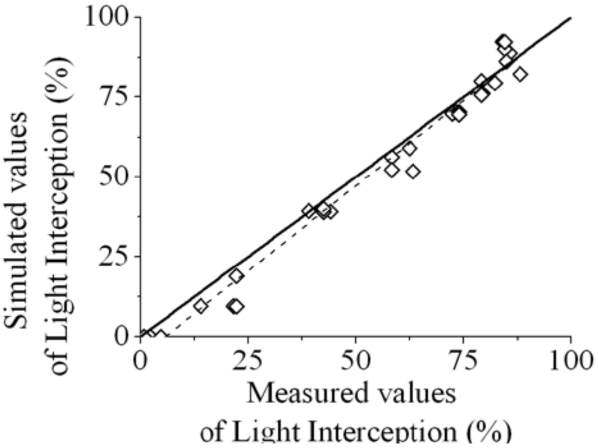

Fig 2.2.5. Measured vs. simulated values of light interception. Values are from 6 weeks of measurements in 8 different canopy heights. Continuous line is 1:1.

In a comparison between EXPL, CONST and ELLIP models, no differences were found in total light interception, but differences were observed for the middle of the canopy. The use of a constant angle led to a 17% underestimation of light interception under diffuse light conditions and a 23.6% underestimation under direct light conditions compared to the EXPL model. Ellipsoidal distribution led to a 7.6% and an 11% underestimation under diffuse light conditions and direct light conditions respectively (Table 2.2.1). These differences were observed only in the middle canopy layer.

In order to investigate the effect of the row crop on the horizontal light distribution and the simulation capabilities of the model, virtual sensors were

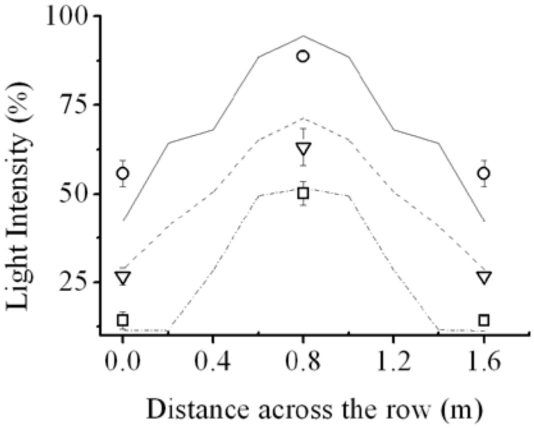

Fig 2.2.6. Measured and simulated horizontal light distribution in a row tomato crop. The light intensity is plotted against the plant row length at three different plant canopy heights (50 cm (○), 100 cm (∇) and 175 cm (□)). Lines represent simulated values while symbols represent measured values ± SEmean. Plant rows are located at 20cm and 140cm while the middle of the path is located at 80cm.

positioned parallel to the crop row, in the middle of the space between two rows and in the middle of the path at three heights (Fig 2.2.6). Light intensity decreased from the top to the bottom of the canopy and from the centre of the path to the row (Fig. 2.2.6).

Table 2.2.1. Comparison of three different leaf elevation angle modelling approaches with respect to effect on light interception and photosynthesis. Values for light interception and photosynthetic rate are for the total canopy. Values in brackets refer to the middle of the canopy (0.7 5m-1.25 m from the top of the plant) where differences were observed.

Leaf elevation angle distribution	Light intercepted (%)		Photosynthesis with one set of parameters (μmol m^{-2} s^{-1})		Photosynthesis with two set of parameters (μmol m^{-2} s^{-1})	
	Diffuse	Direct	Diffuse	Direct	Diffuse	Direct
Fixed angle (CONST)	77 (43)	80 (41)	20 (8.8)	27 (12.3)		
Ellipsoidal distribution (ELLIP)	77 (48)	80 (47)	24 (10.3)	29.2 (12.9)	21 (10.7)	29.6 (13.2)
Explicitly described leaf elevation angles (EXPL)	77 (52)	80 (55)	27 (10.6)	30.3 (13.2)	24 (10.9)	30.9 (13.7)

Direct light was calculated for 21 of June. The light intensity for direct light conditions was derived from the 10-yearly average of light incidence on these dates under Dutch conditions (4.6 μmol m^{-2} s^{-1}at sunrise, 3109 μmol m^{-2} s^{-1} at noon and 23 μmol m^{-2} s^{-1} at sunset). For diffuse light conditions a light intensity of 460 μmol m^{-2} s^{-1} was considered. Calculations were done when it was assumed that all leaves of the canopy had the same photosynthetic properties or with two sets of photosynthetic properties, where the properties of the top layer differed from those of the middle and lower layers.

Taking into account the perpendicular positioning of the leaves to the plant row, leaves positioned towards the path absorbed more light per unit leaf area than leaves positioned towards the middle of the plant row (which received 30%, 43% and 88%

less in the upper, middle and bottom canopies, respectively). Simulation data showed an underestimation of the light intensity at the various plant heights compared to the measured data.

Both sun elevation and plant orientation to the sun's trajectory had an effect on light interception (Fig. 2.2.7). Fraction of light intercepted was in all cases higher during winter than summer. Light interception increased substantially for plants rows with a north-south orientation than plant rows with an east-west orientation. This trend was observed for both times of the year.

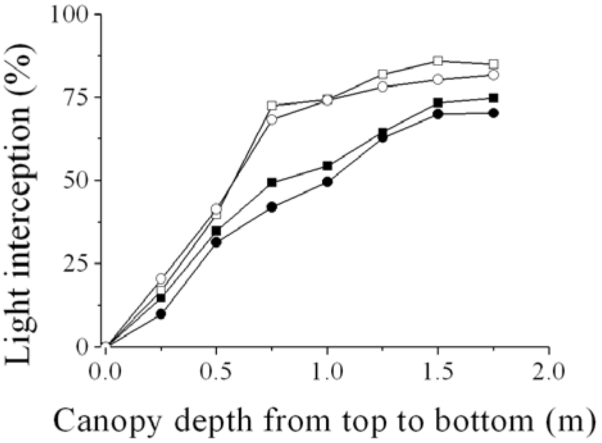

Fig 2.2.7. Seasonal variation in light interception for 21st of December (circles) and 21st June (squares) for a north-south (open symbols) and east-west (closed symbols) row orientation. LAI was 3.1. Calculations were performed for exactly the same canopy structure on both dates.

Photosynthesis

In order to investigate the horizontal distribution of photosynthesis with the model, leaves pointing towards the path were chosen, like those upon which the manual measurements were performed. Leaves located in the higher canopy layer photosynthesized considerably more than those positioned in the middle or the bottom of the canopy. Differences in simulated photosynthesis were not observed between the middle and bottom simulated canopy layers, because the same photosynthetic parameters were used for these two layers and light levels were almost equal. In the higher canopy layer, photosynthesis increased rapidly from 8 to 35 µmol m^{-2} s^{-1} from the inside leaflets to the outer ones, while in the lower canopy layers photosynthesis ranged from 2.5 to 14.8 μmol m^{-2} s^{-1} (Fig. 2.2.8).

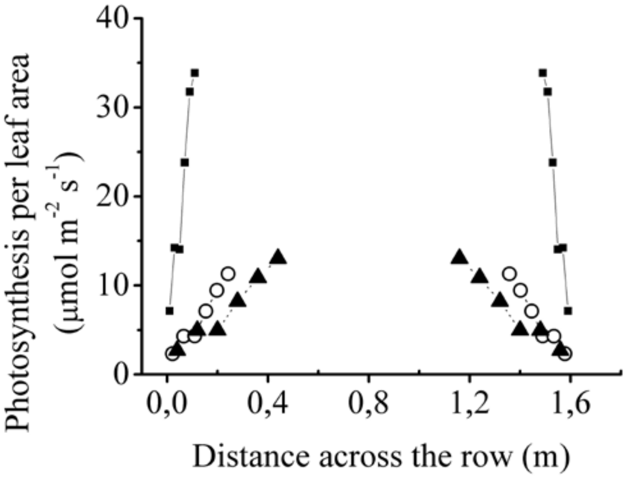

Fig 2.2.8. Horizontal distribution of photosynthesis in the crop path. In this graph only leaves positioned towards the path at three different canopy heights (0.5 m (■), 1 m (○) and 1.75 m (▲)) are used. Each data point is the average of a pair of leaflets and includes the result from 2-3 leaves per plant from 20 plants depending on the leaves position. Simulation was performed under diffuse light conditions.

The rate of the increase from the inside to the outer leaflets was relatively higher in the lower layer (4.3 and 6 for the higher and the lower layer, respectively). Total canopy photosynthesis differed in total 26% (for diffuse light) and 11% (for direct light) between the EXPL and CONST models (Table 2.2.1). Total photosynthesis differences between the EXPL and ELLIP model were 11% (diffuse) and 4% (direct light) respectively. For light interception differences simulated in the middle canopy layer,, the CONST model led to a 16% underestimation of photosynthesis under diffuse light conditions and to a 7% underestimation under direct light conditions in comparison with the EXPL model. The ELLIP model led to a 3% underestimation under diffuse and under direct light conditions compared with the EXPL (Table 2.2.1). The differences in photosynthetic rate when using the same photosynthetic parameters for all leaves compared to the use of two sets of photosynthetic parameters in top and middle leaves was 12.5% and 1.3 % (for diffuse and direct light respectively) for the ELLIP model and 11% and 2 % (for diffuse and direct light, respectively) for the EXPL.

Discussion

The aim of this research was to analyse the temporal dynamics of the horizontal and vertical light distribution and photosynthesis in relation to canopy heterogeneity under direct and diffuse light conditions.

The spatial position of plant organs has been studied in view of their possible adaptation to their local environment. Such strategies can aim either at maximization of plant production efficiency or at minimization of the impact of stress-inducing conditions, such as drought or light inhibition (Björkman and Powles, 1984). Leaf dimensions and especially leaf elevation angles are important in assessing these plant strategies, as they are directly linked to the acquisition of light. Smaller and more upright leaves are found in the top of the canopy, which allows light penetration to the lower layers, while lower leaves have a higher area so as to ensure maximum light absorption (Pearcy *et al.*, 1990). Since leaf elevation angle is an important architectural phenotypic characteristic of a plant, it should be explicitly incorporated in functional–structural plant models. Dong *et al.*, (2008) proposed a functional–structural tomato model in which leaf elevation angle is randomized according to an ellipsoidal distribution. Najla *et al.*, (2009) and Higashide (2008) used a fixed value to describe all leaf elevation angles independent of their position in the canopy. These approaches assume a leaf distribution that is not affected by cultivation practices or the specific plant genotype. However, Sinoquet *et al.*, (2005) showed that this is not the case and that likely factors for the deviation from the randomness in leaf positioning in a canopy can be linked to leaf size and angle. In this study, we compared three leaf elevation angle distributions (CONST, ELLIP and EXPL) and showed that the previous approaches to modeling leaf elevation angle can lead to an underestimation of light interception in the middle canopy ranging from 4% to 15%, depending on the light conditions. Although light interception in the canopy is the same for all models, the change in the middle canopy layer led in photosynthesis simulation to differences of 3-8%. Therefore, in order to correctly model the heterogeneity of plant canopy, leaf elevation angles should be explicitly described.

Another point of importance in terms of plant architecture is the azimuth leaf orientation. Atheron and Rudich (1986) reported that in a single tomato plant, leaves were evenly distributed around the stem with a phyllotaxis angle of 135°. Our data show that tomato plants grown in a row crop system tend to rearrange their leaves in a more systematic way, namely almost perpendicular to the plant row. Similar phenomena, where leaves are turned away from shady spots, have been reported for maize (Maddoni *et al.*, 2001), trees (Cournède *et al.*, 2007) and cucumber (Kahlen *et al.*, 2008). Dauzat *et al.* (2008) observed that branch placement was density-dependent in cotton and that at high densities sympodial and monopodial branches tended to orient towards the space between rows. This placement of leaves and branches is probably due to the plant's strategy for maximizing light interception and should also be taken into account when modelling plant architecture.

Row crop systems are the most common cropping systems used in horticultural and agronomic crops. This system, which was developed mainly to facilitate harvest and crop management, allows higher light penetration inside the plant canopy. In our experiment, light intensity increased towards the middle of the path, as also observed by Stewart *et al.* (2003) in maize and Louarn *et al.* (2008) in grapevines. Our simulation showed that of the amount of light reaching the top of the canopy, 50% reaches the ground floor in the middle of the path. Light direction combined with light intensity has a direct effect on light interception. A seasonal pattern in fraction of light intercepted has been reported for many species (Gilbert *et al.*, 2003, Cassela and Sinoquet, 2007). Light interception follows a seasonal pattern with, on average, a lower fraction of light intercepted during summer than during winter. A main factor is the change in solar elevation changing during the year. The higher solar elevation in summer months, results in an orientation of light rays more perpendicular to the plant canopy, resulting in a higher light penetration and lower interception. Interestingly, row orientation seems to affect substantially light interception with north-south orientation giving a higher light interception than east-west orientation. The same phenomenon has been reported by Palmer (1989) and Borger *et al.* (2010). Kahlen *et al.*, 2008 reported that light direction and intensity are linked to a possible growth advantage of certain plants positions inside the canopy, mainly by leaf rearrangement towards the unshaded patches of the canopy or leaf photosynthetic acclimation to altered light status. Architectural adaptations of plants to the seasonal light patterns would, in this context, be worth investigating.

Leaf elevation angle and vertical leaf distribution are highly relevant for the daily amount of photosynthesis as was shown in the results. Increase in light in lower canopy layers resulted in a higher relative increase in photosynthesis. A probable explanation for this is the leaf acclimation to lower light intensities and the physiological age in lower layers in the canopy (Niinemets, 2007). Leaves situated within the two rows received a substantially lower amount of light than leaves at the same height situated towards the path. If lower photosynthesis is partly an effect of acclimation to lower light levels, it stands to reason that leaves oriented towards the middle of the plant rows will have a lower photosynthetic rate than leaves at the same height that are oriented towards the path, and very likely different photosynthetic potential. A common experimental approach for photosynthesis is to take measurements only in the upper and middle canopy and only of leaves oriented towards the path. However, model calculations showed that the use of one more set of photosynthetic parameters can lead to a 7%-10% difference in photosynthesis prediction. So it stands to reason that when a significant part of the canopy is oriented towards the intra-row space with concomitant higher photosynthetic potential, predictions of crop photosynthesis will be inaccurate. Virtual plant models are able to cope with this, given the proper data. Chelle (2005) also pointed out the need for a new modelling approach that will combine the organ microclimate with the general plant environment. He demonstrated the temperature differences that can be measured at various plant organs and how the use of FSPMs can improve our understanding of the effect of these differences on the plant processes. A similar approach should be used for photosynthesis modelling as it would improve our understanding of the impact of various crop strategies on photosynthesis.

Conclusions

Leaf elevation angles of heterogeneous canopies should be explicitly described as they have a big impact both on light interception and on photosynthesis. Comparisons between 3D models with explicitly described leaf elevation angles and models with standard leaf elevation angle distributions resulted in differences of 4-15%, depending on the light conditions and the number of the sets of photosynthetic parameters. In this frame, functional-structural models can play an important role in our understanding of light distribution along vertical and horizontal gradients caused by crop architecture. Such a tool can be useful in practise not only in yield prediction, but also in experimentation planning as well. However, steps should be taken to move from a static to a dynamic crop so as to incorporate the seasonal adaptation of the plants.

Chapter 2.3

Searching for the ideal plant structure with a functional-structural plant model. How plant architecture affects light absorption and photosynthesis in tomato

Accepted as:

Sarlikioti V., de Visser P.H.B., Buck-Sorlin G.H., Marcelis L.F.M. 2011**.** How plant architecture affects light absorption and photosynthesis in tomato: Towards an ideotype for plant architecture using a functional-structural plant model. Annals of Botany *in press.*

Chapter 2.3 Using FSPM to optimize plant structure in tomato

Abstract

Manipulation of plant structure can strongly affect light distribution in the canopy and photosynthesis. The aim of this paper is to find a plant ideotype for optimization of light absorption and canopy photosynthesis. Using a static functional structural plant model (FSPM), a range of different plant architectural characteristics was tested for two different seasons in order to find the optimal architecture with respect to light absorption and photosynthesis. Simulations were performed with a FSPM of a greenhouse-grown tomato crop. Sensitivity analyses were carried out for leaf elevation angle, leaf phyllotaxis, leaflet angle, leaf shape, leaflet arrangement and internode length. From the results of this analysis two possible ideotypes were proposed. Four different vertical light distributions were also tested, while light absorption cumulated over the whole canopy was kept the same. Photosynthesis was augmented by 6% in winter and decreased by 7% in summer, when light absorption in the top part of the canopy was increased by 25%, while not changing light absorption of the canopy as a whole. The measured plant structure was already optimal with respect to leaf elevation angle, leaflet angle and leaflet arrangement for both light absorption and photosynthesis while phyllotaxis had no effect. Increasing the length-to-width ratio of leaves by 1.5 or increasing internode length from 7 to 12 cm led to an increase of 6 – 10% for light absorption and photosynthesis. At high light intensities (summer) deeper penetration of light in the canopy improves crop photosynthesis, but not at low light intensities (winter). In particular internode length and leaf shape affect the vertical distribution of light in the canopy. A new plant ideotype with more spacious canopy architecture due to long internodes and long and narrow leaves led to an increase in crop photosynthesis of up to 10%.

Introduction

Light absorption is an important factor for determining crop yield, being one of the driving forces behind plant photosynthesis, and at the same time is highly dependent on single-plant architecture as well as on overall canopy structure (Niinemets, 2007). Plant architectural characteristics (such as the number and geometry of organs, i.e. their shape and position within the plant and the canopy), are genotype specific, while at the same time highly dependent on the climatic conditions at the time of their initiation and development (Godin, 2000). Falster and Westoby (2003) have shown that steeper elevation angles in a number of species improve absorption at higher sun elevations and, therefore, carbon gain through assimilation as it allows more light to penetrate to the lower leaves. While the importance of leaf elevation angles for an improved light absorption strategy at the level of the whole plant has been shown in a number of studies (Pearcy and Yang, 1998; Sinoquet *et al.*, 2005), reports about the importance of leaf phyllotaxis are contradictory as some studies did and some did not observe effects on the light absorption of the canopy (Brites and Valladares, 2005). Furthermore, aspects such as the elevation angles of the leaflets of composite leaves have not been, to the best of our knowledge, previously investigated. Both leaf shape and size are important aspects of leaf morphology affecting mutual shading of leaves and light absorption of the canopy (Falster and Westoby, 2003).

The quantitative exploration of the specific effects of each plant architectural characteristic on light absorption and photosynthesis was hardly possible until the introduction of spatially explicit models considering plant architecture at the organ level (Vos *et al.*, 2010). General crop models are powerful tools towards a better understanding of plant processes and for testing case scenarios (Marcelis *et al.*, 1998; Vos *et al.*, 2007). More specifically, functional-structural plant models (FSPM) have been introduced as a relatively recent paradigm in plant modelling where physiological processes are coupled with an explicit 3D plant structure (Vos *et al.*, 2010), often supplied with a mutual feedback between physiology and structure. Modelling on the basis of a 3D structure gives the opportunity to investigate more in-depth the effect of specific architectural characteristics such as leaf angle, leaf length, or leaf shape (Vos *et al*., 2010). Sinoquet *et al.* (2005) used FSPMs to show the effect of leaf and branch distribution on light absorption of trees. Sarlikioti *et al*. (2011) developed a static tomato FSPM to explore the spatial distribution of light absorption and photosynthesis in a tomato canopy. They demonstrated the importance of an explicitly described leaf angle distribution for simulating light absorption and photosynthesis. Accurately calibrated FSPMs can convey a better understanding of the light distribution inside the canopy and also provide us with a tool to define the optimal set of architectural characteristics for maximizing canopy photosynthesis, allocation of assimilates to growing organs and ultimately crop yield.

Donald (1968) defined the "crop ideotype" in the context of cereal breeding as an idealized plant type with a specific combination of characteristics favourable for photosynthesis, growth, and grain production based on knowledge of plant and crop physiology and morphology. He argued that it would be more efficient to define a plant type that was theoretically efficient and then breed for this. The crop ideotype is thus an idealized crop consisting of a plant type with a specific combination of characteristics based on the detailed knowledge of morphological and physiological plant traits (Peng *et al.*, 2008) as well as mutual interactions among plants of the canopy. These traits often are also contributing to plant architecture. Modifications of the arrangement and size of leaves can affect light availability especially in the lower parts of the canopy and alter leaf photosynthetic activity by adjusting light harvesting efficiency (Werner *et al.*, 2001). As an example, a decrease in leaf clustering can increase light absorption and enhance photosynthetic productivity at canopy level (De Castro and Fetcher, 1999). Morphological characteristics such as leaf inclination and leaf shape are often inherited as simple traits (i.e. under the influence of one or a few major genes) in the plant (Thurling, 1991) and can be used to create a more open canopy structure. These breeding traits can be strongly affected by the environment under stress conditions (Valladares and Niinemets, 2007). Tomato (*Solanum lycopersicum* L.) is a species that exhibits a high variability in vegetative morphology (Peralta and Spooner, 2000) ranging from small leaves with a few leaflets to big ones with many leaflets. Lately, studies of the genetic basis of this variation at the leaf level have shown that there are leaf-specific genes that control its shape and morphology (Frary *et al.*, 2004). This genetic background knowledge in combination with the detailed information on the effect of leaf topology and geometry on light absorption and photosynthesis could help to identify or approximate the theoretical optimum of plant architecture.

The objective of this study is to define a plant ideotype for greenhouse-grown tomato with respect to optimization of light absorption and photosynthesis at the canopy level. A range of different plant architectural characteristics were tested under light conditions of a typical summer and winter day in order to define the ideal for each case. We tested two hypotheses: 1) The manipulation of plant structure of a greenhouse-grown tomato crop can lead to substantial improvement in crop photosynthesis even when leaf area index and leaf photosynthetic characteristics remain unaltered. 2) A more spacious canopy architecture improves crop photosynthesis. For this purpose we used the static functional structural tomato model developed by Sarlikioti *et al*. (2011).

Material and methods

Model description

Simulations were performed with a functional structural tomato model. This model was built as a parametric open L-system using the *cpfg* language within the platform L-Studio (Prusinkiewicz, 1999) consisting of three modules (model structure and parameterization has been described in detail in Sarlikioti *et al*. (2011):

Architectural module

This is essentially a static 3D reconstruction of tomato plant architecture, in which each plant consists of 27 phytomers with the first phytomer being the one at the top of the plant (basipetal ranking). A phytomer is the basic architectural unit consisting of a leaf and an internode, the leaf itself being made up of a leaf rachis, a central midrib, and 13 individual leaflets, each one of them consisting of a blade and a petiole, of which one is terminal and 12 are lateral. The blade of each leaflet is described as a flat polygon instead of one with a curvature for reasons of calculation efficiency. Characteristics are: leaf elevation angle (defined as the angle between the leaf rachis, at its insertion point to the stem, and the horizontal plane), leaf length, (defined as the distance from the leaf insertion point at the stem to the tip of the terminal leaflet), and leaf width (defined as the distance between the tips of two longest lateral leaflets). The construction of the leaf leads to a 3D object. The architectural model was parameterized during a summer period for a fully grown tomato crop (*Solanum lycopersicum* L., var. ‘Aranca’) grown in a high-wired greenhouse system in Bleiswijk, The Netherlands (52°01'N, 4°32'E) with a density of 4.1 stems m^{-2} (see Sarlikioti *et al.*, 2011). LAI was equal to 3.6 m^2 of leaf area per m^2 floor area. Every week the lowest leaves were removed, the plants were lowered and kept at the same height throughout the season as it is common practice in the Netherlands. Therefore plant architecture remained “static” at least during each season (summer, winter). For that reason a static modeling approach was used.

Light module.

Light calculations are based on a nested radiosity approach developed by Chelle and Andrieu (1998). The module requires as an input the amount of light absorbed by each plant organ, the leaf transmittance and reflectance coefficients for the upper and lower side of the leaf (upper side reflectance = 0.17, upper side transmittance = 0.06, lower side reflectance = 0.12, lower side transmittance = 0.03) and the light from the light sources that were used to simulate the sky. In the Netherlands, greenhouses are usually larger than 1 ha. In order to better approach real greenhouse cultivation conditions an infinite canopy was assumed in which the basic unit is theoretically reproduced in all directions, thus avoiding a border effect during the calculations. The basic unit consisted of 20 plants arranged in two double rows with five plants per row. The distance between the double rows was 1.2 m (path), the distance between each row of the double row (within the row distance) was 0.4 m and the distance between plants within the row was 0.3 m.

Photosynthesis module

Photosynthesis calculations are based on the biochemical model of Farquhar *et al.* (1980). This model requires as input the computed light absorbed per leaflet in the model and photosynthetic parameters that were derived from the experimental data (Sarlikioti *et al.*, 2011). All photosynthetic parameters (V_{max}, α, θ, *etc.*) were assumed invariate with canopy depth, except for J_{max}, which followed a logistic pattern from the top to the bottom of the canopy (J_{max} at the top = 265 µmol e^- m^{-2} s^{-1}, J_{max} at the bottom= 180 µmol e^- m^{-2} s^{-1}).

Case studies

In this study we attempted to define the importance of each architectural component for optimum light absorption and canopy photosynthesis by performing a sensitivity analysis of the most important components as they appear in the literature. Each component is described in detail below.

Simulations were performed under summer and winter light conditions and for two distinctive days (December 21 and June 21) using ten-yearly (2000-2010) averages, with an hourly time step, of direct and diffuse light incident at these dates under Dutch conditions. Daily global radiation outside the greenhouse was equal to 3.5 MJ m^{-2} d^{-1} in winter and 26 MJ m^{-2} d^{-1} in summer. Transmissivity of the greenhouse was 60%. For direct light conditions light sources were given the same Cartesian co-ordinates as the solar trajectory. To simulate diffuse light, light sources were positioned on a virtual hemisphere around the architectural mock-up. In total 48 directional light sources were used, with daily intensity equal to 1.5 MJ m^{-2} d^{-1} in winter and 7.5 MJ m^{-2} d^{-1} in summer. The fraction of diffuse light was equal to 0.3 for winter and 0.22 for summer. Leaf temperature was 23 ^{o}C and calculations were performed for CO_2 levels equal to 400 ppm. Computed hourly assimilation rates were integrated to daily amount of assimilates produced.

Vertical light distribution scenario

In order to understand the effect of vertical light distribution on canopy photosynthesis, we constructed four light absorption curves (Fig. 3.2.1A) that were based on the light absorption of the reference structure for each date. Light absorption was increased and decreased from the first to the 8th phytomer by 10% and 25%, respectively, while the light absorption of the canopy as a whole remained the same. Canopy photosynthesis was calculated for each curve.

Leaf elevation angle

Here, we studied the effect of modifying leaf elevation angle with respect to the original angular distribution. As a starting value we assumed an explicitly described leaf angle distribution (Sarlikioti *et al.*, 2011). Simulations were carried out by adding or subtracting 15 ^{o} or 30 ^{o} to the measured value of each leaf in the canopy. In the reference crop the leaf angle distribution ranged from -23 ^{o} for bottom leaves to 15 ^{o} for top leaves.

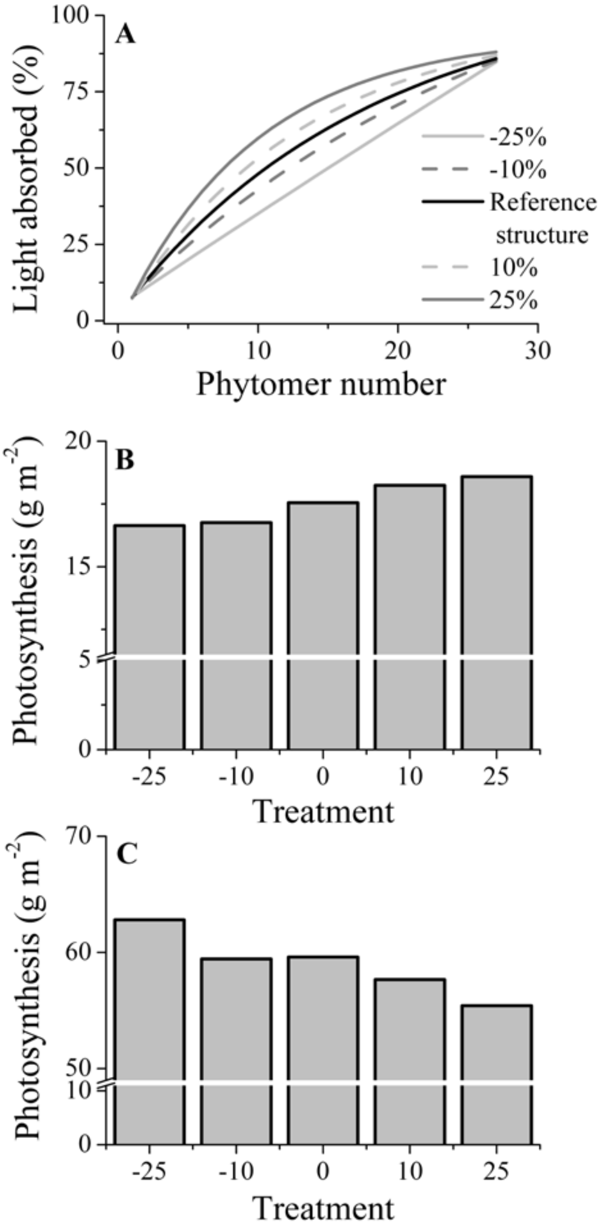

Fig. 2.3.1. Five different vertical light absorption profiles (top graph) were imposed as treatments for studying the optimal profile for maximum crop photosynthesis. The absorbed light is the cumulated value from top (phytomer 1) to bottom of the canopy. Photosynthesis at 1200 h (g CH_2O m^{-2} h^{-1}) is presented for each treatment for winter (middle graph) and summer (lower graph) light conditions. Treatments for absorption profile are -25%, -10%, 0 , +10% and +25%. These values refer to the percentage change in light absorption in the upper eight phytomers of the canopy while light absorption cumulated over the whole canopy was kept constant.

Leaf phyllotaxis

The phyllotactic angle is defined as the angle between two subsequent leaves along the plant stem. Atherton and Rudich (1986) reported that tomato plants follow a common 135 ^{o} phyllotaxis. In reality, however, plants tend to rearrange their leaves, thereby deviating from this value. In previous experiments we found that phyllotactic angle on a tomato row crop is about 160 ^{o} (Sarlikioti *et al*., 2011). Here we assess the effect of leaf phyllotaxis for values equal to 110 ^{o}, 135 ^{o}, 150 ^{o} and 180 ^{o}.

Leaflet angle

As leaflet angle we define the angle between the leaflet petiole and the leaf rachis. In a composite leaf, leaflet angles steadily decrease from the proximal to the terminal leaflet. On average the measured leaflet angle of tomato leaves was 22 ^{o}, ranging from 35 ^{o} for the basal leaflets to 0 ^{o} for the terminal leaflet (Sarlikioti *et al*., 2011). In the present study this original distribution (reference crop) was compared with leaves having all leaflets oriented at an angle of 22 ^{o} or 0 ^{o}.

Leaf shape

The ratio between leaf length and leaf width was used as a convenient measure of leaf shape. In the model the default value of the length-to-width ratio is equal to 1.02. Leaf ratios of 0.5, 0.75, 1.25, 1.5 and 2 times the original ratio were tested for their effect on light absorption and canopy photosynthesis. These ratios resulted in a range of leaf shapes from wide and short leaves to long and narrow ones. For all simulations the leaf area index per plant was kept constant at 3.6 m^{2} leaf area per m^{-2} floor area.

Leaflet arrangement on a leaf

The area of each leaf is equal to the sum of the areas of each of its leaflets. In the leaf of a typical tomato cultivar, pairs of big and small leaflets alternate. A wide range of leaf types can be found in tomato. We investigated the (crop scale) effect of leaf types with all leaflets having the same area as well as the effect of having fewer but bigger leaflets (Fig. 2.3.2). Also the effect of an increase of petiole length by 20% of the original value was investigated. In all simulations the area per leaf (sum of all leaflets) was kept the same.

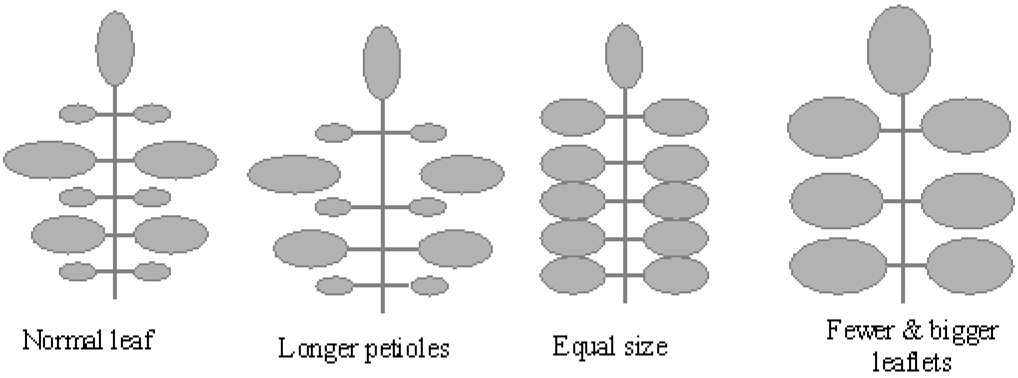

Fig. 2.3.2**.** Schematic representation of leaflet arrangement scenarios. The total area per composite leaf was kept constant.

Internode length

As internode length we define the distance between the insertion points of two consecutive plant organs (leaves or trusses). Measured average internode length was 7.45 cm (st.dev= 0.8) (Sarlikioti *et al.*, 2011). Here we investigated the effect of internode lengths of 3 cm, 5 cm, 10 cm and 12 cm. In all simulations leaf number (13) and area per leaf (m^2) were kept constant.

Ideotyping scenarios

After the above assessment of architectural characteristics of the tomato plant we found that a number of parameters improve the light absorption as well as canopy photosynthesis. Based on these results, we designed the following two scenarios:

- Scenario A: For this scenario architectural characteristics were chosen, which by themselves had produced a minimum increase in canopy photosynthesis by 4%. Internode length was set to 10 cm, length-width ratio to 1.5, all other architectural characteristics were kept the same as in the reference structure.
- Scenario B: This scenario aimed at reconstructing an open structure of the canopy: The internode length was again set to 10 cm and for leaflet arrangement the leaves with longer petioles (Fig. 2.3.2) were chosen. All other architectural characteristics remained the same as in the reference structure.

Results

Vertical light distribution scenario

Imposing five different vertical light absorption profiles while keeping light absorption cumulated over the whole canopy constant (Fig 2.3.1A), showed some distinct effects on canopy photosynthesis. Under winter light conditions at noon, canopy photosynthesis increased by 6% when absorption in the top part of the canopy (upper 8 phytomers) was increased by 25% (Fig. 2.3.1B). In contrast, under summer light conditions, for the same increase of light absorption canopy photosynthesis was decreased by 7% (Fig. 2.3.1C).

Leaf elevation angle

Highest light absorption was achieved with the original leaf angle distribution (leaf angle decreasing from 15 ^{o} degrees for top leaves to -23 ^{o} degrees for bottom leaves) under winter light conditions (Fig. 2.3.3A) while under summer conditions it was maximum when elvation angle was changed by 30 ^{o} (Fig. 2.3.3B). Changes in leaf angle of ±15 ^{o} degrees decreased light absorption by 5% and an increase of the leaf angle to +30 ^{o} led to a decrease of 6% under winter conditions (Fig. 2.3.3A). A decrease in leaf angle by 30 ^{o} distinctively decreased light absorption by 18%. Under

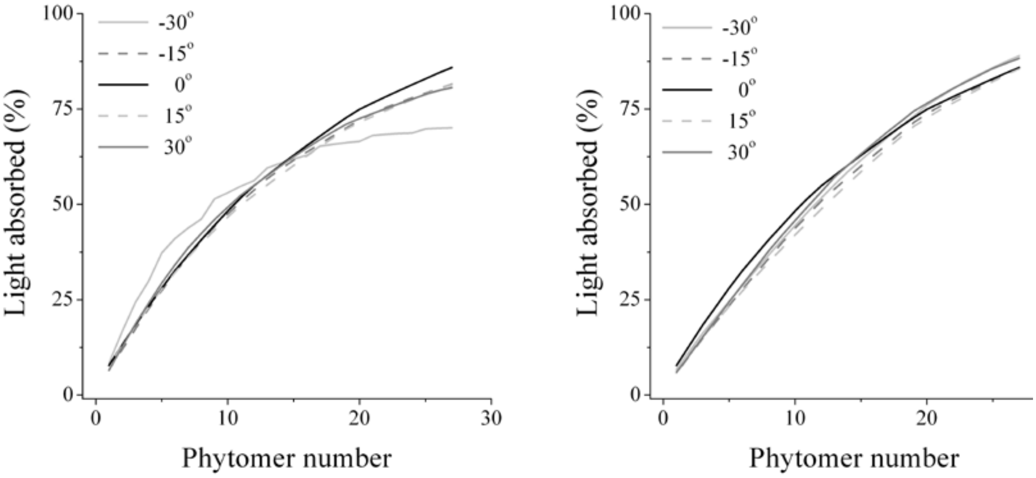

Fig. 2.3.3. Effect of leaf elevation angle on cumulated light absorption from top (phytomer 1) to bottom of the canopy under winter (left) and summer (right) light conditions. 0^o refers to the reference structure with an angle distribution ranging from 15^o (top leaves) to -23^o (bottom leaves).

summer light conditions (Fig. 2.3.3B), the increase was equal to 3% for ±30 ^{o} treatments. Interestingly in winter simulations, the decrease in elevation angle by 30 ^{o} did not only decrease light absorption but also changed the vertical distribution of light absorption leading to a higher absorption at the top part of the canopy (Fig. 2.3.3).

In general a change in elevation angle decreased canopy photosynthesis by 6% to 7% in winter, which is similar to the reduction in light absorption (Table 1). However, when the angle was decreased by 30 ^{o} canopy photosynthesis decreased only by 11% while absorption was decreased by 18%.

Leaf phyllotaxis

Changes in phyllotactic angle hardly affected light absorption (data not shown) and canopy photosynthesis (Table 1).

Leaflet angle

An increase in leaflet angle from completely horizontal leaflets (0 ^{o}) to an angle of 22 ^{o} resulted in an increase in light absorption by 2% in winter (Fig. 2.3.4A) and 8% (Fig. 2.3.4B) in summer. The subsequent increase in canopy photosynthesis was 2% under winter and 8% under summer light conditions (Table 1). When all leaflet angles were 22 ° light absorption was 2% lower under winter and 4% under summer light conditions compared to the reference plants (angle decreasing from 35 ° for basal leaves to 0 ° for the terminal leaflet) (Fig. 2.3.4), while canopy photosynthesis exhibited the same increase for both seasonal conditions.

Leaf shape

The simulations showed that longer and narrower leaves (LW>1) increased cumulative light absorption of the canopy in comparison to shorter and wider leaves when leaf area was kept constant. LW ratio was positively correlated with light absorption (Fig. 2.3.5). Light absorption was decreased by 8% and 12%, respectively, in winter (Fig. 2.3.5A), and 14% and 23%, respectively, in summer for a leaf ratio decrease of 0.25 and 0.5 (Fig. 2.3.5B). An increase in the LW ratio to 1.25 of the reference value resulted in an increase in light absorption by 5% in winter and 4% in summer. When the ratio was 1.5 times as high as the reference ratio, light absorption

Table 2.3.1. Assimilation rate (g CH_2O m^{-2} d^{-1}) in response to leaf elevation angle, phyllotaxis, leaflet angle and arrangement, leaf length/ width ratio, internode length scenarios as well as the optimized architectures.

	Total canopy photosynthesis	
	Winter	**Summer**
Leaf elevation angle		
-30^o	4.7	40.5
-15^o	5	40.5
0 (reference structure)	5.3	42
$+15^o$	5	40.5
$+30^o$	4.9	41.7
Leaf azimuth angle		
110^o	5.2	42.3
135^o	5.3	41.8
160^o (reference structure)	5.3	42
180^o	5.1	41.7
Leaflet angle		
0^o	5.1	37.3
22^o	5.2	40.5
From 35^o to 0° (reference structure)	5.3	42
Leaflet arrangement		
Longer leaflet petioles	5.2	39.4
Same area leaflets	5	39.3
Fewer/bigger leaflets	4.9	41.8
reference structure	5.3	42
Leaf length/ width ratio		
0.5	4.7	34.3
0.75	4.8	39.8
1.02 (reference structure)	5.3	42
1.25	5.5	44.2
1.5	5.6	44.6
Internode length		
3 cm	4.3	37
5 cm	5	38.8
7 cm (reference structure)	5.3	42
10 cm	5.6	44.2
12 cm	5.7	44.6
Optimized canopy architecture		
Scenario A	5.6	45.2
Scenario B	5.9	46.7

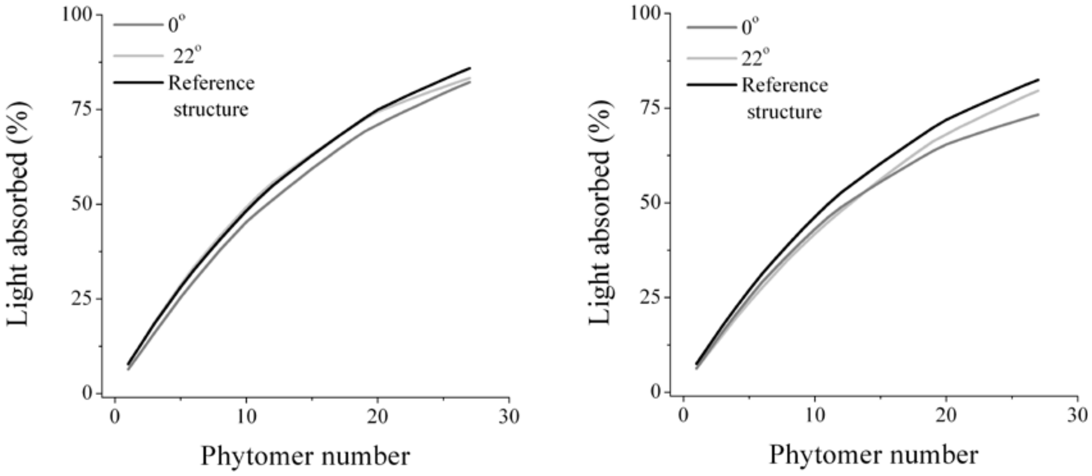

Fig. 2.3.4. Effect of leaflet angle on cumulated light absorption from top (phytomer 1) to bottom of the canopy under winter (left) and summer (right) conditions. In the reference structure the angle is decreasing from 35° for basal leaflets to 0° for the terminal leaflet.

was increased by 8% in winter and 10% in summer. An increase in LW ratio to 2 did not further increase light absorption (data not shown). The effect of the different treatments on canopy photosynthesis followed the trends observed in light absorption but the total increase was 1- 2% smaller compared to the increase in light absorption (Table 1).

Leaflet arrangement

Changing the arrangement of leaflets within a leaf while keeping leaf area constant (Fig. 2.3.2) affected the vertical light distribution as well as the total light absorption. The reference structure showed the maximum absorption under both summer and winter conditions (Fig. 2.3.6A and 2.3.6B). Plants with the reference leaves, i.e. exhibiting irregular leaflet size, absorbed more light in the top portion of the canopy (0.5 – 1 m) than leaves with equally sized leaflets. Leaves with fewer but bigger leaflets absorbed more light than leaves with more but smaller leaflets (Fig. 2.3.6). An increase in petiole length slightly diminished both light absorption and

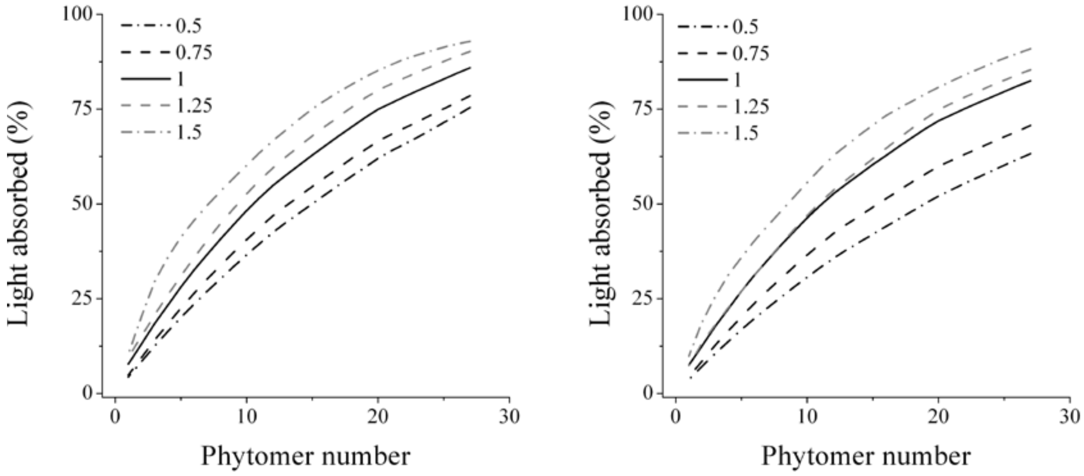

Fig. 2.3.5. Effect of leaf shape on cumulated light absorption from top (phytomer 1) to bottom of the canopy under winter (left) and summer (right) conditions. Leaf shape is referring to length-to-width ratio with reference structure having a ratio of 1.

canopy photosynthesis in both seasonal conditions. The effect of the difference in vertical light distribution between the scenarios was reflected on canopy photosynthesis where the leaf with leaflets of the same area exhibited 2% less canopy photosynthesis in winter and 6% in summer than the other two scenarios (Table 1).

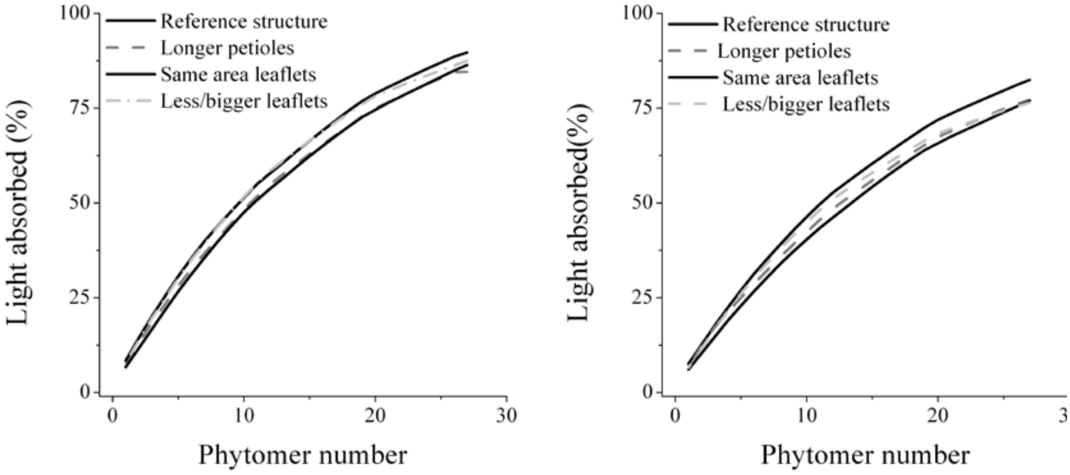

Fig. 2.3.6. Effect of leaflet arrangement on cumulated light absorption from top (phytomer 1) to bottom of the canopy under winter (left) and summer (right) light conditions.

Internode length

Light absorption and canopy photosynthesis were strongly affected by internode length. A decrease in the average internode length from 7 to 3 cm reduced cumulative light absorption by 14% in winter and by 12% in summer. An increase in internode length from 7 cm to 10 cm resulted in an increase of light absorbed of 4% in winter and 5% in summer, while a further increase in internode lengths to 12 cm resulted in a further increase in light absorption by 2% for each date. With respect to canopy photosynthesis, the increase in internode length to 10 cm increased canopy photosynthesis by 5 to 6% in summer and winter (Table 1). An increase to 12 cm increased photosynthesis by 8 and 6% for winter and summer, respectively.

Ideotyping scenarios

According to the above assessment of architectural characteristics of the tomato plant two ideotyping scenarios were designed. In scenario A, internode length was set to 10 cm, LW ratio to 1.5, while all other architectural characteristics were kept the same as in the reference structure. In scenario B, plant structure consisted of the same long internodes (10 cm) and a leaflet arrangement with long leaf petioles was chosen, while all other architectural characters remained the same as in the reference structure. Scenario A improved both light absorption (data not shown) and canopy photosynthesis by 6% (Table 1) in winter and by 8% in summer, respectively. In scenario B, the construction of a more open structure of the canopy resulted in an increase in light absorption of 11% in both winter and summer, with a similar increase in canopy photosynthesis.

Discussion

From the result of this work, we can conclude that both light absorbed and the vertical distribution of light in the canopy are very important for crop photosynthesis. Analysing the effects of vertical light distribution showed that these effects strongly differ between winter and summer light conditions. Deep penetration of light into the canopy has positive effects in summer, while these effects are negative in winter. Under summer light conditions photosynthesis of the upper leaves may be close to saturation, which explains the positive effects of deep penetration of light. This is in contrast to the photosynthesis rate of lower leaves, which remains unsaturated. Under winter light conditions, photosynthesis of the upper leaves is far from saturation. With the same light intensity at leaf level, upper leaves have a higher rate of photosynthesis than lower leaves (e.g. Sarlikioti *et al.,* 2011). This effect dominates in winter, resulting in the lower crop photosynthesis when the light penetrates deeply into the canopy.

Leaf elevation angles are maybe among the best investigated traits. It has been shown that a change in the elevation angles significantly influences light captured in different environments (Niinemets and Fleck, 2002, Valladares and Pearcy, 1998). In our previous study (Sarlikioti *et al.,* 2011) we showed that changes in leaf angles could have a substantial effect on both light absorption and photosynthesis. In the current study optimal results were achieved when leaf elevation angle distribution ranged between 15 ^{o} (top) and -23 ^{o} (bottom), indicating that during the cultivation period the plant orientates its leaves in such a way as to maximize light absorption and therefore photosynthesis. Deviations from that range failed to distinctly increase both light absorption and photosynthesis, though the vertical light distribution was affected.

Modifying the phyllotactic angle resulted in no improvement of light absorption and photosynthesis. These findings agree with those of Niklas (1998) who reported that phyllotactic angle in a crop has no effect on light absorption or photosynthesis. In contrast to this, Zotz *et al.* (2002) reported that change of leaf phyllotaxis to a golden angle of 137.5 ^{o} significantly improved the light capture efficiency in an epiphytic plant. Nevertheless, phyllotactic angle is very dependent on light competition. Although a change in the angle as such might have no direct effect on light absorption and photosynthesis, it might be followed by changes in other architectural characteristics that may lead to an increase in plant efficiency with respect to light absorption and photosynthesis.

In our study we found that an increase in LW ratio of leaves by 0.5 boosted light absorption by 8% in winter and 10% in summer. In environments where light is not an inhibiting parameter (e.g. because of an excess in radiation), structures that avoid mutual shading of the leaf components as well as the shading between neighbouring leaves can be advantageous for optimization of light absorption. A high leaf LW ratio has been reported to have a positive effect on light capture and crop photosynthesis in many species (Falster and Westoby, 2003). Other aspects of morphology of composite leaves such as shape and number of leaflets have, to our knowledge, hardly been investigated. In forest species it has been shown that the space and degree of overlap of the leaves in leaf clusters on the same branch has a significant effect on light capture (Planchais and Sinoquet, 1998).

Takenaka *et al*. (1994) showed that in general an increase in internode length causes an increase in light absorption and photosynthetic efficiency in nature: in fact, hormonally mediated internode elongation is among the most important mechanisms of the so-called shade avoidance syndrome, a set of processes that enable a plant to avoid shading by neighbouring plants (Smith and Whitelam, 1997). Our results showed that plants with longer internodes exhibited higher light absorption and photosynthesis both in winter and summer.

The combination of different architectural parameters in the ideotype scenarios resulted in an increase in both light absorption and canopy photosynthesis. Both ideotype scenarios aimed at creating a more open structure with more light absorption. In both cases this combination of characteristics led to an increase in photosynthesis which in turn could potentially result in a yield increase. Modern developments in plant breeding can use this type of information to produce more efficient genotypes in terms of canopy photosynthesis. On the other hand plant architectural characteristics are very plastic and plants are usually dynamically adapting to their environment (Valladares *et al.*, 2007), thereby potentially eluding breeding efforts towards a light absorption ideotype. For example during the course of the day leaf movement has been observed (Kao and Forseth, 1992) or plants tend to readjust their position when facing an intense environmental factor such as, for example, shading (Kahlen et al., 2008). Thus, even if this type of static model can give us a good quantification of the effect of each parameter on light absorption and photosynthesis, the ensuing genotype has to be tested under actual cultivation conditions in order to verify the expectations.

In the simulation model photosynthetic parameters decreased from top to the bottom of the canopy. These parameters were not adjusted with the increase or decrease of light intensities in the middle and lower parts of the canopy. In other words no dynamic adaptation of these parameters to the new light profiles induced by the case studies was considered. In reality leaf photosynthetic potential, of course, adapts to the long term light conditions a leaf has been exposed to (Gonzáles-Real *et al*,. 2007), as well as the short term changes in light climate during the day (Schurr *et al*,. 2006). Experimental data describing these phenomena could help to improve the model calculations by taking into account these adaptations.

Conclusions

The importance of different plant architectural components for light absorption and photosynthesis was investigated in detail using a static virtual plant. Our simulations lead to the conclusion that the most important architectural traits with respect to the optimization of light absorption and photosynthesis are internode length and leaf shape. We also assessed the importance of vertical light distribution for canopy photosynthesis and showed that the advantage of a deeper penetration of light in the canopy depends on the season.

Chapter 3 Monitoring PAR interception and photosynthetic stress

Chapter 3.1

Crop Reflectance as a Tool for Online Monitoring Of LAI and PAR Interception in Two Different Greenhouse Crops

Published as:

Sarlikioti V., Meinen E., Marcelis L.F.M. 2011. Crop reflectance as a tool for the online monitoring of LAI and PAR interception in two different greenhouse Crops. Biosystems Engineering, 108: 113-120.

Abstract

The aim of this study was to explore ways of on-line monitoring of LAI and PAR interception of the canopy, in greenhouse conditions through reflectance measurements on the PAR part of the spectrum for tomato and sweet pepper. LAI and PAR interception were measured at the same moments as reflectance at six wavelengths in different plant developmental stages in greenhouses. Normalized Difference Vegetation Index (NDVI) was also calculated. Relationships between the measured parameters were established in experimental greenhouses and subsequently these were tested in commercial greenhouses. The best estimation for LAI and PAR interception was obtained from reflectance at 460nm for both tomato and sweet pepper. The goodness of the fit validated with data from the commercial greenhouses, was also tested in this study. The divergence of the results from the ones reported from field experiments can be traced on the special greenhouse environment where more sources of reflectance are added due to construction parts and a white plastic covered background. Thus this new approach of estimating LAI and PAR interception from 460 nm is promising and can play a role in the decision support systems of modern greenhouse management.

Introduction

Today's highly industrialized production of greenhouse crops in combination with the increasing demand for a more sustainable production are pointing at the necessity of decision support systems. Methods of online monitoring of the major crop properties should be established. Leaf area is one of the most important crop properties which have a strong physiological impact on plant functioning namely PAR interception and thus photosynthesis. Unfortunately current models of leaf area index (LAI) development in use are not sufficiently correct and may lead to errors in calculating light interception (Jonckheere *et al.*, 2004). PAR interception can currently be measured manually. A light stick is used to take measurements on the top and bottom of the canopy. In order to gain a reliable estimation of PAR interception many measurements at different canopy spots are needed. This method can therefore be applied only for experimental purposes and not for commercial application as it is rather time consuming. Therefore the existence of an online monitoring technique would improve the efficiency of the measurement.

Optical remote sensing has been used in the past for the online monitoring of parameters such as chlorophyll and nitrogen content, plant stress etc. in field grown crops. Attempts to measure LAI and PAR interception with the use of remote sensing have been successful in field crops. Different approaches have been used for the data collection, with sensors either on low range (ground) or far range (low flights or satellites) (Bouman *et al.*, 1992; Clevers, 1997). Efforts in the field to use reflectance measurements for the estimation of the canopy nitrogen, biomass and photosynthesis in wheat (Gitelson *et al.*, 2002) potato (Jongschaap, 2006), sugar beets (Clevers, 1997), miscanthous (Vargas et. al, 2002) and rice (Tian *et al.*, 2005), were successful.

Plant canopy exhibits strong absorption in the red part of the spectrum (around 670nm) where reflectance is less than 3-5% (Gitelson, 2002). Low reflectance in this part of the spectrum is strongly related to high light absorption from chlorophyll and secondary photosynthetic pigments. On the other hand in the near-infrared part of the spectrum about 50% of the light is reflected, 45% is transmitted and only a small part is absorbed Turner *et al.*, 1999). The steep difference of canopy reflectance between red and near-infrared enables the distinction of vegetation from background materials (Bouman *et al.*, 1992).

Many studies aimed at determining combinations of reflectance of different wavelengths for correcting the effect of disturbing factors (such as old leaves and soil background) on the relationship between crop reflectance and crop characteristics such as LAI. These combinations of the reflectance at different wavelength bands are known as vegetation indices (VI's). Vegetation indices are quantitative measures that are used in an attempt to measure vegetation abundance or vigor (Wang *et al.*, 2005).

Some of the vegetation indices proposed during the years are the Weighted Difference Vegetation Index (WDVI) (Bouman *et al.*, 1992), the infrared/ red (IR/R) ratio and the red edge leaf effect, with the most commonly used the Normal Difference Vegetation Index (NDVI) (Wang *et al.*, 2005). NDVI is based on ratios of red (R) to near-infrared (NIR) reflectance where NDVI= (NIR−R)/(NIR+R). R and NIR ratio-based indices are strongly rooted in the contrasting response of R and NIR reflectance to increases in LAI (Chen *et al.*, 2002).

Though remote sensing has been applied quite successfully in the open field, it has not yet been tested in protected cultivations. Open-field methods cannot directly be transported in greenhouses due to complicating conditions such as existence of greenhouse structure and ground covering with white plastic. So research has yet to be done to test, under which conditions remote sensing can be applied for plant monitoring inside a greenhouse. The aim of this paper is to explore an accurate way for an on-line estimation of LAI and PAR interception based on reflectance measurements in two greenhouse grown crops (tomato and sweet pepper).

Materials and methods

Experiments

Seven experiments were conducted with tomato (*Lycopersicon esculentum* L.; Exp 1-7) and two with sweet pepper (*Capsicum annuum* L; Exp 8-9) grown in greenhouses. Experiments were conducted in experimental greenhouses (Exp. 1- 4 for tomato and Exp 8 for sweet pepper) for developing relationships estimating LAI and PAR interception from canopy reflectance. These relationships were afterwards tested in commercial greenhouses (Exp 5-7 for tomato and Exp. 9 for sweet pepper). In Exp 1 plants were evenly distributed in the greenhouse (plants were placed in equal distances). In the other experiments plants were grown in rows. For an overview of the experiments see Table 3.1.1.

Table 3.1.1. Overview of set-up of all experiments, the type of the measurements taken and the plant age for the duration of the experiments. Experimental greenhouse refer to university facilities while commercial greenhouse refer to units managed by growers. LAI, PAR interception of the crop and crop reflectance were measured (+) unless otherwise indicated (-).

Exp No	Crop		Plant age (in days from planting)	PAR interception	Reflectance	LAI
1	tomato	Experimental experiment	0-117	+	+	-
2	tomato	Experimental greenhouse	0-117	+	+	-
3	tomato	Experimental greenhouse	0-126	+	+	-
4	tomato	Experimental greenhouse	0- 80	+	+	-
5	tomato	Commercial greenhouse	2-171	+	+	+
6	tomato	Commercial greenhouse	180-240	+	+	-
7	tomato	Commercial greenhouse	180-240	+	+	-
8	sweet pepper	Experimental greenhouse	58-73	+	+	+
9	sweet pepper	Commercial greenhouse	0-274	+	+	+

Measurements were taken every seven to fifteen days, depending on the weather conditions, from a week after planting till the end of the cultivation cycle (table 1). Diffuse radiation conditions were chosen in order to avoid measuring errors occurring with direct sun conditions, such as shadow stripes caused by the construction parts of the greenhouse, as well as low sun azimuth and insufficient cosine correction of the diffuser.

Reflectance measurements

For the reflectance measurements, Cropscan MSR87 (CropScan, Rochester, USA), sensor was used. The downward facing sensor was equipped with eight spectral bands, each covering approximately 20nm, centered at 460, 510, 560, 610, 660, 710, 760 and 810 nm. The data were recorded with a datalogger. The sensor was positioned parallel to the ground at 4m height. The viewing angle of 28 degrees resulted in a viewing area of 3 m^2. At each measuring date reflectance was measured in 8 independent plots of the crop canopy with 2 measurements per plot. Data were averaged per plot. The effect of the background reflectance (white plastic) was investigated with a small trial with new and old sheets of plastic as background. The difference on reflectance of the two treatments was a maximum of 10% when no crop was present and diminished as the LAI was increasing (data not shown).

PAR interception measurements

PAR interception was measured above and below the canopy in the same spots as reflectance. For each position eight successive readings were taken. For PAR interception measurements a SunScan Canopy Analysis System SS1-UM-1.05 (Delta-T. Cambridge, UK) was used. Sunscan sensor consists of a light sensitive stick that is 1 meter long, containing 64 photodiodes equally spaced along its length measuring incident PAR light. It was coupled with a Beam Fraction sensor (BFS) that measures total light as well as PAR light levels. The BFS sensor was mounted at the trellis on the top of the greenhouse measuring the incident light on the canopy at the same time with the measurements with the stick in the canopy.

PAR interception was calculated as follows:

PAR interception= 1-(PAR below the canopy/ PAR above the canopy) Eqn 3.1.1

In many crop models Lambert- Beer's law (Monsi and Saeki, 1953; Marcelis *et al.* 1998) is used for the simulation of PAR interception by the crop. It assumes uniform and infinite canopy of randomly distributed, absorbing leaves. The amount of photosynthetically active radiation intercepted (I) by a crop is calculated as:

$$I= (1-\rho)*I_0*(1-e^{-k*L})*100 \qquad \text{Eqn 3.1.2}$$

Where ρ stands for canopy reflection coefficient, I_0 is the radiation level at zero canopy depth, L the leaf area index of the canopy, and k is the light extinction coefficient of the crop. Lambert- Beer's law was applied on the data and the extinction coefficient was calculated:

$$k = \mathrm{Ln}(I/I_o)/\ LAI \qquad \text{Eqn 3.1.3}$$

Leaf Area Index measurements

Leaf Area Index was measured destructively. The leaves were sampled on the same dates as the reflectance and PAR interception measurements and their leaf area was measured with LI- COR 3100 meter (LICOR Inc. Lincoln, NE, USA). A minimum of 6 plants was used in each harvest when destructive measurements were possible. Yellowed and brown leaves were not measured.

Full spectrum measurements on leaf level

Reflectance and transmittance of the full spectrum between 380nm-1100nm of individual leaves of tomato and sweet pepper plants were measured with the use of InstaSpec IV CCD spectrometer (Oriel, Stradford, CT, USA**)** and LiCor 1800-12 (LICOR Inc. Lincoln, NE, USA) integrating Sphere equipment. The measurements were performed on young and old leaves in a non- destructive way. Eight leaves per category were measured for each plant. The measurements were repeated after two weeks on the same leaves in order to investigate the spectral differences in time as well as in canopy depth. Data were then used to calculate the crop extinction coefficient:

$$k = k_{bl}\sqrt{(1-\sigma)} \qquad \text{Eqn 3.1.4}$$

Where k_{bl} (0.84) is the extinction coefficient for a crop with spherical leaf elevation angle distribution when the crop is composed of black leaves. and σ is the scattering coefficient. The wavelength dependent scattering coefficient σ equals the sum of reflection and transmission and was derived from the spectrometer measurements per wavelength at the leaf level. Consequently, Eqn 3.1.4 scales up this leaf property to crop level. The above methods were derived from and elaborated upon in detail by Goudriaan and Van Laar (1994). The k values calculated from Eqn 3.1.4 where then compared with the k-values derived from Eqn 3.1.3.

Vegetation Indices (VI's).

Normal Difference Vegetation Index (NDVI) was calculated from the available reflectance data. NDVI was calculated as (Wang *et al.*, 2005):

$$NDVI= (R_{660}-R_{810})/(R_{660}+R_{810}) \qquad \text{Eqn 3.1.5}$$

where R_{660} and R_{810} is the reflectance at 660nm and 810nm, respectively. NDVI was tested against single wavelength reflectance measurements.

Statistical Analysis

For the statistical analysis of the results the GenStat 9th edition software package was used. Regression analysis was applied to the data and linear, polynomial and exponential models were tested in terms of the best fit on the basis of the adjusted R^2 and the standardized residuals ($p<0.05$).

Results

The natural log of I/Io showed a linear relationship with LAI in both tomato and sweet pepper, following the Lambert- Beer law (Eqn 3.1.3, Fig 3.1.1). The extinction coefficient derived from these data, was 0.81 for sweet pepper and 0.68 tomato. Calculation of k from optical properties of individual leaves, resulted to a k factor of 0.8 for sweet pepper and of 0.7 for tomato respectively (Eqn 3.1.4). The higher extinction coefficient of the sweet pepper crop compared to tomato implies structural differences between the two species that affect the way incident light is intercepted.

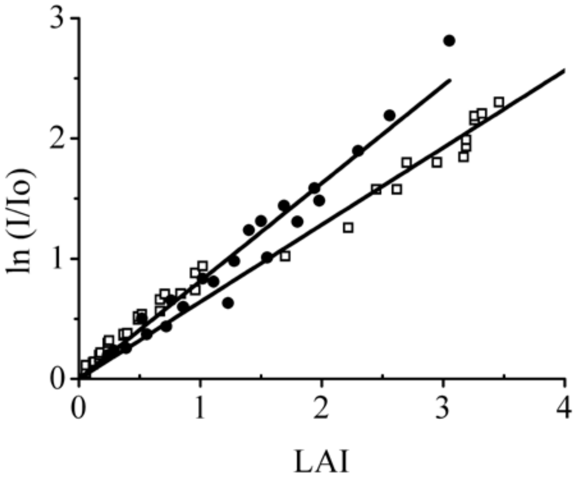

Fig. 3.1.1. Relation between leaf area index and fraction of light intercepted (I/Io) for tomato (□) and sweet pepper (●) (1A). The slope of the line represents the extinction coefficient k. Data are from experiment 1-7 for tomato and 8-9 for sweet pepper.

Single tomato leaves have higher reflectance (6.1%) than sweet pepper (1.8%) for PAR (400 nm to 700 nm) (Fig 3.1.2A and 3.1.2B). The difference is especially high between 510 nm and 660 nm (average reflectance 7.4% and 2.5% for tomato and sweet pepper respectively) that correspond to the green part of the spectrum and beginning of the red part (Fig 3.1.2A and 3.1.2B). Leaf spectrum for tomato and sweet pepper remains almost constant in the area of five out of eight bands measured (460, 610, 660, 760 and 810 nm) by cropscan for measuring crop reflectance. This spectrum consistency enables the use of wider spectrum bands for the calculation of Vegetation indices in general and NDVI in particular.

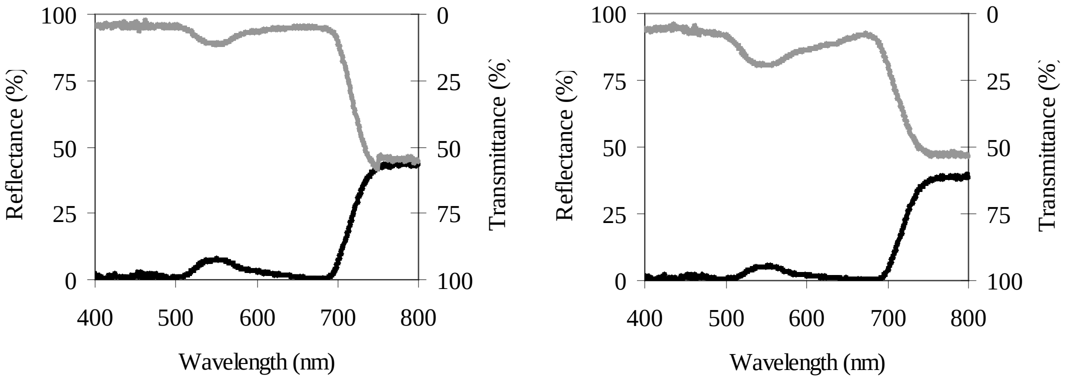

Fig. 3.1.2. Transmittance (black line) and reflectance (gray line) of individual leaves of tomato (A) and sweet pepper (B). Data are from experiment 1 and 8 respectively.

The relationship of LAI and PAR interception with canopy reflectance, was investigated for six wavebands as well as calculated NDVI (Eqn. 3.1.5; table 3.1.2) on tomato (Exp 1) and sweet pepper (Exp 8). The exponential regression model had the best fit for all wavelengths. For tomato crop, PAR interception correlates well with reflectance at three different wavelengths (460nm, 510nm and 560nm) with a slightly better fit at 460nm. The same was observed for LAI. Reflectance at 460nm gave the best estimation for both LAI and PAR interception also for sweet pepper. The use of NDVI did not improve the estimation of PAR interception and LAI for any of the two crops.

Table 3.1.2. Correlation of PAR interception by the crop and LAI (y-values) with crop reflectance at different wavelengths and Normal Difference Vegetation Index (NDVI) (x-values) in tomato and sweet pepper. Fitted function was $y= a+b*e^{-kx}$. The R^2 and Root Mean Square Error (RMSE) are presented. No of observations was 35 for tomato (Exp. 1) and 26 for sweet pepper (Exp. 8).

	Tomato				Sweet pepper			
Wavelengths	PAR Interception (%)		LAI		PAR Interception (%)		LAI	
	R^2	RMSE	R^2	RMSE	R^2	RMSE	R^2	RMSE
460	97.8	4.55	95.4	0.29	96.6	7.47	88.4	0.36
510	97.6	4.74	94.7	0.32	95	5.48	85.7	0.39
560	97.7	4.70	94.3	0.45	94.6	5.68	83.3	0.42
610	85.4	11.8	86.8	0.68	80.9	10.7	61.8	0.64
660	95.4	6.63	92.4	0.60	88.6	8.46	74.5	0.54
710	93.2	8.01	90.1	0.79	64.9	14.5	45.3	0.77
NDVI	87.3	11	85.6	0.52	89.4	8.15	79	0.49

During plant growth LAI increases, resulting in an increase of PAR interception of the crop. A decrease in the amount of reflected light exponentially decreased with the increase of LAI and PAR interception for both crops (Fig 3.1.3). Tomato crop reflects more when LAI is small (<0.5) than sweet pepper.

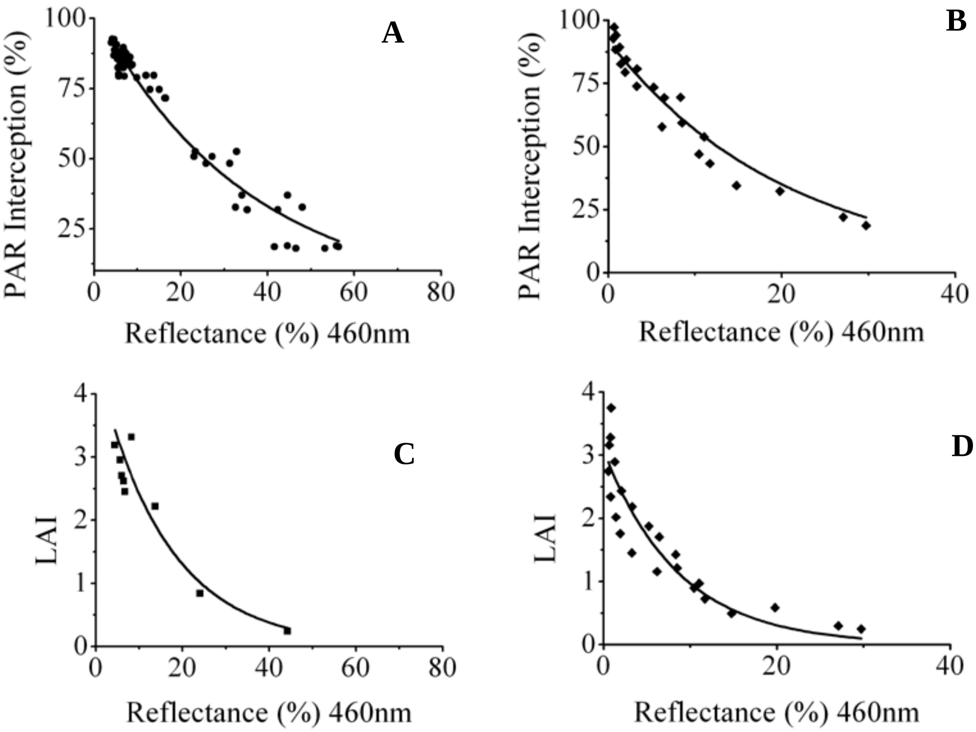

Fig 3.1.3. Correlation of reflectance at 460 nm with PAR interception (A, B) and LAI (C, D) for tomato (A, C) and sweet pepper plants (B, D) in experimental trials. Data points are from experiment 1-4 for tomato and experiment 8 for sweet pepper.

The relationships established from the first trials were then tested for data gathered from commercial greenhouses. The diversity of the cultivation practices of the greenhouses on which those data were taken was a good test for the robustness of the relationship of reflectance at 460nm with LAI and PAR interception (Fig. 3.1.4). For tomato the fit of the established relationships was good for PAR interception (R^2= 95.2, se=6.35) and LAI (R^2= 89.4, se=0.51). For sweet pepper the fitting gave reasonable results for both PAR interception (R^2= 89.4, se=0.51) and LAI (R^2= 79.8, se=0.76).

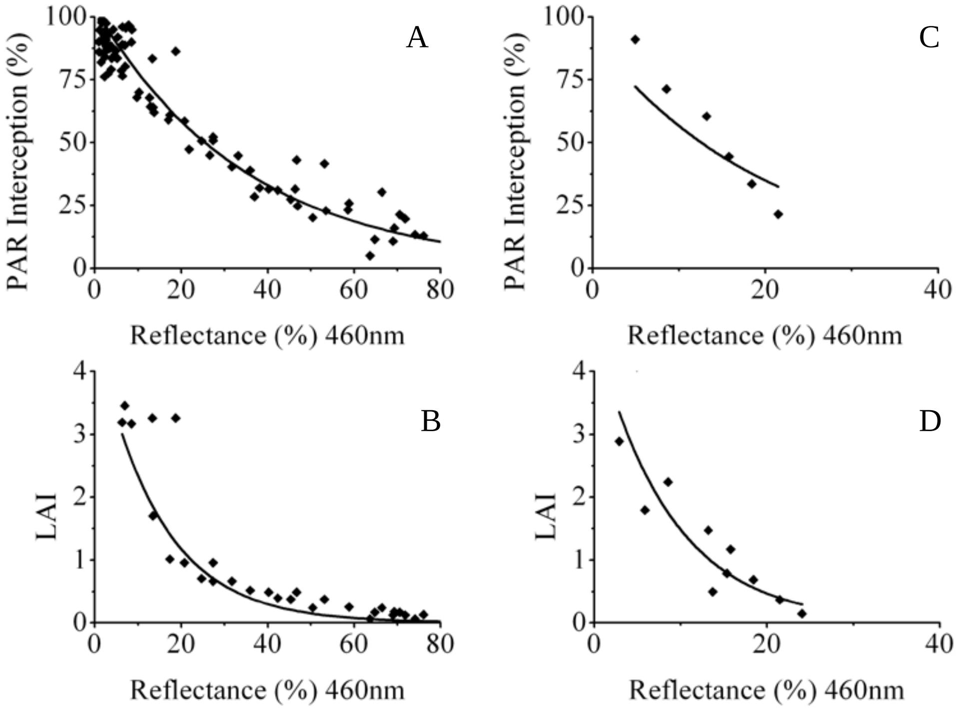

Fig 3.1.4. Validation of the relationship between reflectance at 460 nm with PAR interception and LAI for tomato (Fig 4A and 4C respectively) and sweet pepper (Fig 4B and 4D respectively) plants in commercial greenhouses. The lines are from Fig 3. Based on experimental greenhouses experiments. Data points are from experiment 5-7 for tomato and experiment 9 for sweet pepper

Discussion

This study explores an accurate way for the online estimation of LAI and PAR interception of the crop through remote reflectance measurements as well as the robustness of such a method for tomato and sweet pepper grown in a greenhouse. For that reason six different wavelengths covering an area of the spectrum from blue to red and NDVI were tested. Previous studies performed with field crops showed a correlation of PAR interception and LAI with reflectance in the R/NIR region of the spectrum (Bouman, 1994, Clevers, 1997). In our study nevertheless the **B** best correlation was achieved at 460 **A** or both tomato and sweet pepper.

The difference between field application results and the ones reported here can mainly be explained from the added reflectance inside the greenhouse from the various metallic parts and especially the white plastic that covers the floor. These special characteristics of the greenhouse production system ask for a new approach on remote monitoring than the ones already used in the open-field and is proposed in this study. Relationships established in this study were further validated with data from commercial greenhouses. The relationship between reflectance, PAR interception and LAI was best described by an inverse exponential curve similar to the types of curves used in field experiments of potato (Bouman, 1994) and miscanthus (Varga *et al.*, 2002) for LAI monitoring. In this study the relationships between LAI, PAR interception and crop reflectance were established under diffuse light conditions. Corrections for the bidirectional reflectance can be used for sensor corrections for saturation problems of the sensor under direct light conditions. This method can be successfully applied to the field for remote sensing techniques. In the particular case of the greenhouse measurements under direct light conditions might be highly variable by shadows caused by the construction parts of the greenhouse and reflectance from the glass.

Light in plants is highly absorbed in the red and a little less in the blue part of the spectrum. It is expected that reflectance in these particular regions will increase with the growth as usually more LAI indicates more light interception and less light reflectance. The 460 nm wavelength is located on the blue part of the spectrum partly explaining the good correlation between the data. Reflectance at 460 nm was high (~65%) in an empty greenhouse because of the reflectance of the construction parts and the white plastic on the floor (data not shown). The difference between crop and background reflectance, makes it easier to monitor changes in the leaf area of the plant inside the greenhouse. Also the lack of differences between plastic sheets in different stages of wear, provide an additional robustness to the estimation method presented on this paper. Finally around 460 nm reflectance hardly changes with the wavelength, diminishing any errors arising from the sensor.

After both crops reach a LAI of about 3 and PAR interception of about 90%, variation is high and an accurate estimation of LAI and therefore PAR interception from reflectance is no longer possible. This variation is mainly due to the fact that in the visible region of the spectrum only the reflectance of the upper leaf layers determines the contribution of the canopy to the total measured reflectance. So if LAI is higher than 3 not much more light is intercepted and therefore reflectance changes are no longer perceived. When LAI is higher than three, 90% of PAR is already intercepted by the canopy and further LAI increase has only a small impact on PAR interception. In this context an accurate knowledge of LAI in the early developmental stages is more important than in later stages.

Increasing LAI lead to increasing PAR interception and following Lambert- Beer law. The k value calculated for the extinction coefficient for tomato was similar to the values reported in the literature (Atheron and Rudich, 1986). Sweet pepper seems to have a little higher light interception for the same LAI as tomato which is in accordance with the fact that it also shows lower reflectance both in leaf and canopy level than tomato plants. For sweet pepper a k factor varying from 0.6 to 1.45 have been reported in the literature (Nederhoff and Vegter, 1994). The k factor of 0.81 that was calculated from our data is within this range. Extinction coefficient is an important input parameter in most crop models. Most models assume that extinction coefficient is constant and the value used as an input is either calculated by experimental data or from literature. Because the extinction coefficient depends on canopy structure (for example leaf elevation angle) its value can change with the growth of the plant. With our method of estimating LAI and PAR interception, it is possible to calculate extinction coefficient from the start of the cultivation until the crop is fully grown.

The normalized difference vegetation index (NDVI) is widely used in field applications for the estimation of LAI. NDVI is based on plant physiological processes and particularly in the difference between leaf absorption of red light and non- absorbance of the NIR light. Despite the wide use of this index in the open fields (Wang *et al.*, 2002; Hoffmann and Blomberg, 2004; Elwadie *et al.*, 2005), single waveband at 460nm performed better in our case in estimating PAR interception or LAI in greenhouse conditions. The decreased accuracy of NDVI inside the greenhouse is probably a result of the added reflectance from the construction parts of the greenhouse as well as the background cover in all part of the spectrum and especially the red.

Conclusions

Reflectance measurements offer a non- destructive way to estimate PAR interception and LAI (up to the value of 3) in greenhouse production systems. The relationship established from the current work between reflectance at 460 nm, PAR interception and LAI for both tomato and sweet pepper, can become a good tool for crop online monitoring in greenhouse conditions. Furthermore if information from reflectance sensors are inputted directly into crop models, new opportunities for decision support in greenhouse production could be opened

Chapter 3.2

Photochemical reflectance index as a mean of monitoring early photosynthetic stress

Published as:

Sarlikioti V., Driever S.M., Marcelis L.F.M. 2010. Photochemical reflectance index as a mean of monitoring early water stress. Annals of Applied Biology, 157: 81-89.

Abstract

Water stress in plants affects a number of physiological processes such as photosynthetic rate, stomatal conductance as well as the operating efficiency of PSII and non- photochemical quenching. Photochemical Reflectance Index (PRI) is reported to be sensitive to changes of xanthophyll cycle that occur during stress and could possibly be used to monitor changes in the parameters mentioned before. Therefore the aim of this study was to evaluate the use of PRI as an early water stress indicator. Water stress treatment was imposed in a greenhouse tomato crop. CO_2 assimilation, stomatal conductance, light and dark adapted fluorescence as well as PRI and relative water content of the rooting medium $RWC_s\%$ where repeatedly measured. The same measurements were also performed on well-irrigated plants that acted as a reference. The experiment was repeated in four consecutive weeks. Results showed a strong correlation between $RWC_s\%$ and photosynthetic rate, stomatal conductance, non- photochemical quenching and operating efficiency of PSII but not with PRI when the whole dataset was considered. Nevertheless more detailed analysis revealed that PRI gave a good correlation when light levels where above 700µmol m^{-2} s^{-1}. Therefore, the use of PRI as a water stress indicator cannot be independent of the ambient light conditions.

Introduction

Plant water status is an important aspect of crop management in greenhouse cultivation. Water stress affects plants water regulation, leading to changes in physiological processes. Thornley and Johnson (1990) noted that water stress often reduces plant growth by reducing leaf area development and increasing carbohydrate partitioning to the roots. According to Foolad *et al.* (2003) when water stress was imposed during the reproductive stages of tomato plants, its effects on these processes were found to be dependent on the degree of stress. Moderate water stress imposed just before flowering tended to accelerate flowering and fruit setting, but high and severe water stress retarded flowering and fruit setting, and reduced the number of flowers and fruits set per truss. Nowadays, production of tomato fruits in greenhouses is a highly intensive agricultural industry, where the cost profit limit is marginal and efficient use of resources is vital. The use of fresh water, one of the most important resources for this industry, has been predicted to become more scarce in the future (Hsiao *et al.*, 2007). Improving the efficiency of water use for irrigation is therefore one of the key challenges for tomato growers. Although much progress has been made on optimizing water supply based on greenhouse climate data (Marcelis *et al.*, 2006), only few methods use direct plant-based physiological indicators to detect the occurrence of water stress. An advantage of a reliable plant-based indicator over model predictions based on greenhouse climate is that, when stress can be detected in an early stage, water supply could be optimized.

Water stress may occur when the water supply to the plant is not meeting its transpiration demands. Stomata opening is directly affected by water stress (Cornic, 2000, Lawlor and Tezara, 2009). When stomata are not sufficiently opened, photosynthetic rate starts to decrease as well as the quantum yield of photosystem II (Chaves *et al.*, 2002). As a result of a decreased rate of CO_2 assimilation, light energy absorbed by the leaf cannot be used to drive photosynthetic electron transport and a part of this energy is dissipated as heat increasing the non-photochemical quenching (Krause and Weis, 1991; Baker and Rosenqvist, 2004). Researchers have reported changes in chlorophyll fluorescence corresponding to water stress (Baker and Ort, 1992; Lichtenthaler and Babani, 2000). Changes in plant physiological processes due to water stress have also been linked to changes in plant reflectance in specific wavelengths (Pinter *et al.*, 2003).

Several studies have attempted to quantify water stress through remote sensing (Bowman 1989; Peñuelas *et al.* 1994). Spectral indices have been used to determine real-time crop coefficients to improve irrigation scheduling (Bausch 1995). The relationship between reflectance spectra and leaf water status have been studied in numerous crop species as well as the possibility of estimating relative leaf water content by reflectance in the range of the near infrared (Danson *et al.*, 1992; Danson and Aldakheel 2000). Peñuelas *et al.* (1993) have found that reflectance at 970 nm can be a useful water status indicator but only when the stress is already well developed. Reflectance at 1,400 and 1,900 nm has been shown to correspond directly to water content in plant tissue (Peñuelas *et al.* 1997) as well as physical-based studies in short wave infrared (1,400– 2,500 nm) (Ceccato *et al.*, 2001). Graeff and Claupein (2007) showed, that reflectance in the 510_{780}, 540_{780}, $490_{1,300}$, and $540_{1,300}$ nm wavelength ranges can also become an indicator of plant water status (the values are for the combination of two different reflectance filters used in the analysis). However, infrared reflected radiation is greatly affected by plant architecture, crop density and leaf structure, thus increasing the uncertainty of estimation (Elachi, 1987).

In this paper we define plant water stress as the loss of plant photosynthetic activity induced by water deprivation. The so-called photochemical reflectance index (PRI = (R531 − R570)/ (R531 + R570)), where R531 and R570 are reflectance signals at 531 and 570 nm respectively, has been used to monitor dynamic changes in photosynthesis in water stressed plants (Evain *et al.* 2004). PRI provides a quick and non-destructive assessment on photosynthesis related physiological properties of the leaf and canopy (Peñuelas *et al.* 1994; Evain *et al.* 2004; Weng *et al.* 2006) for a wide range of species (Gamon *et al.* 1997; Guo and Trotter 2004). Changes in absorbance and reflectance values around 531–535 nm as a result of nutrient stress, have been related to ΔpH-mediated chloroplast shrinkage and to changes in the aggregation state of antenna pigment–protein complexes mediated by an accumulation of de-epoxidized forms of the xanthophyll cycle molecules (Morales *et al.* 1990; Ruban *et al.* 1993).Since we defined water stress as the loss of plant photosynthetic activity induced by water deprivation, it comes to reason that PRI could be used for water stress monitoring. The epoxidation and de-epoxidation of the xanthophyll cycle is also strongly related to the light environment (Adams *et al.*, 1992, Lawlor and Tezara, 2009). Although its applicability in detecting plant water stress has been tested in crops grown under conditions of severe stress it is yet to be tested for early stages of water deprivation. Therefore the aim of this paper is to evaluate PRI as an indicator of early water stress.

Materials and methods

Plant materials and water stress imposition

Tomato plants (*Solanum lycopersicon* cv Aranca) were grown on rockwool slabs (at field capacity contains 80% solution, 15% air pore space and 5% rockwool fibers) in a glasshouse in Bleiswijk, the Netherlands (52^o 1'N). Two week old plants were planted on the rockwool slabs, with three plants per slab on 16 June. After plants reached a height of 1.8m (17 July), measurements commenced. Each plant was irrigated by one dripper. Plants were watered automatically (water flow 4.5 l/h) with a nutrient solution [major nutrients: K 8.0, Ca 10.0, Mg 4.5, NO_3 23.0, SO_4 6.8, H_2PO_4 1.25 (mM); minor elements: Fe 25, Mn 7, Zn 7, B 50, Cu 0.7, Mo 0.5 (μM)] with a pH 5.5 and an EC of 4 ms/cm. At the start of each experiment, stress treatment was applied by withholding water through removal of drippers from a rockwool slab with three plants (hereafter referred to as stressed plants). Leaf measurements on these three stressed plants and three normally irrigated plants (hereafter referred to as reference plants) were performed on the first young fully developed leaf in full exposure to the sun for three consecutive days after the dripper removal. The experiment was repeated in four consecutive weeks. In each repetition, drought stress treatment was applied on a different slab with three new stressed plants and a different slab was used as reference plants. Measurements were performed every day in all plants of each treatment. Each day measurements were taken every half hour or every hour from 9:00 till 15:00.

Growth conditions

Incident light at leaf level was measured every 5 minutes by LI-190 quantum sensors (Licor, Inc., Lincoln, NE, USA.) positioned at the same height as the measured leaf. The relative water content (RWC_s%) in the rooting medium (rockwool slab) was monitored by WET sensors (Delta-T, UK) positioned vertically in the slab. Five WET sensors per slab were used both in reference and stress treatment.

Inside greenhouse temperature as well as the vapour pressure deficit (VPD) where also monitored, every 5 min during the experiment (table 3.2.1).

Table 3.2.1. Climate conditions in the greenhouse. Average values ±s.e of incident PAR, the inside greenhouse temperature (Tair$_{in}$) and VPD are presented for each repetition. The repetitions were performed in 4 consecutive weeks. Each repetition lasted 3 days.

Repetition	*Day*	*PAR* *(µmol m^{-2} s^{-1})*	*Tair$_{in}$* *(C^o)*	*VPD* *(kPa)*
	1	222.1 ±41.79	22.4 ±0.5	0.34 ±0.17
Rep 1	2	348.6 ±45.5	22.4 ±0.5	0.54 ±0.2
	3	128.82 ±16.69	21.2 ±0.12	0.27 ±0.03
	1	764.1 ±19.33	21.5 ±0.03	1.14 ±0.08
Rep 2	2	566.3 ±28.66	21.5 ±0.04	0.07 ±0.04
	3	652.5 ±28.01	23.5 ±0.03	0.09 ±0.17
	1	556.6 ±26.96	25.9 ±0.06	0.1 ±0.05
Rep 3	2	609.6 ±33.41	25.9 ±0.08	0.08 ±0.02
	3	199.6 ±37.82	25.6 ±0.09	0.23 ±0.11
	1	424.6 ±57.57	19.9 ±0.04	0.11 ±0.20
Rep 4	2	408.8 ±85.97	19.9 ±0.04	0.24 ±0.30
	3	221.1 ±29.73	20.5 ±0.54	0.26 ±0.29

PRI

The photochemical reflectance index of a leaf was measured by a PlantPen PRI 200 instrument (Photon Systems Instruments Ltd, Czech Republic). The PlantPen was clipped to a lateral leaflet. Light from an internal dual wavelength light source at 531 and 570 nm was emitted to the leaf and reflectance was measured through a PIN photodiode with a 500 to 600 nm bandpass filter. Data of reflectance at 531 and 570 nm as well as the calculated PRI value was collected. Measurements were taken every half hour from three plants from the stress treatment and three reference plants. The instrument measures an area of 0.5 cm^2. The average of eight measurements from randomly chosen positions on each selected leaf were taken for a representative measurement of PRI.

CO_2 assimilation rate

The CO_2 assimilation rate was measured every half hour with the use of a portable open gas exchange measurement equipment (LCpro$^+$, ADC, UK). The lateral leaflet of the chosen leaf was clamped in the chamber (6.25 cm^2 leaf area). The measurement conditions were 800 ppm CO_2 and 700 μmol m^{-2} s^{-1} PAR with a vapour pressure deficit (VPD) of 6.35 kPa. Vapour pressure deficit and CO_2 concentrations were similar to those in the greenhouse environment. The light intensity given was the average value of ambient irradiance on a sunny day. Constant light was used in the measurement to ensure that light intensity was sufficiently high and allowed comparison between treatments. Measurements were taken every minute for 12 minutes and CO_2 assimilation rate was then calculated as the average of the last three measured rates (i.e. the measurements at 10, 11 and 12 minutes). The period of the 12 minutes was previously determined as the time needed for stabilization of stomatal conductance and CO_2 assimilation rate.

Stomatal conductance

Stomatal conductance of a leaf was measured with an AP4 porometer (Delta-T, UK). The set relative humidity was the same as the greenhouse condition. The porometer was calibrated every 3 hours or when changes in environmental conditions in the greenhouse prevented accurate measurements (temperature difference ±2^{o}C from calibration temperature). Stomatal conductance was measured every half an hour. Three measurements per leaf from three plants per treatment were taken to obtain a representative measurement.

Chlorophyll Fluorescence

Chlorophyll fluorescence parameters were measured every hour with a Dual-PAM-100 (Heinz Walz GmpH, Germany). A leaflet was clamped into the optical unit and was continuously illuminated with white actinic light at an irradiance of 700 μmol m^{-2} s^{-1} (same level as for CO_2 assimilation measurements). The steady-state fluorescence (F') was recorded after 6 minutes, before a saturation pulse was imposed to the leaf in order to determine the maximal fluorescence level in the light adapted state (F'_m). The leaf was then darkened for 25 min, thereafter minimal fluorescence (F_o) was measured. During dark adaptation, the modulated measuring light was sufficiently low (<0.1 μmol m^{-2} s-^{1}) not to induce any significantly variable fluorescence.

Maximal fluorescence (F_m) was measured after saturating light was applied for 0.8s at 3000 μmol m^{-2} s^{-1} to the already dark adapted leaf. These measurements were used to calculate the operating efficiency of PSII ($F_q'/F'_m = (F'_m - F')/F'_m$), the maximum quantum efficiency of PSII (Fv/Fm) as a relative measure of the maximum quantum yield of PSII primary photochemistry (Björkman and Demmig, 1987) and the non-photochemical quenching (NPQ, calculated as (F_m/F'_m) − 1) (Bilger and Björkman, 1990; Baker, 2008).

Statistical analysis

The data were analysed with Genstat 11 software. Regression analysis using linear and polynomial models were used to assess the relationship between RWC_s% and the other response variables. The significance of model terms was tested using the F-test at the $p = 0.05$ level of significance. The goodness of fit of models was considered using the adjusted (for the number of model terms) R^2.

Results

Environmental conditions in general and incident light intensity in particular varied greatly between repetitions (weeks), days of a week and during the course of a day during the experiment. The inside temperature varied as a day average from 20°C to 26°C, while VPD fluctuated from 0.07 to 1.14 kPa (table 3.2.1).

Water stress was induced gradually in tomato plants by withholding water (Fig. 3.2.1A). Although the way of withholding water was identical for all repetitions, the rate at which the relative water content of the rockwool slab (RWC_s%) decreased, differed. The final RWC_s% that was reached at the end of each repetition varied from 23% to 75%. This variation was due to variable transpiration as a result of the variation in climatic conditions Light levels varied on average from 128.8 μmol m^{-2} s^{-1} to 764.1 μmol m^{-2} s^{-1}(Table 1). Withholding water reduced CO_2 assimilation by up to 52%, while stomatal conductance was reduced by up to 70%. CO_2 assimilation (Fig. 3.2.1A) as well as stomatal conductance (Fig. 3.2.1B) showed a high and positive correlation with RWC_s%.

The operating efficiency of the PSII (F'_q/F'_m) was decreased up to 20% (the percentage is expressing the maximum difference between the lowest (0.45) and the maximum value (0.525 - 0.57)) in response to lowered RWC_s% for all repetitions

(Fig. 3.2.1C), while *NPQ* increased considerably with the decrease of RWC$_s$% (Fig. 3.2.1D), with an increase in the range of 100%-216% for the different repetitions.

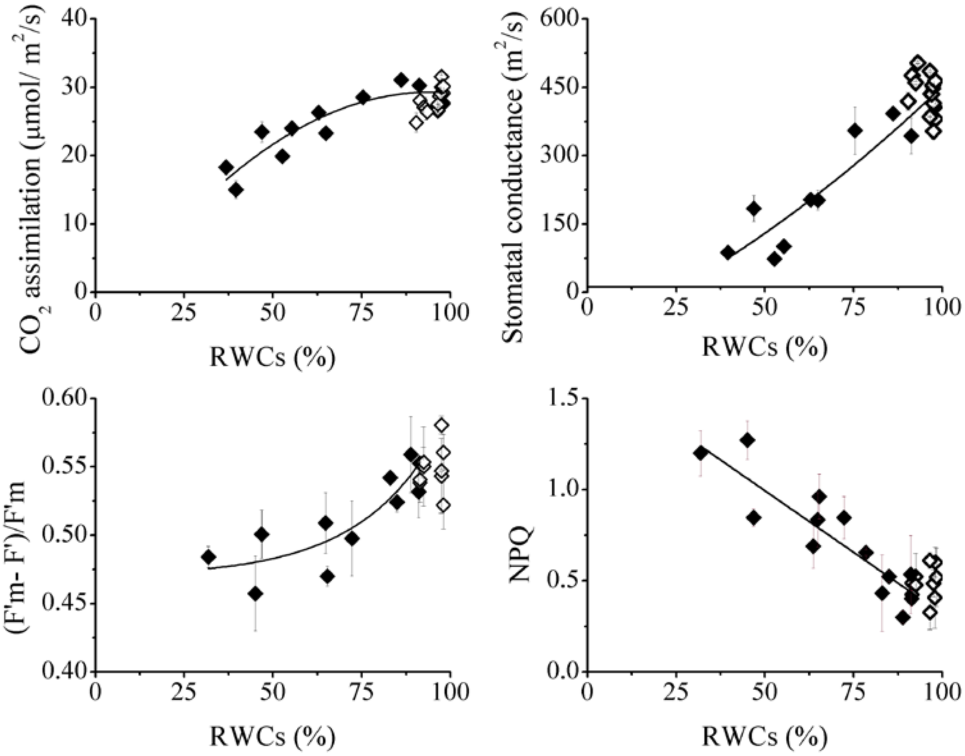

Fig 3.2.1. Correlation of CO_2 assimilation (s.e of intercept= 8.47, of B1= 0.26 and B2= 0.001 ,R^2= 0.79 ,s^2= 5.49 with 10 d.f), Stomatal conductance (s.e of intercept= 240.88, of B1= 7.1 and B2= 0.049 ,R^2= 0.86 ,s^2= 2691.9 with 9 d.f), Quantum yield of PSII efficiency (F'm- F')/ F'm) (s.e of intercept= 12.4 and of B= 0.00104, R^2= 0.71 ,s^2= 3.61 with 8 d.f), and NPQ (s.e of intercept= 0.0826, of B1= and B2= ,R^2= 0.68 ,s^2= 5.53 with 11 d.f), with relative water content of the rooting medium. Data points represent daily averages (n=12 for CO_2 assimilation and stomatal conductance, n=6 for (F'm- F)/ F'm and NPQ). Closed symbols (♦) represent values of plants with imposed water stress and open symbols represent (◊) control plants. Vertical bars indicate standard error of mean.

Although all chlorophyll fluorescence parameters mentioned above showed a direct response to reduction of water availability, this response was not clearly observed for PRI (Fig 3.2.2). PRI gradually decreased with RWC$_s$%.

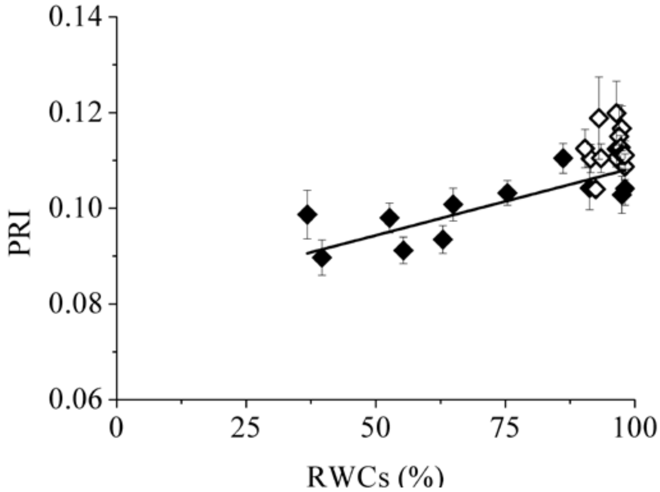

Fig 3.2.2. Correlation between PRI and slab relative water content (s.e of intercept= 0.0037 and of B1= 5 10^{-5}, R^2= 0.61, $s^2$1.79 with 10 d.f), t. PRI values correspond to daily averages (n=12). Closed symbols (♦) represent values of plants with imposed water stress and open symbols represent (◊) control plants. Vertical bars indicate standard error of mean

Previous studies show that PRI is sensitive to changes in light intensity. To investigate the relationship between RWC$_s$% and PRI in relation to light intensity, the data were divided into four different categories on the basis of the occurring light levels during the repetitions. These levels were: i. 0-299 µmol m^{-2} s^{-1}, ii. 300-499 µmol m^{-2} s^{-1}, iii. 500-699 µmol m^{-2} s^{-1} and iv. 700-850 µmol m^{-2} s^{-1}. PRI showed no statistical relationship with RWC$_s$% for light intensities from 0-699 µmol m^{-2} s^{-1} (Fig 3.2.3A-3.2.3C). Nevertheless when radiation was above 700 µmol m^{-2} s^{-1}, PRI showed a good correlation with RWC$_s$% (R^2= 0.83, $p< 0.01$) (Fig. 3.2.3D). In this category PRI decreased as much as 35%.

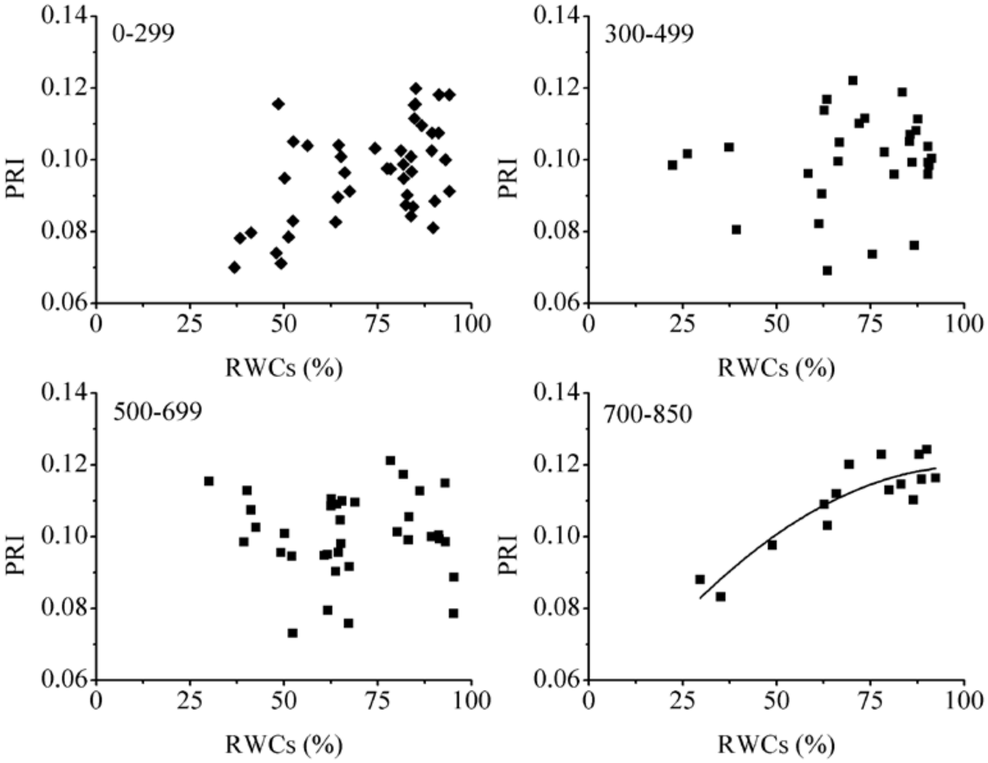

Fig 3.2.3. Correlation of PRI and slab relative water content for light intensities varying from 0-299 μmol m^{-2} s^{-1}, 300- 499μmol m^{-2} s^{-1}, 500-699 μmol m^{-2} s^{-1} and 700-850 μmol m^{-2} s^{-1} (s.e of intercept= 0.015, of B1= $5.2*10^{-4}$ and B2= $4.1*10^{-6}$, R^2= 0.80 ,s^2= $2.98*10^{-5}$ with 12 d.f),. Data points represent half-hour measurements.

For light levels above 700 μmol m^2 s^{-1}, PRI showed a statistically significant correlation with CO_2 assimilation (R^2= 0.66, p<0.01), stomatal conductance (R^2= 0.63, p< 0.01), the operating efficiency of PSII (R^2= 0.7, p< 0.01) and NPQ (R^2= 0.69, p< 0.01) (Fig. 3.2.4). This correlation was not observed in lower light levels.

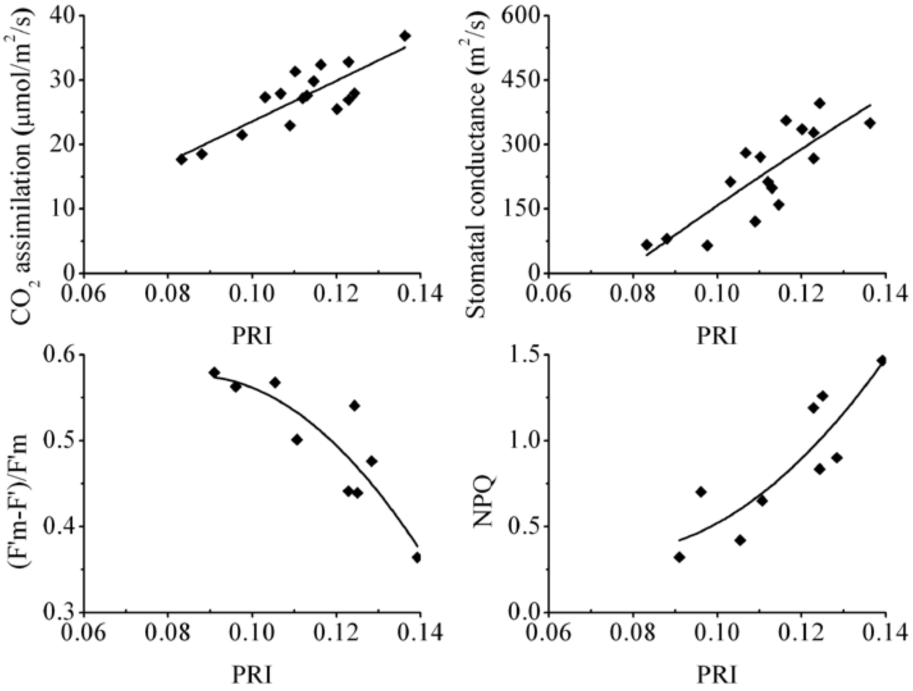

Fig 3.2.4. Correlation of CO_2 assimilation (s.e of intercept=6.15 and of B=54.9, R^2= 0.68, s^2= 8.42 with 14 d.f), Stomatal conductance (s.e of intercept=135.33 and of B=1207.9, R^2= 0.66,s^2= 4079.5 with 14 d.f), Quantum yield of PSII efficiency (F'm- F')/ F'm) (s.e of intercept= 0.81, of B1= 14.45 and B2= 63.33 ,R^2= 0.71 ,s^2= 0.00155 with 6 d.f), and NPQ (s.e of intercept= 4.45, of B1= 79.23 and B2= 347 ,R^2= 0.68 ,s^2= 0.046 with 6 d.f), with PRI for light intensities of 700-850 μmol m^{-2} s^{-1}. Data points represent half-hour measurements for CO_2 assimilation and stomatal conductance and hour measurements for (F'm- F)/ F'm and NPQ).

Discussion

The objective of this study was to evaluate PRI as a method for early detection of water stress in a greenhouse tomato crop.

The RWC_S% is a factor regularly used in literature for indication of water stress imposed on plants. In the current study it was shown that when RWC_S% decreased plant physiological processes are affected. More analytically a reduced RWC_S% results in stomatal closure, diminishing of the photosynthetic rate and quantum yield as well as the increase of *NPQ*. These results are in accordance with the observations of Flexas and Medrano (2002). According to Cornic (2000) the stomatal closure is the primary cause of the reduction of photosynthesis under mild water stress and is also dependent on the environmental conditions during drought (Schulze and Hall 1982), and the velocity of the drought imposition (Flexas *et al.* 1999). A significant correlation between CO_2 assimilation and stomatal conductance was found in the currently presented experiment. The development of water stress differed between repetitions and was directly linked to changes in light.

Chlorophyll fluorescence parameters have been reported to be sensitive to the plant water status and plant stress in general (Lichtenthaler and Babani, 2000; Baker and Rosenqvist, 2004). Increase of water stress generally decreases the F'_q/F'_m. In order to protect the photosystems from oxidation, excess energy is dissipated as heat, resulting in a substantial increase of NPQ (Baker, 2008). In the currently presented study, NPQ increased with decreasing RWC_s% which indicated its sensitivity to plant water stress. The decrease of quantum yield found here was similar to decreases observed by Evain *et al.* (2004). In stressed conditions excess energy on photosystems is dissipated as heat from secondary pigments as the xanthophylls in order to protect the photosystems from damage. When fluorescence parameters are changed, changes should also be observed in PRI. Sun *et al.* (2007) and Penuẽla *et al.* (1994) reported a decrease in PRI with time of water withholding. Also Suarez et al (2009) reported an indirect relationship between PRI and water stress through a correlation of PRI with canopy temperature. Contrary to the aforementioned papers, a direct effect of water stress on PRI was not observed in the current study, unless light levels were above 700 µmol m^{-2} s^{-1}.

When plants are exposed to abiotic and biotic stresses in the light, decreases in F_v/F_m are frequently observed. Zarco-Tejada *et al.*(2000) observed a diurnal pattern in F_v/F_m. Its value decreased during the middle of the day. In most studies (Gamon *et al.* 1997; Flexas and Medrano, 2002), dark adapted fluorescence parameters are measured under pre-dawn conditions and assumed to be steady for the rest of the day. Measurements done in the currently presented study showed variability in the dark adapted maximal PSII efficiency (F_v/F_m) during the day. In early morning F_v/F_m was equal to 0.82 and decreased up to 10% by 13:00. A similar reduction on F_v/F_m values during the day was also reported by Sobrado (2008). This indicates that the common practice of measuring F_v/F_m only in predawn conditions in stress experiments is not representative for the variability of F_v/F_m during the day and hence not suitable for calculation of parameters using either dark adapted F_m or F_o, which could lead to errors of estimation,

When a plant is under stressed conditions, non-photochemical quenching is increased. In the currently described experiment NPQ increased as the stress progressed indicating a loss of energy from the photosytems as heat (Baker, 2008). It is well established that xanthophyll cycle contributes essentially to the dissipation of the excess excitation energy in the PSII antenna under stressed plant conditions (Jahns *et al.*, 2009). The increase in NPQ was more pronounced in days with high light levels (>700μmol m^{-2} s^{-1}), indicating that the RWC_s% plays a synergetic role to light (photoinhibition) affecting photosynthesis and stomatal conductance. Water stress is often indiscernible from photoinhibition in plants. Lu and Zhang (1998) reported that the extent of photoinhibition became more pronounced when the stress progressed. Similar synergistic effects of water and light stress on the photochemistry of the plants are reported in the literature (Masojidek *et al.* 1991; Giardi *et al.* 1996; Genty *et al.* 1987; Jefferies 1994). Many authors indeed argue that the degree of the damage to the photosynthetic properties of the plant by water stress can be amplified by high irradiancies (Powles 1984; Björkman and Powles, 1983).

Gamon *et al.* (1990) as well as Ruban et al (1993) showed that PRI corresponds to fast changes of incident light. PRI is connected with the de-epoxidation state of the xanthophylls in the photosynthetic cycle. The xanthophyll cycle role is mainly to protect the photosystem from damage under stress conditions. Therefore the xanthophyll pool and the epoxidation state of the xanthophyll are also sensitive to the incident light on the leaf level (Maxwell *et al.*, 1994; Schindler and Lichtenthaler, 1994). The de- epoxidation cycle starts only when light absorbed by chlorophyll reaches excessive levels. In the Mediterranean region where the light is in abundance especially in summer period, PRI was used to track successfully the crop water status. Gamon *et al.* (1997) reported that PRI correlated well with light use efficiency only under high light conditions (PPFD<500μmol m^{-2} s^{-1}). On the other hand Sims *et al.* (2006) did not observe a direct correlation between PRI and LUE during the day. In our experiment we showed that PRI is strongly correlating with RWC_s% only when PPFD became higher than 700μmol m^{-2} s^{-1}, indicating that light is of prime importance when it comes to PRI. As Barton and North (2001) showed with a model, PRI is not only dependent on light levels but on plant structure (Suarez *et al.*, 2008), plant optical properties and sun position as well. This assumption was confirmed also by Peguero-Pina (2008) who indicated that early day measurements of PRI, when the sun is still low, do not correlate well with fluorescence as expected. Since sun position plays a role on PRI changes it comes to reason that greenhouse structure prohibits the correct PRI functioning at certain hours of day. Diversion between our data and the published work at PRI can also be attributed to changes in latitude and consequently to solar angle.

Conclusions

In this paper it was shown that PRI can be used as an early water stress indicator only when light intensity at crop level is above 700 μmol m^{-2} s^{-1}. At lower values of light intensity the relationship of PRI to RWCs% was poor in comparison to photosynthesis or fluorescence parameters that showed a high correlation to RWC_s%. For that reason we can conclude that PRI as water stress indicator cannot be independent of the ambient light conditions and its use can make sense only under conditions of high light**.**

Chapter 4

General discussion

The aim of this thesis was to explore different techniques to simulate and monitor light interception and photosynthesis by a greenhouse grown tomato canopy. For that purpose a three dimensional model for accurate light interception and photosynthesis was developed and different methodologies to monitor online these physiological properties by means of remote sensing were explored. In this chapter the advantages of these approaches in the broader scientific field are discussed, as well as their shortcomings and how these can be surmounted.

Modelling light interception and photosynthesis

Modelling light interception and photosynthesis are basic to plant modelling. Both these parameters are highly affected by plant architecture. Light absorption is dependent on both the optical properties of the leaf as well as the leaf position with respect to the incoming light. The importance of leaf position for light absorption is such that, an accurate representation of canopy structure in space is necessary for correct light absorption simulations. This need has been addressed recently by adding one more dimension in plant modelling (Vos *et al.*, 2010).

Adding a dimension in plant modelling involves a number of tedious measurements of plant architecture that are time consuming. Even when the third dimension is added to the model, assumptions are introduced in order to simplify the structure. Leaf positioning towards the incoming light is dependent on the elevation angle (angle of the single leaf to the horizontal). A common assumption in 3D modelling however involves the use of an elevation angle distribution on the entire canopy rather than attribute a specific elevation angle to the leaves that is dependent on each age and relative rank in the canopy. In this thesis (Chapter 2.2) we showed clearly that an explicitly described leaf elevation angle distribution can improve modelling accuracy up to 8% in light absorption in comparison to the ellipsoidal distribution that is the normal approach of most models (Farque *et al.*, 2001).

The importance of a three- dimensional model is not confined to an accurate simulation output but it can also improve our understanding of the impact of the chosen experimental approaches on the acquired data. In this thesis (Chapter 2.2) we used the model to investigate the vertical light distribution inside a double row canopy. The results showed that leaves positioned towards the path absorbed more light per unit leaf area than leaves positioned towards the middle of the plant row. This difference is important in the context of the common experimental approach to measure leaf photosynthesis only on leaves facing the path and not in the middle of the plant row. This approach can lead to serious errors of estimation if these data are used for an estimation of canopy photosynthesis or the calibration of a plant model. The evaluation of experimental techniques through 3D modelling is starting to be explored in the field, since the addition of one more dimension can help improve the microclimate simulation around different plant organs (Sinoquet *et al.*, 2005).

The general characteristics of plant architecture are defined by the plant genotype. Allowing for the environmental plasticity of the structure, an ideotype structure can be identified for each plant grown under certain environmental conditions. The idea of a plant ideotype is hardly new (Donald, 1965), but the use of 3D models in exploring plant phenotypes can prove a very important tool in plant breeding by cutting the time and costs needed on the traditional approach. A plant structure as any architectural structure is a synthesis of a number of smaller components, whose unique topology and geometry can have a direct effect on light absorption and photosynthesis. In the thesis (Chapter 2.3) we identified as the main components of tomato architectural structure the leaf elevation angle, phyllotactic angle, leaflet angle, leaf length to width ratio, leaflet arrangement and internode length. Then we proceeded to quantify the effect of the change of each of these components both on light absorption and photosynthesis. From our analysis we found that the highest impact had the leaf length:width ratio and internode length depending on the light conditions. From the study we conclude that a more spatial arrangement of architectural components increased both light absorption and photosynthesis. Based on this result we proposed two types of plant ideotypes for the particular conditions that both aimed at a more spatial arrangement. The model predicted a possible increase of photosynthesis of up to 10%. Highly interesting was the exploration of the effect of the vertical light absorption profiles on photosynthesis for summer and winter. Simulation data presented in Chapter 2.3 showed that during summer higher photosynthesis was expected when more light was absorbed from the lower leaves of the canopy while the opposite was observed for the winter period. This was caused by both overall light intensity as well as the different light angle fot the different seasons. Therefore, it becomes apparent that the vertical light distribution should not be ignored and that total light interception alone is not enough for simulating photosynthesis.

In the two dimensional models plant structure is approached by using the extinction coefficient (k) as it is defined by Lambert-Beer law. Although this factor is a good approximation it fails to explore the effect of the variations in vertical light distributions. As we showed in this thesis (Chapter 2.1) the extinction coefficient changes not only with canopy depth but also with the increase of the row spacing. Therefore a single coefficient cannot be used for one crop or different cultivation practises.

In this thesis we used a static approach in modelling plant structure. The choice to use a static structure was based on the Dutch cultivation system that results in a uniform plant structure throughout the seasons. Nevertheless the model does not take into account the daily changes of plant structure. For example plants tend to rearrange their structure during the day as well as during the growing season. These dynamic adaptations of plant structure should be taken into account in the model since they are part of the plant strategy for optimum growth. Some possible effects of the dynamic leaf adaptation during the day are for example under low light conditions an increasing photosynthetic production or under high light conditions avoiding photo inhibition. Studies have reported leaf movement during the day (Kao and Forseth, 1992). These phenomena are quite important for our understanding of plant interactions with their environment and should therefore be taken into account also in a modelling approach especially if we use this type of modelling for proposing possible optimum genotypes for particular environments.

Online crop monitoring

Monitoring leaf area index and light interception

Leaf area index as well as light interception is an important component of plant modelling. Experimental methods to measure light interception are usually time consuming and can only take a snapshot in a certain time period and therefore miss the dynamics of this component. Leaf area index is usually estimated using distractive measurements, which are not always possible to do, and they limit the number of samples being processed. In order to avoid these pitfalls an effort has been made to develop online non- destructive methods. Remote sensing that monitors the light that is reflected from the crop canopy has successfully been applied in the field (Clevers, 1997, Gitelson *et al.*, 2002). Nevertheless this approach poses special challenges inside the greenhouse environment, as we have to take into account the interference of scattering light from the construction parts of the greenhouse as well as reflectance coming from the white plastic covering the soil (Chapter 3.1). Although it is not possible to directly apply these methodologies from the field, in this thesis we showed that we can develop new ones that can succesfully measure LAI and light interception in the greenhouse environment.

In field crops it is customary to monitor LAI by using a normalized index based on the red area of the spectrum i.e. Normalized Difference Vegetation Index (NDVI) (Wang *et al.,* 2005). The rationale behind using an index based on the red part of the spectrum is based on the fact that light emitted in this part of the spectrum has a strong link with structural and physiological processes of the plant. In this thesis we showed that this index cannot be used under greenhouse conditions and that reflectance in the blue part of the spectrum gives a much better correlation with both LAI and light interception. On the contrary reflectance around 460nm showed a good correlation with both LAI and light interception in different environments and in several plant species. An application of this findings in a permanent sensor that was positioned on the top of the greenhouse and took reflectance measurements at 460nm in real time, gave also a good correlation between light interception and reflectance (Janssen *et al.*, 2006). This differentiation between our result and what is usually applied in the field can have a number of explanations. One reason could be the resolution of the sensor used in this experiment as the broad bands in which reflectance was measured could miss the subtle changes in the reflectance both during the day and during the season. A sensor with higher resolution could address this problem. Another reason could be the background light scattering in the greenhouse is so strong that is impossible to monitor reflectance changes in the red part of the region.

Monitoring photosynthetic stress

The ability to monitor photosynthetic activity is important not only for model parameterisation but also for a dynamic assessment of plant status. During the cultivation period climatic stress parameters such as high light conditions or water deprivation can cause a decrease in plant photosynthetic efficiency. Different non-destructive methods have been explored in recent years in order to monitor plant stress for example canopy reflectance (Evain *et al.* 2004), infrared cameras (Langton *et al.*, 2002), fluorescence meters (Rosema *et al.*, 1998), pressure probes (Zimmermann *et al.*, 2008), sap flow sensors (de Swaef and Steppe, 2010), emitted plant volatiles (Janssen *et al.*, 2009). In this thesis we studied the use of the photochemical reflectance index (PRI) as photosynthetic stress indicator. The photosynthetic stress was induced by applying water stress on the plants. The results of our experiment were positive under high light conditions where the stress effect from the water deprivation was enhanced by photoinhibition. The results from our experiments showed that PRI can be used as an indicator of photosynthetic stress in places where high light conditions occur. it could be a useful tool in arid areas with ubiquitous high irradiance levels, however on a regular base its use has a limited applicability for the relatively short summer period in the Netherlands.

PRI uses wavelengths that have been related to the de-epoxidation of xanthophylls. During this process it was found that light is emitted from 500-545nm. The wavelength of 531nm used by PRI is linked with the fast-changes of the xanthophyll cycle and possible is not the best measure for gradual changes of the photosynthetic status. A few studies have stated that instead of using 531nm a PRI calculation based on spectra from 521-550nm (Gamon *et al.*, 1997, Wu *et al.*, 2008) would give a better correlation with photosynthetic efficiency in some species. This shift in wavelength can be caused by the leaf morphology and the optical properties (Noomen *et al.*, 2006).

Possibilities for further research

In this thesis we presented the advantages of a three-dimensional model approach as well as the applicability of canopy reflectance as a tool for monitoring non-destructively the leaf area index, light interception and photosynthetic stress in the plant canopy. Based on the results of this thesis, further steps to improve this work and forward the research would include:

- Incorporate a dynamic component in plant architecture so as plant development can be taken into account. As discussed plant structure can show high plasticity depending on the environmental conditions. Functions that predict plant palsticity would be helpful in our understanding of the plant adapatation on its environment. Such a dynamic adaptive model could also prove a useful tool for investigating the performance of a number of phenotypes in different environments for a whole cultivation season without having to resort to expensive and time consuming experimental methods.

- In this thesis we measured PRI only in two specific wavelength. However as it was mentioned earlier in the discussion the wavelengths we used to calculate PRI, monitor only the fast kinetic changes of the xanthophylls, while the leaf morphology and the possible effect of the epidermis is not taken into account. Therefore, the improvement of PRI efficiency should be investigated in relation to the wavelengths related to the slow kinetics of the xanthophyll cycle as well as determine the role of the leaf morphology.
- Finally the model should be coupled to the monitoring sensors. This online feedback of the model in real time would be a powerful tool to improve the model accuracy and also boost the efficiency of decision support systems which in turn will enable the better management of the crop in terms of yield and quality.

References

Adams W.W.III, Volk M., Hoehn A., Demmig- Adams B. 1992. Leaf orientation and the response of the xanthophyll cycle to incident light. Oecologia, 90, 404–410.

Atherton J.G.; Rudich J. 1986. The tomato crop: a scientific basis for improvement. Chapman and Hall, London.

Baker N.R., Ort D.R. 1992. Light and crop photosynthetic performance. In Crop Photosynthesis: Spatial and Temporal Determinants, pp. 289-312. London: Elsevier Science Publishers B.V.

Baker N.R., Rosenqvist E. 2004. Applications of chlorophyll fluorescence can improve crop production strategies: an examination of future possibilities. Journal of Experimental Botany, 55: 1607–1621.

Baker N.R. 2008. Chlorophyll Fluorescence: A Probe of Photosynthesis In Vivo. Annual Review of Plant Biology, 59: 89–113.

Baldocchi D.D., Finnigan J.J, Wilson K.W., Paw K.T., Falge E. 2000. On measuring net ecosystem carbon exchange in complex terrain over tall vegetation. Boundary Layer Meteorology, 96: 257-291.

Barthélémy D., Caraglio Y. 2007. Plant architecture: a dynamic, multilevel and comprehensive approach to plant form, structure and ontogeny. Annals of Botany, 99: 375-407.

Barton C.V.M., North P.R.J, 2001. Remote sensing light use efficiency using the photochemical reflectance index. Remote sensing of Environment, 78: 264–273.

Bausch W.C. 1995. Remote sensing of crop coefficients for improving the irrigation scheduling of corn. Agricultural Water Management, 27: 55–68.

Bilger W., Björkman O. 1990. Role of the xanthophylls cycle in photoprotection elucidated by measurements of light-induced absorbance changes, fluorescence and photosynthesis in Hedera canariensis. Photosynthetic Research, 25: 173–85.

Björkman O, Powles B. 1984. Inhibition of photosynthetic reactions under water stress: interaction with light level. Planta, 161: 490-504.

Björkman O., Demmig B. 1987. Photon yield of O2 evolution and chlorophyll fluorescence characteristics at 77 K among vascular plants of diverse origins. Planta, 170: 489–504.

Boonen C., Samson R., Janssens K., Pien H., Lemeur R., Berckmans D. 2002. Scaling the spatial distribution of photosynthesis from leaf to canopy in a plant growth chamber, Ecological Modelling, 156: 201-212.

Boote K.J., Scholberg J.M.S. 2006. Developing, parameterizing, and testing of dynamic crop growth models for horticultural crops. Acta Horticulturae, 718:23-34.

Borger C.P.D., Hashem A., Pathan S. 2010. Manipulating Crop Row Orientation to Suppress Weeds and Increase Crop Yield. Weed Science, 58: 174-178.

Bouman B.A.M.; Uenk D.; Haverkort A.J. 1992. The estimation of ground cover of potato by reflectance measurements. Potato research, 35: 111- 125

Bouman B.A.M. 1994. A framework to deal with uncertainty in soil and management parameters in crop yield stimulation: A case study in rice. Agricultural systems, 46: 1- 17

Bowman W.D. 1989. The relationship between leaf water status, gas exchange, and spectral reflectance in cotton leaves. Remote Sensing of Environment, 30: 49–255.

Brites D. Valladares F. 2005. Implications of opposite phyllotaxis for light interception efficiency of Mediterranean woody plants. Trees, 19: 671-679.

Buck-Sorlin G., de Visser P.H.B., Henke M., Sarlikioti V., van der Heijden G.W.A.M., Marcelis L.F.M., Vos J. 2011. Towards a functional-structural plant model of cut-rose - simulation of light environment, light absorption, photosynthesis and interferences with the plant structure. Annals of Botany, in press

Casella E., Sinoquet H. 2007. Botanical determinants of foliage clumping and light interception in two-year-old coppice poplar canopies: assessment from 3-D plant mock-ups. Annals of Forest Science, 64: 395- 404.

Ceccato P., Flasse S., Tarantola S., Jacquemoud S., Gregoire J-M. 2001. Detecting vegetation leaf water content using reflectance in the optical domain. Remote Sensing of Environment, 77: 22–33.

Chaves M.M., Pereira J.S., Maroco J., Rodrigues M.L., Ricardo C.P.P., Osório M.L., Carvalho I., Faria T., Pinheiro C. 2002. How Plants Cope with Water Stress in the Field? Photosynthesis and Growth. Annals of Botany, 89: 907–916.

Chelle M., Andrieu B. 1998. The nested radiosity model for the distribution of light within plant canopies. Ecological Modelling, 111: 75-91.

Chelle M. 2005. Phylloclimate or the climate perceived by individual plant organs: What is it? How to model it? What for? New Phytologist, 166: 781-90.

Chen J.M., Pavlic G., Brown L., Cihlar J., Leblanc S.G., White H.P., Hall R.J., Peddle D., King D.J., Trofymow J.A., Swift E., Van der Sanden J., Pellikka P. 2002. Validation of Canada-wide leaf area index maps using ground measurements and high and moderate resolution satellite imagery. Remote Sensing of Enviroment, 80: 165-184.

Clevers J.G.P.W. 1997. A simplified approach for yield prediction of sugar beet based on optical remote sensing data. Remote Sensing of Environment, 61: 221-228.

Cornic G., Briantais J.M. 1991. Portioning of photosynthetic electron flow between CO2 and O2 reduction in C3 leaf (Phaseolus vulgaris L.) at different CO2 concentrantions and during drought stress. Planta, 183: 178–184.

Cornic G. 2000. Drought stress inhibits photosynthesis by decreasing stomatal aperture- not by affecting ATP synthesis. Trends in Plant Science, 5: 187–188.

Cournède P.H., Mathieu A., Houllier F., Barthélémy D., de Reffye P. 2008. Computing competition for light in the GREENLAB model of plant growth: a contribution to the study of the effects of density on resource acquisition and architectural development. Annals of Botany, 101: 1207–1219.

Danson F.M., Steven M.D., Malthus T.J., Clark J.A. 1992. High-spectral resolution data for determining leaf water content. International Journal of Remote Sensing, 13: 461–470.

Danson F.M., Aldakheel Y.Y. 2000. Diurnal water stress in sugar beet: spectral reflectance measurements and modeling. Agronomie, 20: 31–39.

Dauzat J., Clouvel P., Luquet D., Martin P. 2008. Using virtual plants to analyse the light-foraging efficiency of a low-density cotton crop. Annals of Botany, 101: 1153–1166.

De Castro F., Fetcher N. 1999. The effect of leaf clustering in the interception of light in vegetal canopies: theoretical considerations. Ecological Modelling, 116: 125-134.

de Swaef T., Steppe K. 2010. Linking stem diameter variations to sap flow, turgor and water potential in tomato. Functional Plant Biology, 37: 429–438.

Donald C.M. 1965. The breeding of crop ideotypes. Euphytica 17: 385-403.

Dong Q.X., Wang X.L., Yang L.L., Barczi J.F., De Reffye F. 2007. Greenlab tomato: a 3D architectural model of tomato development. New Zealand Journal of Agricultural Research, 50: 1229-1233.

Drouet J.L., Pagès L. 2007. GRAAL-CN: A model of Growth, Architecture and Allocation for Carbon and Nitrogen dynamics within whole plants formalized at the organ level. Ecological Modelling, 206: 231-249.

Elachi C. 1987. Introduction to the Physics and Techniques of Remote Sensing, pp. 66–71. New York: John Wiley.

Elwadie M.E.; Pierce F.J.; Qi J. 2005. Remote Sensing of Canopy Dynamics and Biophysical Variables Estimation of Corn in Michigan. Agronomy Journal, 97: 99-105.

Evain S., Flexas J., Moya I. 2004. A new instrument for passive remote sensing: 2. Measurement of leaf and canopy reflectance changes at 531 nm and their relationship with photosynthesis and chlorophyll fluorescence. Remote Sensing of Environment, 91: 175–185.

Evers J.B., Vos J., Fournier C., Andrieu B., Chelle M., Struik P.C. 2005. Towards a generic architectural model of tillering in Gramineae, as exemplified by spring wheat (Triticum aestivum). New Phytologist, 166: 801-812.

Falster D.S., Westoby M. 2003. Leafs size and angle vary widely across species: what consequences for light interception? New Phytologist,158: 509-525.

Farque L., Sinoquet H., Colin F. 2001. Canopy structure and light interception in Quercus petraea seedlings in relation to light regime and plant density. Tree Physiology, 21: 1257–1267.

Farquhar G.D., von Caemmerer S., Berry J.A. 1980. A biochemical-model of photosynthetic CO2 assimilation in leaves of C3 species. Planta, 149: 78-90.

Flexas J., Escalona J.M., Medrano H. 1999. Water stress induces different photosynthesis and electron transport rate regulation in grapevine. Plant, Cell and Environment, 22: 39–48.

Flexas J., Medrano H. 2002. Drought-inhibition of photosynthesis in C3 plants: stomatal and non-stomatal limitations revisited. Annals of Botany, 89, 183–189.

Foolad M.R., Zhang L.P., Subbiah P. 2003. Genetics of drought tolerance during seed germination in tomato: Inheritance and QTL mapping. Genome, 46: 536–545.

Forseth N. 1990. Function of leaf movements. 238–261 pp. In: Satter RL, Gorton HL, Vogelmann TC (eds) The pulvinus: motor organ for leaf movement.

Fourcaud T., Zhang X., Stokes A., Lambers H., Körner C. 2008. Plant growth modelling and applications: the increasing importance of plant architecture in growth models. Annals of Botany, 101: 1053-1063.

Frary A., Fritz L.A., Tanksley S.D. 2004. A comparative study of the genetic basis??? of natural variation in tomato leaf, sepal, and petal morphology. Theoretical and Applied Genetics, 109: 523-533.

Gamon J.A., Field C.B., Bilger W., Björkman O., Fredeen A.L., Peñuelas J. 1990. Remote sensing of the xanthophyll cycle and chlorophyll fluorescence in sunflower leaves and canopies. Oecologia, 85: 1–7.

Gamon J.A., Serrano L., Surfus J.S. 1997. The photochemical reflectance index: An optical indicator of photosynthetic radiation use efficiency across species, functional types, and nutrient levels. Oecologia, 112: 492–498.

Gary C., Barczi J.F., Bertin N., Tchamitchian M. 1995. Simulations of individual organ growth and development on a tomato plant: a model and user-friendly interface, Acta Horticulturae, 399: 199–205.

Gayler S., Grams T.E.E., Kozovits A.R., Winkler J.B., Luedemann G., Priesack E. 2006. Analysis of competition effects in mono- and mixed cultures of juvenile beech and spruce by means of the plant growth simulation model PLATHO. Plant Biology, 8: 503-514.

Genty B., Briantais J.M., Viera da Silva J.B. 1987. Effects of drought on primary photosynthetic processes of cotton leaves. Plant Physiology, 83: 360–364.

Giardi M.T., Cona A., Geiken B., Kucera T., Masojidek J., Mattoo A.K. 1996. Long- term drought stress induces structural and functional reorganisation of photosystem II. Planta, 199: 118–125.

Gilbert R.A., Heilman J.L., Juo A.S.R. 2003. Diurnal and Seasonal Light Transmission to Cowpea in Sorghum–Cowpea Intercrops in Mali. Journal of Agronomy and Crop Science, 189: 21- 29.

Gitelson A.A. 2002. Wide Dynamic Range Vegetation Index for remote quantification of biophysical characteristics of Vegetation. Journal of Plant Physiology, 161: 165-173.

Godin C. 2000. Representing and encoding plant architecture: a review. Annals of Forest Science, 57: 413-438.

Godin C., Sinoquet H. 2005. Functional-structural plant modelling. New Phytologist, 166: 705-708.

González-Real M.M., Baille A., Gutiérrez Colomer RP. 2007. Leaf photosynthetic properties and radiation profiles in a rose canopy (Rosa hybrida L.) with bent shoots. Scientia Horticulturae, 114: 177–187.

Goudriaan J., van Laar H.H. 1994. Modelling potential crop growth processes. Kluwer Academic Publishers, Dordrecht.

Graeff S., Claupein W. 2007. Identification and discrimination of water stress in wheat leaves (Triticum aestivum L.) by means of reflectance measurements. Irrigation Science, 26: 61–70.

Guo J., Trotter C.M. 2004. Estimating photosynthetic light-use efficiency using the photochemical reflectance index: variations among species. Function Plant Biology, 31: 255–265.

Hanan J. 1997. Virtual plants: integrating architectural and physiological models. Environmental Modelling and Software, 12: 35-42.

Heuvelink E. 1999. Evaluation of a dynamic simulation model for tomato crop growth and development. Annals of Botany, 83: 413-422.

Higashide T. 2009. Light interception by tomato plants (Solanum lycopersicum) grown on a sloped field. Agricultural and Forest Meteoreology, 149: 756-762.

Hoffmann C.M; Blomberg M. 2004. Estimation of Leaf Area Index of Beta vulgaris L. Based on Optical Remote Sensing Data. Journal of Agronomy and Crop Science, 190: 197-204.

Hsiao T., Steduto P., Fereres E. 2007. A systematic and quantitative approach to improve water use efficiency in agriculture. Irrigation Science, 25: 209-231.

Jahns P., Latowski D., Strzalka K. 2009. Mechanism and regulation of the violaxanthin cycle: The role of antenna proteins and membrane lipids. Biochimica et biophysica acta-bioenergetics, 1787: 3–14.

Jansen R.M.C., Hofstee J.W., Wildt J., Verstappen F.W.A., Bouwmeester H.J., Posthumus M.A., Van Henten E.J.V. 2009. Health monitoring of plants by their emitted volatiles: trichome damage and cell membrane damage are detectable at greenhouse scale. Annals of Applied Biology, 154: 441–452.

Janssen H.J.J., Sarlikioti V., Gieling Th.H., Meurs E.J.J., Marcelis L.F.M., Van Dugteren J.R. 2008. A prototype sensor for estimating light interception by plants in a greenhouse. Acta Horticulturae, 801: 621-627.

Jefferies R.A. 1994. Drought and chlorophyll fluorescence in the fieldgrown potato (Solanum tuberosum). Physiologia Plantarum, 90: 93–97.

Jonckheere I.; Fleck S.; Nackaerts K.; Muys B.; Coppin P.; Weiss M.; Baret F. 2004. Review of methods for in situ leaf area index determination: Part I. Theories, sensors and hemispherical photography. Agricultural Forest Meteorology, 121: 19- 35.

Jongschaap R.E.E. 2006. Run- time calibration of simulation models by integrating remote sensing estimates leaf area index and canopy nitrogen. European Journal of Agronomy, 24: 316-324.

Kahlen K. 2006. 3D architectural modeling of greenhouse cucumber (Cucumis sativus l.) Using l-systems. Acta horticulturae, 718: 51-58.

Kahlen K., Wiechers D., Stützel H. 2008. Modelling leaf phototropism in a cucumber canopy. Functional Plant Biology, 35: 876-884.

Kao W.Y., Forseth I.N. 1992. Diurnal leaf movement, chlorophyll fluorescence and carbon assimilation in soybean grown under different nitrogen and water availabilities. Plant, Cell and Environment, 15: 703-710.

Krause G.H., Weis E. 1991. Chlorophyll Fluorescence and Photosynthesis: The Basics Annual Review Plant Physiology Plant Molecular Biology, 42: 313–349.

Lai C.T., Katul G., Ellsworth D., Oren R. 2000. Modelling vegetation–atmosphere CO2 exchange by a coupled eulerian–lagrangian approach, Boundary-Layer Meteorology, 95: 91-122.

Langton F.A., Horridge J.S., Holdsworth M.D., Hamer P.J.C. 2004. Control and optimization of the greenhouse environment using infra-red sensors. Acta Horticulturae, 633: 145–152.

Lawlor D.W., Tezara W. 2009. Causes of decreased photosynthetic rate and metabolic capacity in water-deficient leaf cells: a critical evaluation of mechanisms and integration of processes. Annals of Botany, 103: 561-579.

Lichtenthaler H.K., Babani F. 2000. Detection of photosynthetic activity and water stress by imaging the red chlorophyll fluorescence. Plant Physiology and Biochemistry, 38: 889–895.

Lindenmayer, A., Prusinkiewicz P. 1990. The algorithmic beauty of plants. New York, Springer-Verlag.

Lintz J., Simonett D.S. 1976. Remote Sensing of the Environment. 713 p. Addison-Wesley, Reading, Massachusetts.

Louarn G., Lecoeur J., Lebon E. 2008. A three-dimensional statistical reconstruction model of grapevine (Vitis vinifera) simulating canopy structure variability within and between cultivar/training system pairs. Annals of Botany, 101:1167–1184.

Lu C., Zhang J. 1998 Effects of water stress on photosynthesis, chlorophyll fluorescence and photoinhibition in wheat plants. Australian Journal of Plant Physiology, 25: 883–892.

Maddonni G.A., Otegui M.E., Andrieu B., Chelle M., Casal J.J. 2002. Maize Leaves Turn Away from Neighbors. Plant Physiology, 130: 1181-1189.

Marcelis L.F.M., Heuvelink E., Goudriaan J. 1998. Modelling of biomass production and yield of horticultural crops: a review. Scientia Horticulturae, 74: 83-111.

Marcelis, L.F.M., Van den Boogaard R., Meinen, E. 2000. Control of crop growth and nutrient supply by the combined use of crop models and plant sensors. 351-356 pp. Proc. Int. Conf. Modelling and control in agriculture, horticulture and post-harvested processing. IFAC.

Marcelis L.F.M., Dieleman J.A., Kittas C., De Groot F., Buschmann C., Van Loon A., Boulard T., Brajeul E., Kocsanyi L., Garate A., Wieringa G. 2006. CLOSYS: Closed system for water and nutrient management in horticulture. Acta Horticulturae, 718: 375–382.

Masojidek J., Trivedi S., Halshaw L., Alexiou A., Hall D.O. 1991. The synergistic effect of drought and light stresses in sorghum and pearl millet. Plant Physiology, 96: 198–207.

Maxwell C., Griffiths H., Young A.J. 1994. Photosynthetic acclimation to light regime and water stress by the C3-CAM epiphyte Guzmania monostachia: gas-exchange characteristics, photochemical efficiency and the xanthophyll cycle. Functional Ecology, 8: 746–754.

Meir P., Kruijt B., Broadmeadow M., Barbosa E., Kull O., Carswell F., Nobre A., Jarvis P.G. 2002. Acclimation of photosynthetic capacity to irradiance in tree canopies in relation to leaf nitrogen concentration and leaf mass per unit area. Plant, Cell & Environment, 25: 343–357.

Méthy M., Joffre R., Rambal S. 1999. Remote Sensing of Canopy Photosynthetic Performances: Two Complementary Ways for Assessing the Photochemical Reflectance Index. Photosynthetica, 37: 239–247.

Monsi M.; Saeki T. 1953. Über den lichtfaktor in den pflanzengesellschaften und seine bedeutung für die stoffproduktion. Japanese Journal of Botany, 14: 22-52.

Morales F., Abadía A., Abadía J. 1990. Characterization of the xanthophyll cycle and other photosynthetic pigment changes induced by iron deficiency in sugar beet (Beta vulgaris L.). Plant Physiology, 94: 607–613.

Najla S., Vercambre G., Pagès L., Grasselly D., Gautier H., Génard M. 2009. Tomato plant architecture as affected by salinity: Descriptive analysis and integration in a 3-D simulation model. Botany, 87: 893-904.

Nederhoff E.M.; Vegter J.G. 1994. Canopy Photosynthesis of Tomato, Cucumber and Sweet Pepper in Greenhouses: Measurements Compared to Models. Annals of Botany, 73: 421-427.

Niinemets Ü., Fleck S. 2002. Petiole mechanics, leaf inclination, morphology, and investment in support in relation to light availability in the canopy of Liriodendron tulipifera. Oecologia, 132: 21-33.

Niinemets U. 2007. Photosynthesis and resource distribution through plant canopies. Plant Cell and Environment, 30: 1052-1071.

Niklas K.J. 1998. Light harvesting “fitness landscapes” for vertical shoots with different phyllotactic patterns. 759-773 pp. In: Jean RV, Barabé D, eds. Symmetry in plants. Singapore: World Scientific Publishing Co. Pte. Ltd..

Noomen M.F., Skidmore A.K., van der Meer F.D., Prins H.H.T. 2006. Continuum removed band depth analysis for detecting the effects of natural gas, methane and ethane on maize reflectance, Remote Sensing of Environment, 105:262–270.

Palmer J.W. 1989. The effects of row orientation, tree height, time or year and latitude on light interception and distribution in model apple hedgerow canopies. Journal of Horticultural Scence, 64: 137- 145.

Papadopoulos A.P., Pararajasingham S. 1997. The influence of plant spacing on light interception and use in greenhouse tomato (Lycopersicon esculentum Mill.) : A review. Scientia Horticulturae, 69: l-29.

Pearcy R., Roden J., Gamon J. 1990. Sunfleck dynamics in relation to canopy structure in a soybean (Glycine max (L.) Merr.) canopy. Agricultural and Forest Meteorology, 52: 359-372.

Pearcy R.W., Yang W. 1998. The functional morphology of light capture and carbon gain in the Redwood forest understorey plant Adenocaulon bicolor Hook. Functional Ecology, 12: 543-552.

Peguero-Pina J., Morales F., Flexas J., Gil-Pelegrín E., Moya I. 2008. Photochemistry, remotely sensed physiological reflectance index and de-epoxidation state of the xanthophyll cycle in Quercus coccifera under intense drought. Oecologia, 156: 1–11.

Peng S., Khush G.S., Virk P., Tang Q., Zou Y. 2008. Progress in ideotype breeding to increase rice yield potential. Field Crops Research, 108: 32-38.

Peñuelas J., Biel C., Serrano L., Savé R. 1993. The reflectance at the 950– 970 nm region as an indicator of plant water stress. International Journal of Remote Sensing, 14: 1887–1905.

Peñuelas J., Gamon J.A., Freeden A.L., Merino J., Field C.B. 1994. Reflectance indices associated with physiological changes in nitrogen- and water-limited sunflower leaves. Remote Sensing of Environment, 48: 135–146.

Peñuelas J., Filella I., Araus J.L. 1997. Visible and near-infrared reflectance assessment of salinity effects on barley. Crop Science, 37: 198–202.

Peralta I.E., Spooner D.M. 2000. Classification of wild tomatoes: A review. Kurtziana, 28: 45-54.

Pinter Jr.P.J., Hatfield J.L., Schepers J.S., Barnes E.M., Moran M.S., Daughtry C.S.T., Upchurch D.R. 2003. Remote sensing for crop management. Photogrammetric Engineering and Remote Sensing, 69: 647–664.

Planchais I., Sinoquet H. 1998. Foliage determinants of light absorption in sunny and shaded branches of Fagus sylvatica (L.). Agricultural and Forest Meteorology, 89: 241-253.

Powles S.B. 1984. Photoinhibition of photosynthesis induced by visible light. Annual Review of Plant Physiology, 35: 15–44.

Prusinkiewicz P. 1999. A look at the visual modelling of plants using L-systems. Agronomie, 19: 211-224.

Rakocevic M., Sinoquet H., Christophe A., Varlet-Grancher C. 2000. Assessing the geometric structure of a white clover (Trifolium repens) canopy using 3D digitizing. Annals of Botany, 86:519-526.

Rosema A., Snel J.F.H., Zahn H., Buurmeijer W.F., Van Hove L.W.A. 1998. The relation between laser-induced chlorophyll fluorescence and photosynthesis. *Remote* Sensing of Environment, 65**:** 143–154.

Ruban A.V., Young A.J., Horton P. 1993. Induction of nonphotochemical energy dissipation and absorbance changes in leaves. Plant Physiology, 102: 741–750.

Schindler C., Lichtenthaler H.K. 1994. Is there a correlation between light induced zeaxanthin accumulation and quenching of variable chlorophyll a fluorescence? Plant Physiology Biochemistry, 32: 813–823.

Schulze E.-D., Hall A.E. 1982. Stomatal responses, water loss and CO2 assimilation rates of plants in contrasting environments. In Encyclopedia of plant physiology Vol. 12. Physiological plant ecology ecosystem processes, 263–324 pp. Eds OL Lange, PS Nobel, CB Osmond and H Ziegler. Berlin, Germany: Springer-Verlag.

Schurr U., Walter A., Rascher U. 2006. Functional dynamics of plant growth and photosynthesis – from steady-state to dynamics – from homogeneity to heterogeneity. Plant, Cell and Environment, 29: 340–352.

Sievänen R., Nikinmaa E., Nygren P., Ozier-Lafontaine H., Perttunen J., Hakula H. 2000. Components of functional–structural tree models. Annals of Forest Science, 57: 399-412.

Sims D.A., Luo H., Hastings S., Oechel W.C., Rahman A.F., Gamon, J.A. 2006. Parallel adjustments in vegetation greenness and ecosystem CO2 exchange in response to drought in a Southern California chaparral ecosystem. Remote Sensing of the Environment, 103: 289-303.

Sinoquet H., Sonohat G., Phattaralerphong J., Godin C. 2005. Foliage randomness and light absorption in 3-D digitized trees: an analysis from multiscale discretization of the canopy. Plant Cell and Environment, 28: 1158-1170.

Smith H., Whitelam G.C. 1997. The shade avoidance syndrome: multiple responses mediated by multiple phytochromes. Plant, Cell and Environment, 20: 840-844.

Sobrado M.A. 2008. Leaf characteristics and diurnal variation of chlorophyll fluorescence in leaves of the ‘bana’ vegetation of the amazon region. Photosynthetica, 46: 202-207.

Stewart D.W., Costa C., Dwyer L. M., Smith D. L., Hamilton R. I., Ma B. L. 2003. Canopy Structure, Light Interception, and Photosynthesis in Maize. Agronomie Journal, 95: 1465-1474.

Suárez L., Zarco-Tejada P.J., Sepulcre-Cantó G., Pérez-Priego O., Miller J.R., Jiménez-Muñoz J.C., Sobrino, J. 2008. Assessing canopy PRI for water stress detection with diurnal airborne imagery. Remote Sensing of the Environment, 112: 560-575.

Suárez L., Zarco-Tejada P.J., Berni J.A.J., Gonzalez-Dugo V., Ferreres E. 2009. Modeling PRI for water stress detection using radiative transfer models. Remote Sensing of the Environment, 113: 730-744.

Sun P., Grignetti A., Liu S., Casacchia R., Salvatori R., Pietrini F., Loreto F., Centritto M. 2007. Associated changes in physiological parameters and spectral reflectance indices in olive (Olea europaea L.) leaves in response to different levels of water stress. International Journal of Remote Sensing, 29: 1725–1743.

Takenaka A. 1994. Effects of leaf blade narrowness and petiole length on the light capture efficiency of a shoot. Ecological Research, 9: 109-114.

Thornley J.H.M., Johnson I.R. 1990. Plant and Crop Modelling: In A Mathematical Approach to Plant and Crop Physiology. 669 pp. Oxford, UK: Clarendon Press

Thurling N. 1991. Application of the ideotype concept in breeding for higher yield in the oilseed brassicas. Field Crops Research, 26: 201-219.

Tian Y.Q., Zhu Y., Cao W. 2005. Monitoring leaf photosynthesis with canopy spectral reflectance in rice. Photosynthetica, 43: 481-489.

Toler J.E., Murdock E.C., Stapleton G.S., Wallace S.U. 1999. Corn leaf orientation effects on light interception, intraspecific competition, and grain yields. Journal of production agriculture, 12: 396-399.

Turner D.P., Cohen W.B., Kennedy R.E., Fassnacht K.S., Briggs J.M. 1999. Relationships between Leaf Area Index and Landsat TM Spectral Vegetation Indices across Three Temperate Zone Sites. Remote Sensing of Environment, 70: 52-68.

Valladares F., Pearcy R.W. 1998. The functional ecology of shoot architecture in sun and shade plants of Heteromeles arbutifolia M. Roem., a Californian chaparral shrub. Oecologia, 114: 1-10.

Valladares F. 2003. Light heterogeneity and plants: from ecophysiology to species coexistence and biodiversity. Progress in Botany. 439–471 pp. Springer Verlag, Berlin, Germany.

Valladares F., Gianoli E., Gómez J.M. 2007. Ecological limits to plant phenotypic plasticity. New Phytologist, 176: 749–763.

Valladares F., Niinemets Ü. 2007. The architecture of plant crowns: from design rules to light capture and performance. 101-149 pp. In: Pugnaire F, Valladares F, eds. Functional plant ecology. New York: Taylor and Francis.

Vargas L.A., Andersen M.N., Jensen C.R., Jørgessen U. 2002. Estimation of leaf area index, light interception and biomass of Miscanthus sinensis ‘Goliath’from radiation measurements. Biomass and bioenergy, 22: 1-14.

Vos J., Marcelis L.F.M., Evers J.B. 2007. Functional-structural plant modelling in crop production: adding a dimension. 1-12 pp. In: Vos, J., Marcelis, L.F.M., De Visser, P.H.B., Struik, P.C. and Evers, J.B. eds. Functional-structural plant modelling in crop production. Springer, Dordrecht.

Vos J., Evers J.B., Buck-Sorlin G.H., Andrieu B., Chelle M., de Visser P.H.B. 2010. Functional-structural plant modelling: a new versatile tool in crop science. Journal of Experimental Botany, 61: 2101-2115.

Wang D., Wilson C., Shannon M.C. 2002. Interpretation of salinity and irrigation effects on soybean canopy reflectance in visible and near-infrared spectrum domain. International Journal of Remote Sensing, 23: 811-824.

Wang Q., Adiku S., Tenhunen J., Granier A. 2005. On the relationship of NDVI with leaf area index in a deciduous forest site. Remote Sensing of Environment, 94: 244-255.

Weng J.H., Chen Y.N., Liao T.S. 2006. Relationships between chlorophyll fluorescence parameters and photochemical reflectance index of three species adapted to different temperature regimes. Functional Plant Biology, 33: 241–246.

Werner C., Ryel R.J., Correia O., Beyschlag W. 2001. Structural and functional variability within the canopy and its relevance for carbon gain and stress avoidance. Acta Oecologica, 22: 129-138.

Zarco-Tejada P.J., Miller J.R., Mohammed G.H., Noland T.L. 2000. Chlorophyll Fluorescence Effects On Vegetation Apparent Reflectance: I. Leaf-Level Measurements And Model Simulation. Remote Sensing of Environment, 74: 582–590.

Zheng B., Shi L., Ma Y., Deng Q., Li B., Guo Y. 2008. Comparison of architecture among different cultivars of hybrid rice using a spatial light model based on 3-D digitizing. Functional Plant Biology, 35: 900-910.

Zimmermann D., Reuss R., Westhoff M., Geßner P., Bauer W., Bamberg E., Bentrup F.-W., Zimmermann U. 2008. A novel, non-invasive, online-monitoring, versatile and easy plant-based probe for measuring leaf water status. Journal of Experimental Botany, 59: 3157–3167.

Zotz G., Reichling P., Valladares F. 2002. A simulation study on the importance of size and related changes in leaf morphology and physiology for carbon gain in an epiphytic Bromeliad. Annals of Botany, 90: 437-443.

Summary

A greenhouse crop can be approached as an open system that can be affected by a number of parameters such as light, climate or nutrient supply. In the last decades efforts have been made to understand the functioning of this system and the interaction between the different parameters. The intensive nature of greenhouse cultivation combined with the economic necessity to enlarge the farm size makes the development of decision support systems (DSS) imperative to help the growers in managing their farms efficiently. The foundation of DSS are plant models and in order to work more efficiently they should be able to receive information in real time from sensors that measure different plant parameters such as light interception, leaf area index and photosynthetic stress in a non-destructive way. In order to develop functional DSS it is imperative to develop accurate models and monitoring techniques applied in the specific greenhouse environment.

The aim of this thesis was to explore different techniques to simulate and monitor light interception and photosynthesis by a greenhouse grown tomato canopy. Since photosynthesis is directly linked to light absorption we opted to develop a three dimensional model that takes into account the explicit plant architecture. Different methodologies to monitor these physiological properties online by means of remote sensing were also explored.

A number of physiological tomato models have been proposed the last decades, their main challenge being the correct simulation of fruit yield. For this, an accurate simulation of light interception, and thus photosynthesis, is of primary importance. At present most process-based models and the majority of three dimensional models, include simplifications of plant architecture that can compromise the accuracy of light interception simulations and, accordingly, canopy photosynthesis. In **Chapter 2.1** the first steps towards the development of the model are presented. Light interception is highly dependent on the canopy structure, which is affected, among others, by the distance between plant rows, the distance of plants within the row, leaf pruning and crop variety. The model was used to test different crop planting scenarios on their effect on light interception. Light interception from the planting scenarios was then compared with results of a totally homogeneous canopy. Also analysis of differences between manual measurements of leaf length, width, elevation angle and leaf orientation was conducted. Changes of leaf elevation

angles at two different times of the day were also measured. In tomato differences in leaf length, width and elevation angle of the leaves were mainly observed in the upper 90cm of the plant, in the still developing zone. Changes of the architectural characteristics of structural plant characteristics affected directly light interception by the crop canopy. Nevertheless even if plant structure stayed the same, light penetration could easily be manipulated by changing the row spacing in the crop, thus affecting light interception and potentially plant production.

In **Chapter 2.2** the development and calibration of a functional-structural tomato model is fully described. The model was used to investigate the canopy heterogeneity of an explicitly described tomato canopy in relation to temporal dynamics of horizontal and vertical light distribution and photosynthesis under direct and diffuse light conditions. The model consists of an architectural static virtual plant coupled with a nested radiosity model for light absorption and a leaf photosynthesis module. Different scenarios for horizontal and vertical distributions of light interception, incident light and photosynthesis were investigated under diffuse and direct light conditions. Simulated light interception showed a good correspondence to the measured values. Explicitly described leaf elevation angles resulted in higher light interception in the middle of the plant canopy compared to fixed and ellipsoidal leaf elevation angle distribution models, although the total light interception remained the same. The fraction of light intercepted at a north-south orientation of rows differed from an east-west orientation by 10% in winter and 23% on summer days. The horizontal distribution of photosynthesis differed significantly between the top, middle and lower canopy layer. Taking into account the vertical variation of leaf photosynthetic parameters in the canopy, led to ca. 8% increase on simulated canopy photosynthesis.

Manipulation of plant structure can strongly affect light distribution in the canopy and photosynthesis. In **Chapter 2.3** the idea of identifying different plant ideotypes for optimization of light absorption and photosynthesis was explored. Using the functional-structural tomato model presented in the previous chapters, a range of different plant architectural characteristics were tested for two different seasons in order to find the optimal architecture with respect to light absorption and photosynthesis. Sensitivity analyses were carried out for leaf elevation angle, leaf phyllotaxis, leaflet angle, leaf shape, leaflet arrangement and internode length. From the results of this analysis two possible ideotypes were proposed. Increasing light absorption in the top part of the canopy by 25 %, without changing light absorption of the canopy as a whole, augmented photosynthesis by 6 % in winter and decreased it by 7 % in summer. The measured plant structure was already optimal with respect to leaf elevation angle, leaflet angle and leaflet arrangement for both light absorption and photosynthesis while phyllotaxis had no effect. Increasing the length-to-width ratio of leaves by 1.5 or increasing internode length from 7 to 12 cm led to an increase of 7 – 10 % for light absorption and photosynthesis. The most important architectural traits found were the internode length and the leaf shape as they affect vertical light distribution in the canopy distinctly. A new plant ideotype with more spacious canopy architecture due to long internodes and long and narrow leaves led to an increase in photosynthesis of up to 10 %.

In **Chapter 3.1** ways to monitor on-line LAI and PAR interception of the canopy, under greenhouse conditions, through reflectance measurements, were explored. LAI and PAR interception were measured at the same moments as reflectance at six wavelengths in different developmental stages of tomato and sweet pepper plants. Normalized Difference Vegetation Index (NDVI) was calculated. Relationships between the measured parameters were established in experimental greenhouses and subsequently these were tested in commercial greenhouses. The best estimation for LAI and PAR interception was obtained from reflectance at 460nm for both tomato and sweet pepper. The goodness of the fit was validated with data from the commercial greenhouses and was also tested in this study. The divergence of the results from the ones reported from field experiments can be traced back to the special greenhouse environment, where more sources of reflectance are added due to construction parts and a white plastic covered background. Reflectance measurements offer a non- destructive way to estimate PAR interception and LAI (up to the value of 3) in greenhouse production systems. The relationship established between reflectance at 460 nm, PAR interception and LAI for both tomato and sweet pepper, can become a good tool for crop online monitoring in greenhouse conditions. Furthermore if information from reflectance sensors is used as input directly into the crop models, new opportunities for decision support systems in greenhouse production could be opened up.

Photosynthetic stress induced by water deprivation in plants affects a number of physiological processes such as photosynthetic rate, stomatal conductance as well as the operating efficiency of PSII and non- photochemical quenching. Photochemical

Reflectance Index (PRI) is reported to be sensitive to changes of xanthophyll cycle that occur during stress and could possibly be used to monitor changes in the physiological parameters mentioned before. In **Chapter 3.2** the use of PRI as an early photosynthetic stress indicator was evaluated. A water stress treatment was imposed on a greenhouse tomato crop. CO_2 assimilation, stomatal conductance, light and dark adapted fluorescence as well as PRI and relative water content of the rooting medium RWC_s% where repeatedly measured. The same measurements were also performed on well-irrigated plants that acted as a reference. The experiment was repeated in four consecutive weeks. Results showed that PRI can be used as an early stress indicator only when light intensity at crop level was above 700μmol m^{-2} s^{-1}. At lower values of light intensity the relationship of PRI to RWCs% was poor in comparison to photosynthesis or fluorescence parameters that showed a high correlation to RWC_s%. For that reason we can conclude that PRI as water stress indicator cannot be independent of the ambient light conditions and its use can make sense only under conditions of high light.

Finally in **Chapter 4** the main achievements and limitations of this study are discussed and directions for future research are proposed.

Samenvatting

De afgelopen decennia is kennis over het functioneren van kasgewassen en de interactie met teeltfactoren sterk toegenomen. De intensieve teeltwijze in kassen gecombineerd met de schaalvergroting maken de ontwikkeling en toepassing van beslissingsondersteunende systemen, ofwel decision support systemen (DSS) noodzakelijk om telers te ondersteunen bij het verhogen van de doelmatigheid en efficiëntie van het teeltproces. De basis van een DSS wordt veelal gevormd door een model. Om dit model effectief te laten functioneren zou het real time data moeten krijgen van sensoren die plant-parameters meten zoals lichtonderschepping, bladoppervlakte-index en fotosynthetische stress op een niet-destructieve wijze. Dus om een goed functionerende DSS te ontwikkelen moeten nauwkeurige modellen en monitoring technieken voor toepassing in kassen, ontwikkeld worden.

Het doel van dit proefschrift was om verschillende methodologieën te onderzoeken ten behoeve van het simuleren en monitoren van lichtonderschepping en fotosynthese van een tomatengewas in de kas. Aangezien fotosynthese direct gekoppeld is aan lichtonderschepping, is er voor gekozen om een functioneel-structureel model te ontwikkelen. Een functioneel-structureel model is een model dat de drie-dimensionale plant architectuur expliciet in beschouwing neemt evenals de belangrijkste fysiologische processen. Verschillende methodologieën om fysiologische eigenschappen on-line en contactloos te meten zijn onderzocht.

Afgelopen decennia zijn enkele fysiologische groeimodellen voor tomaat ontwikkeld om de vruchtproductie te kunnen simuleren. Hierbij zijn een nauwkeurige simulatie van lichtonderschepping en fotosynthese van primair belang. Momenteel bevatten de meeste proces-gebaseerde modellen en zelfs de drie-dimensionale modellen sterke vereenvoudigingen van de plantarchitectuur die een negatieve invloed hebben op de nauwkeurigheid van de simulatie van lichtonderschepping en als gevolg daarvan van gewasfotosynthese.

In **Hoofdstuk 2.1** worden de eerste stappen gezet om een functioneel-structureel tomatenmodel te ontwikkelen. Lichtonderschepping is sterk afhankelijk van de gewasstructuur. De gewasstructuur hangt onder andere af van de afstand tussen de plantrijen, de afstand tussen de planten in de rij, bladsnoei en ras. Het model werd gebruikt om het effect van verschillende scenario’s voor plantafstanden op lichtonderschepping te testen. Lichtonderschepping van deze scenario’s werd vergeleken met een volledig homogeen gewas zonder plantrijen. De bladhoek bleek tijdens de dag te veranderen . Deze verandering tijdens de dag werden vooral waargenomen in de bovenste 90cm van de plant; dit is in het deel van de plant dat zich nog aan het ontwikkelen is. Veranderingen van de plant architectuur hadden direct effect op de lichtonderschepping van het gewas. Desalniettemin, zelfs als de plant structuur hetzelfde bleef, kon de lichtdoordringing eenvoudig veranderd worden door bijvoorbeeld de rijafstand te veranderen.In **Hoofdstuk 2.2** wordt de ontwikkeling en calibratie van een functioneel-structureel tomaten model volledig uitgewerkt. Met het model werd de heterogeniteit van horizontale en verticale lichtverdeling en fotosynthese in een tomatengewas onderzocht voor zowel direct als diffuus licht. Het model bestond uit een statisch architectuurmodel, gekoppeld aan een ‘nested radiosity’ model voor lichtabsorptie en een bladfotosynthesemodel. Verschillende scenario’s voor horizontale en verticale verdeling van de lichtonderschepping, invallend licht en fotosynthese werden onderzocht voor direct en diffuus licht. Gesimuleerde lichtonderschepping vertoonde een goede overeenkomst met gemeten waarden. Een expliciete beschrijving van bladhoeken in het model resulteerden in hogere lichtonderschepping in het midden van het gewas in vergelijking met een vaste of ellipsoïdale bladhoekverdeling. De totale lichtonderschepping van het gewas bleef wel gelijk. De fractie van het licht dat onderschept werd door het gewas was bij een noord-zuid oriëntatie van de gewasrijen in de winter 10% en in de zomer 23% hoger dan bij oost-west oriëntatie. De horizontale verdeling van fotosynthese verschilde significant tussen boven, midden en onder in het gewas. Door in het model rekening te houden met de verticale verdeling van bladfotosyntheseparameters in het gewas, veranderde de gesimuleerde gewasfotosynthese met circa 8%. Het veranderen van de gewasstructuur kan sterke invloed hebben op de lichtverdeling in het gewas en de fotosynthese. In **Hoofdstuk 2.3** zijn plant ideotypes gezocht met een optimale lichtabsorptie en fotosynthese. Met behulp van het functie-structuurmodel voor tomaat zoals in vorige hoofdstukken beschreven, zijn effecten van een groot aantal architectuureigenschappen van de plant onderzocht op de lichtabsorptie en fotosynthese in twee verschillende seizoenen (zomer en winter). Er is een gevoeligheidsanalyse uitgevoerd voor bladhoek van het blad en van deelblaadjes, fyllotaxie, lengte/breedte verhouding van bladeren, opbouw van het blad uit deelblaadjes en internodiumlengte. Naar aanleiding van de analyse van deze resultaten werden twee mogelijke ideotypes voor optimale lichtonderschepping en fotosynthese geformuleerd. Als de verticale verdeling van het geabsorbeerde licht in het gewas werd veranderd waarbij de lichtabsorptie in de top van het gewas met 25% toenam terwijl de totale licht absorptie van het gewas gelijk gehouden werd, dan nam de gewasfotosynthese met 6% toe in de winter, maar nam af met 7% in de zomer. De waargenomen plantstructuur was al optimaal voor lichtabsorptie en fotosynthese, voor zover het ging om de volgende architectuureigenschappen: bladhoek van blad en deelblaadjes en opbouw van het samengestelde blad. De meest belangrijke architectuur eigenschappen van een plant bleken de internodiumlengte en lengte-breedte verhouding van het blad. Verhoging van de lengte-breedte verhouding van bladeren met 50% of vergroten van internodiumlengte van 7 naar 12 cm leidde tot een toename van lichtabsorptie en fotosynthese met 7 tot 10%. Een nieuw plant ideotype met een meer open gewasstructuur als gevolg van langere internodiën en lange smalle bladeren leidde tot een toename van de gewasfotosynthese tot 10%.

In **Hoofdstuk 3.1** zijn methoden onderzocht om onder kascondities de bladoppervlakte-index (LAI) en lichtonderschepping van het gewas on-line te monitoren met behulp van reflectiemetingen. LAI en lichtonderschepping werden tegelijk met reflectie van 6 verschillende golflengten gemeten aan paprika- en tomatenplanten in verschillende ontwikkelingsstadia. Relaties tussen de gemeten parameters werden bepaald in onderzoekskassen en vervolgens werden deze relaties getest in praktijkkassen. De beste schatting voor LAI en lichtonderschepping werd verkregen met behulp van reflectie van 460nm licht voor zowel tomaat als paprika. Reflectiemetingen bieden hiermee een goede niet-destructieve mogelijkheid om LAI (tot een waarde van circa 3) en lichtonderschepping te monitoren bij de teelt van tomaten en paprika in kassen. Fotosynthetische stress geïnduceerd door watertekort in planten heeft invloed op een aantal fysiologische processen, zoals fotosynthesesnelheid, huidmondjesgeleidbaarheid, efficiëntie van PSII en niet-fotochemische doving (non-photochemical quenching). De fotochemische reflectie index (PRI) is gevoelig voor veranderingen in de xanthofyl cycli die optreden tijdens stress. Dit zou mogelijk gebruikt kunnen worden om veranderingen in fysiologische parameters te kunnen monitoren. In **Hoofdstuk 3.2** werd het gebruik van de PRI meting geëvalueerd als vroegtijdige indicator voor fotosynthetische stress. Een behandeling met watertekort werd aangelegd in een kas met een tomatengewas. CO_2 mol mμassimilatie, huidmondjesgeleidbaarheid, licht en donker geadapteerde fluorescentie alsmede PRI en relatief watergehalte van het wortelmedium (RWC%) werden gemeten. Dezelfde metingen werden verricht aan goed geïrrigeerde planten die dienden als referentiebehandeling. Het experiment werd herhaald in vier opeenvolgende weken. De resultaten lieten zien dat PRI alleen gebruikt kan worden als vroegtijdige stressindicator wanneer lichtintensiteit meer dan 700 $^{-2}$ s^{-1} bedraagt. Bij lagere lichtintensiteiten was de relatie tussen PRI en RWC% zwak in vergelijking met de goede relaties tussen RWC% en fotosynthese of fluorescentieparameters. Op basis van deze resultaten kan geconcludeerd worden dat PRI als stressindicator niet onafhankelijk van lichtniveau gebruikt kan worden. PRI metingen kunnen alleen als stressindicator gebruikt worden bij voldoende hoge lichtniveaus.

Tenslotte worden in **Hoofdstuk 4** de belangrijkste resultaten en de beperkingen van dit onderzoek bediscussieerd en worden richtingen voor vervolgonderzoek voorgesteld.

Περίληψη

Η καλλιέργεια του θερμοκηπίου μπορεί να προσεγγιστεί ως ένα ανοικτό σύστημα το οποίο μπορεί να επηρεαστεί από μια σειρά παραμέτρων όπως το φως, το κλίμα ή η παροχή θρεπτικών συστατικών. Τις τελευταίες δεκαετίες έχουν καταβληθεί προσπάθειες για την κατανόηση της λειτουργίας του συστήματος αυτού καθώς και της αλληλεπίδρασης μεταξύ των διαφόρων παραμέτρων. Η εντατική φύση της καλλιέργειας του θερμοκηπίου σε συνδυασμό με την οικονομική ανάγκη να διευρυνθεί το μέγεθος της γεωργικής εκμετάλλευσης καθιστά επιτακτική την ανάπτυξη συστημάτων υποστήριξης αποφάσεων (Desicion Support Systems), για την πιο αποτελεσματική διαχείριση των αγροκτημάτων. Βάση των DSS αποτελούν τα μοντέλα φυτών (plant models). Τα μοντέλα αυτά προκειμένου να είναι αποτελεσματικά, θα πρέπει να μπορούν να λαμβάνουν πληροφορίες σε πραγματικό χρόνο από αισθητήρες οι οποίοι μετρούν διαφορετικές παραμέτρους όπως η πρόσληψη φωτός, ο δείκτης της φυλλικής επιφάνειας ή το φωτοσυνθετικό στρες με μη καταστροφικό τρόπο. Για την ανάπτυξη λειτουργικών DSS είναι επιτακτική ανάγκη να αναπτυχθούν ακριβή μοντέλα και τεχνικές παρακολούθησης που μπορούν να εφαρμόστούν στο συγκεκριμένο θερμοκηπιακό περιβάλλον.

Σκοπός αυτής της διατριβής ήταν να διερευνήσει διαφορετικές τεχνικές για την προσομοίωση και την παρακολούθηση της πρόσληψης του φωτός και της φωτοσύνθεσης σε μια καλλιέργεια θερμοκηπιακής ντομάτας. Δεδομένου ότι η φωτοσύνθεση είναι άμεσα συνδεδεμένη με την πρόσληψη του φωτός, στα πλαίσια αυτής της μελέτης επιλέχτηκε η ανάπτυξη ενός τρισδιάστατου μοντέλου ντομάτας που θα λαμβάνει υπόψιν του τη ρητή αρχιτεκτονική των φυτών. Σε δεύτερο πλάνο, διευρήνθηκαν και αξιολογήθηκαν κάτω από τις ειδικές συνθήκες του θερμοκηπίου διαφορετικές μεθοδολογίες για την παρακολούθηση των φυσιολογικών παραμέτρων των φυτών σε πραγματικό χρόνο, μέσω τηλεπισκόπησης,.

Τις τελευταίες δεκαετίες έχουν προταθεί διάφορα φυσιολογικά μοντέλα ντομάτας, όπου η βασική πρόκληση που αντιμετωπίζουν είναι η σωστή προσομοίωση της απόδοσης της καλλιέργειας. Γι 'αυτό, μια ακριβής προσομοίωση της πρόσληψης φωτός, και συνεπώς, και της φωτοσύνθεσης, είναι πρωταρχικής σημασίας. Προς το παρόν τα περισσότερα μοντέλα περιλαμβάνουν απλουστεύσεις της αρχιτεκτονικής του φυτού που θέτει σε κίνδυνο την ακρίβεια της προσομοίωσης της πρόσληψης του

φωτός και, κατά συνέπεια της φωτοσύνθεσης. Στο **Κεφάλαιο 2.1** παρουσιάζονται τα πρώτα βήματα για την ανάπτυξη του μοντέλου. Η πρόσληψη του φωτός εξαρτάται σε μεγάλο βαθμό από τη δομή του φυλλώματος, και επηρεάζεται, μεταξύ άλλων, από την απόσταση μεταξύ των σειρών των φυτών, την απόσταση των φυτών στο εσωτερικό της σειράς, το κλάδεμα των φύλλων και την φυτική ποικιλία. Το μοντέλο χρησιμοποιήθηκε για τη μελέτη της επίδρασης διαφόρων σεναρίων φύτευσης στην πρόσληψη του φωτός. Στη συνέχεια πραγματοποιήθηκε σύγκριση αυτών των σεναρίων με μια καλλιέργεια με ομοιογενές φύλλωμα. Πραγματοποιήθηκε επίσης ανάλυση μετρήσεων του μήκους των φύλλων, του πλάτους, της γωνίας ανύψωσης και του προσανατολισμού των φύλλων. Η μέτρηση της μεταβολής της γωνίας ανύψωσης των φύλλων πραγματοποιήθηκε για δύο ώρες της ημέρας. Στη ντομάτα οι διαφορές στο μήκος των φύλλων, το πλάτος και τη γωνία ανύψωσης παρατηρήθηκαν κυρίως στα άνω 90 εκατοστά του φυλλώματος, στη ζώνη δηλαδή του φυτού που συνεχίζει να αναπτύσσεται. Αλλαγές των αρχιτεκτονικών χαρακτηριστικών του φυτού επηρεάζουν άμεσα την πρόσληψη του φωτός από το φύλλωμα. Παρ 'όλα αυτά, ακόμη και αν η αρχιτεκτονική δομή του φυτού παρέμενε η ίδια, η πρόσληψη του φωτός θα μπορούσε εύκολα να χειραγωγηθεί από την αλλαγή της απόστασης των γραμμών φύτευσης, επηρεάζοντας έτσι την πρόσληψη του φωτός και, ενδεχομένως, τη φυτική παραγωγή.

Στο **Κεφάλαιο 2.2** παρουσιάζεται η ανάπτυξη και βαθμονόμηση ενός τρισδιάστατου μοντέλου ντομάτας. Το μοντέλο χρησιμοποιήθηκε για να διερευνηθεί η επίδραση της ετερογένειας του φυλλώματος στη δυναμική της οριζόντιας και κάθετης κατανομής του φωτός και της φωτοσύνθεσης, κάτω από συνθήκες άμεσου και διάχυτου φωτισμού. Το μοντέλο αποτελείται από τρία υπομοντέλα: α) ένα στατικό αρχιτεκτονικό μοντέλο, β) ένα μοντέλο για τον υπολογισμό της πρόσληψης του φωτός και γ) ένα μοντέλο υπολογισμού της φωτοσύνθεσης σε επίπεδο φύλλου. Διαφορετικά σενάρια για την οριζόντια και κάθετη κατανομή της πρόσληψης του φωτός, της προσπίπτουσας ακτινοβολίας και της φωτοσύνθεσης διερευνήθηκαν κάτω από συνθήκες άμεσου και διάχετου φωτισμού. Η προσομοίωση της πρόσληψης του φωτός έδωσε μια καλή αντιστοιχία με τις μετρούμενες τιμές. Η ρητή περιγραφή των γωνιών των φύλλων οδήγησε σε υψηλότερη πρόσληψη φωτός σε σύγκριση με μοντέλα που χρησιμοποιούν σταθερές ή ελλειψοειδείς κατανομές για την περιγραφή των γωνιών των φύλλων αλλά η συνολική πρόσληψη φωτός παρέμεινε η ίδια. Το ποσοστό του φωτός που προσλήφθηκε ήταν υψηλότερο κατά 10% το χειμώνα και 23% το καλοκαίρι όταν ο προσανατολισμός των σειρών σποράς ήταν από βορρά προς νότο σε συγκρισή με αυτόν από ανατολή προς δύση. Η οριζόντια κατανομή της φωτοσύνθεσης διέφερε σημαντικά μεταξύ της κορυφής, της μέσης και του χαμηλότερου στρώματος του φυλλώματος.

Η χειραγώγηση της αρχιτεκτονικής δομής του φυτού μπορεί να επηρεάσει σημαντικά την κατανομή του φωτός στο φύλλωμα και τη φωτοσύνθεση. Στο **Κεφάλαιο 2.3** διερευνάται η χρήση φυτικών ιδεοτύπων (ideotypes) για τη βελτιστοποίηση της απορρόφησης του φωτός και της φωτοσύνθεσης. Χρησιμοποιώντας το μοντέλο της ντομάτας που παρουσιάστηκε στα προηγούμενα κεφάλαια, εξετάζεται ένα φάσμα διαφορετικών φυτικών αρχιτεκτονικών χαρακτηριστικών για δύο διαφορετικές εποχές του έτους, ούτως ώστε να οριστεί η βέλτιστη αρχιτεκτονική δομή για την απορρόφηση του φωτός και την φωτοσύνθεση. Συγκεκριμένα μελετήθηκαν οι επιδράσεις της γωνίας ανύψωσης των φύλλων, της φυλλοταξίας, της γωνίας των φύλλαρίων, του σχήματος των φύλλων, της διαρύθμισης των φυλλάριων καθώς και του μήκους του μεσογονατίου διαστήματος. Από τα αποτελέσματα αυτής της ανάλυσης, προέκυψαν δύο πιθανοί ιδεότυποι. Αύξηση της απορρόφησης του φωτός στο πάνω μέρος του φυλλώματος κατά 25%, χωρίς αλλαγή της συνολικής απορρόφησης αύξησε τη φωτοσύνθεση κατά 6% το χειμώνα και την μείωσε κατά 7% το καλοκαίρι. Η αρχική αρχιτεκτονική δομή ήταν ήδη βέλτιστη σε σχέση με τη γωνία ανύψωσης του φύλλου, τη γωνία και τη διαρύθμιση του φυλλαρίου τόσο για την απορρόφηση του φωτός όσο και για τη φωτοσύνθεση, ενώ η φυλλόταξη δεν είχε καμία επίδραση. Η αύξηση του μήκους και πλάτους των φύλλων κατά 1,5 ή η αύξηση του μήκους των μεσογονατίων διαστημάτων από 7 έως 12 cm οδήγησε σε αύξηση κατά 7 - 10% για την απορρόφηση του φωτός και τη φωτοσύνθεση. Τα πιο σημαντικά αρχιτεκτονικά χαρακτηριστικά που διαπιστώθηκαν ήταν το μήκος του μεσογονατίου διαστήματος και το σχήμα των φύλλων που επηρέασαν σημαντικά την κάθετη κατανομή του φωτός στο φύλλωμα. Ο νέος προτεινόμενος ιδεότυπος με πιο ανοιχτό φύλλωμα λόγω αύξησης των μεσογονάτιων διαστημάτων και μακριών και λεπτών φύλλων οδήγησε σε αύξηση της φωτοσύνθεσης μέχρι και 10%.

Στο **Κεφάλαιο 3.1** μελετήθηκαν τρόποι για παρακολούθηση on-line της φυλλικής επιφάνειας και της πρόσληψης του φωτός μέσω μετρήσεων ανάκλασης. Η φυλλική επιφάνεια και η πρόσληψη του φωτός μετρήθηκαν την ίδια στιγμή με την ανάκλαση σε έξι διαφορετικά μήκη κύματος σε διάφορα στάδια της ανάπτυξής φυτών ντομάτας και πιπεριάς και υπολογίστηκε ο Normalized Difference Δείκτης Βλάστησης (NDVI).

Οι σχέσεις μεταξύ των μετρούμενων παραμέτρων που βρέθηκαν στις αρχικές μετρήσεις σε πειραματικά θερμοκήπια, στη συνέχεια εξετάστηκαν για την ακρίβεια τους και σε εμπορικά θερμοκήπια. Την καλύτερη σχέση για την εκτίμηση της φυλλικής επιφάνειας και της πρόσληψης του φωτός έδωσε η ανάκλαση στα 460nm και για τις δύο καλλιέργειες. Η σχέση αυτή επικυρώθηκε και από τα αποτελέσματα των μετρήσεων από τα εμπορικά θερμοκήπια. Αποκλίσεις των αποτελεσμάτων μεταξύ των εμπορικών και των πειραματικών θερμοκηπίων οφείλονται στο ιδιαίτερο περιβάλλον του θερμοκηπίου, όπου η ανάκλαση μπορεί να προέρχεται επίσης από τα κατασκευαστικά μέρη του θερμοκηπίου. Η σχέση μεταξύ της ανάκλασης στα 460 nm, τον δείκτη φυλλικής επιφάνειας και της πρόσληψης του φωτός μπορεί να αποτελέσει ένα καλό εργαλείο για την σε πραγματικό χρόνο παρακολούθηση των καλλιεργειών σε συνθήκες θερμοκηπίου. Επιπλέον, εάν οι πληροφορίες από τους αισθητήρες ανάκλασης χρησιμοποιηθούν απευθείας απο τα φυτικά μοντέλα, θα άνοιξουν νέες προοπτικές στα συστήματα υποστήριξης αποφάσεων στην παραγωγή του θερμοκηπίου.

Το φωτοσυνθετικό στρες που προκαλείται από στέρηση νερού στα φυτά επηρεάζει έναν αριθμό φυσιολογικών διεργασιών όπως ο φωτοσυνθετικός ρυθμός, η στοματική αγωγιμότητα καθώς και η λειτουργική απόδοση του PSII και της μη-φωτοχημικής απόσβεση(NPQ). Ο Φωτοχημικός Δείκτης Ανάκλασης (PRI) φέρεται να είναι ευαίσθητος στις μεταβολές του κύκλου της ξανθοφύλλης, που συμβαίνουν κατά τη διάρκεια του στρες και θα μπορούσε ενδεχομένως να χρησιμοποιηθεί για την παρακολούθηση των αλλαγών στις φυσιολογικές παραμέτρους που αναφέρθηκαν παραπάνω. Στο **Κεφάλαιο 3.2** αξιολογείται η χρήση του PRI ως μια πρώιμη ένδειξη φωτοσυνθετικού στρες. Για τους σκοπούς αυτής της έρευνας υδατικό στρες εφαρμόστηκε σε μια καλλιέργεια ντομάτας. Κατά τη διάρκεια του πειράματος μετρήθηκαν η πρόσληψη του CO_2, η στοματική αγωγιμότητα, ο φθορισμός κάτω από σθνθήκες φωτός ή μη, καθώς και το PRI και η σχετική περιεκτικότητα σε νερό του ριζώματος επί τοις %. Οι ίδιες μετρήσεις διεξήχθησαν επίσης σε καλά ποτισμένα φυτά που ενήργησαν ως σημείο αναφοράς. Το πείραμα επαναλήφθηκε τέσσερις διαδοχικές εβδομάδες. Τα αποτελέσματα έδειξαν ότι το PRI μπορεί να χρησιμοποιηθεί ως μια πρώιμη ένδειξη στρες μόνο όταν η ένταση του φωτός ήταν πάνω από 700μmol m-2 s-1. Σε χαμηλότερες τιμές της έντασης του φωτός η σχέση του PRI με τη σχετική περιεκτικότητα σε νερό του ριζώματος επί τοις % ήταν φτωχή, σε σύγκριση με τη φωτοσύνθεση ή το φθορισμό με τις οποίες έδειξαν υψηλή συσχέτιση. Για το λόγο αυτό μπορούμε να συμπεράνουμε ότι η χρήση του PRI σαν δείκτης φυτικού στρες, δεν μπορεί να είναι ανεξάρτητη από τις συνθήκες φωτισμού του περιβάλλοντος και η χρήση του μπορεί να έχει νόημα μόνο σε συνθήκες υψηλού φωτός.

Τέλος το **Κεφάλαιο 4** συζητιούνται τα κύρια επιτεύγματα και οι περιορισμούς της μελέτης αυτής και προτείνονται κατευθύνσεις για μελλοντική έρευνα.

Acknowledgements

The PhD thesis is a road with a lot of twists and turns that is impossible to complete without the input of many people that are all important in their different and unique way. So at this part of the thesis I would like to thank everyone that was there for me all these years and gave me the strength and support I needed to reach the end of the road.

First of all I would like to thank my promotor and main supervisor prof. dr. Leo F.M. Marcelis for his patience, understanding and the fact that he never gave up hope on my thesis, even when the direction was not clear for me. Although we did not always agree, I want you to know that I appreciate and always will be grateful for all your efforts during these years.

I would also like to thank my co-promotor prof. dr. Olaf van Kooten for being a source of constant support. Meetings with you, however brief, always made me feel confident with my research and ready to solve any problems ahead.

I would like to thank my daily supervisor, dr. Pieter de Visser foremost for being the first to teach me about modelling and the fun it can be. I much enjoyed all discussions with you.

To my former roommates Bram, Julienne, Didi, Fleur, Roberta, Andreas and Pavlos thanks for creating a playground environment for me to work into. To Didi special thanks for all the conversations especially the non scientific ones as they helped me to relax and go back to work with more energy.

I would like to thank my good friend and colleague Gerhard Buck-Sorlin who with his constant support and faith in my helped me a lot at the final stages of this thesis. Gerhard our talks were literately out of this earth.

To all current and former members of the Greenhouse Horticulture thank you for making me feel like home while at work. Time spent with you was always stimulating. Anja thank you for all the good advise you gave me during the years. Steven the collaboration with you was more than productive, it was fun. To current and former colleagues of Horticulture Supply Chains Group thank you for the nice times I had with you at all the outings together. I especially appreciated the challenges to give or attending presentations in your group. Also to the members of the FLOP meeting thank you for all the nice conversations and the good and creative criticism on all my writing efforts. To the members of the VPN group a big thank you for the initiating me to the fun of 3D modelling although I did not manage to participate as much I wanted.

To all people that helped me with my experiments during the years I would like to say a big thank you. Especially to Esther that showed me the importance of being methodical, to Johan and Dick from whom I learned a lot about dutch culture and some not always polite dutch words.

I would also like to thank all the numerous friends I made in Wageningen for making my stay here more pleasant. In particular I would like to thank Dimitrios for introducing me to everyone, to Pavlos and Andreas for creating a greek buble inside the office, Niovi and Ioannis for their frendship. Yiannis thank you for all those escapes from the PhD reality when I most needed it and for your constant encouragement and good advice.

Last but not least I would like to thank my parents and brother for never questioning, always trusting, always hoping, always supporting. Thank you for making me what I am in life today. I would have never made it without you.

Curriculum vitae

Vaia Sarlikioti was born on 19th of May 1981 in Amarousio Attikis, Greece. She attended secondary school at the 1st Lyceum in Palaio Phaliro where she received her diploma in 1999. At the same year she started her studies in University of Thessaly, School of Agricultural Sciences, Department of Agriculture Crop Production and Rural Environment, where she obtained her Bachelor degree in 2004, and graduating first of her year. In 2004 she started her Msc studies in the Master program of Agricultural Engineering and Management of Natural Resources were she graduated in 2006. In May 2006 she started her PhD project in Wageningen University, the Netherlands after receiving a scholarship from the Greek Scholarships Foundation, that focused on modelling and monitoring of canopy light interception and plant stress in greenhouse crops. The project was executed at the institute Wageningen UR Greenhouse Horticulture and at the Horticultural Supply Chains group of Wageningen University. Since April 2011 Vaia works as a post-doc in INRA Avignon France.

List of publications

Peer-reviewed publications:

Buck-Sorlin G, de Visser PHB, Henke M, **Sarlikioti V**, van der Heijden GWAM, Marcelis LFM, Vos J ., 2011. Towards a functional-structural plant model of cut-rose - simulation of light environment, light absorption, photosynthesis and interferences with the plant structure. Annals of Botany, in press

Sarlikioti, V., de Visser, P.H.B., Buck-Sorlin, G.H, Marcelis, L.F.M. How plant architecture affects light absorption and photosynthesis in tomato: Towards an ideotype for plant architecture using a functional-structural plant model. Submitted ***(Chapter 2.3)***

Sarlikioti, V., de Visser, P.H.B., Marcelis, L.F.M., 2011. Exploring the spatial distribution of light interception and photosynthesis of canopies by means of a functional-structural plant model. Annals of Botany, 107:875-883. ***(Chapter 2.2)***

Sarlikioti V., Meinen E.., Marcelis L.F.M., 2011. Crop reflectance as a tool for the online monitoring of LAI and PAR interception in two different greenhouse crops. Biosystems Engineering, 108. :113-120. ***(Chapter 3.1)***

Sarlikioti V., Driever S.M., Marcelis L.F.M., 2010. Photochemical reflectance index as a mean of monitoring early water stress. Annals of Applied Biology, 157: 81-89. ***(Chapter 3.2)***

Conference proceeding and reports:

Sarlikioti V., de Visser P.H.B., Marcelis L.F.M., 2011. Towards a 3D structural tomato model for calculating light interception. Acta Horticulturae, 893:721-728. ***(Chapter 2.1)***

Janssen H.J.J., **Sarlikioti V.,** Gieling Th.H., Meurs E.J.J., Marcelis L.F.M., Van Dugteren J.R. 2008. A prototype sensor for estimating light interception by plants in a greenhouse. Acta Horticulturae, 801: 621-627

Dueck T.A.., Nederhoff E.M., Nieboer S., Scheffers C.P., Steenhuizen, J.W., Chizhmak S., Uenk D., **Sarlikioti V.,** de Visser P.H.B., 2010. Verbetering van de lichtonderschepping in een tomatengewas door aanpassing van de rijstructuur: Effecten van de rijstructuur op lichtverdeling, fotosynthese en productie. 44pp. Rapport GTB-1029. Wageningen UR

PE&RC PhD Education Certificate

With the educational activities listed below the PhD candidate has complied with the educational requirements set by the C.T. de Wit Graduate School for Production Ecology and Resource Conservation (PE&RC) which comprises of a minimum total of 32 ECTS (= 22 weeks of activities)

Review of literature (4.5 ECTS)

- Modelling and remote sensing of canopy light interception and plant stress in greenhouses

Writing of project proposal (4.5 ECTS)

- Modelling and remote sensing of canopy light interception and plant stress in greenhouses

Post-graduate courses (6 ECTS)

- Basic and advanced statistics; PE&RC (2007-2008)
- The art of modelling; PE&RC (2008)

Invited review of (unpublished) journal (2 ECTS)

- Annals of botany: 3D modelling (2010)
- Journal of horticultural science: photosynthesis and biomass production (2011)

Deficiency, refresh, brush-up courses (10.5 ECTS)

- Greenhouse technology (2008)
- Crop ecology (2009)

Competence strengthening / skills courses (3 ECTS)

- The art of writing; WGS (2007)
- Techniques of writing and presenting a scientific paper; WGS (2007)

PE&RC Annual meetings, seminars and the PE&RC weekend (1.8 ECTS)

- PE&RC Weekend (2008)
- PE&RC Day (2007)
- PE&RC Day (2008)
- PE&RC Day (2009)

Discussion groups / local seminars / other scientific meetings (6 ECTS)

- Frontier Literature in Plant Physiology (FLOP)

International symposia, workshops and conferences (7.5 ECTS)

- Hortimodel; Wageningen, the Netherlands (2006)
- Greensys; Quebec, Canada (2009)
- Greensys; Halkidiki, Greece

Funding

Vaia Sarlikioti received a grant from the Greek State Scholarship Foundation for conducting the research described in this thesis. Additionally the research was supported by Powerhouse.

Financial support for the printing of this thesis by Wageningen Univerisity is gratefully acknowledged.

ΕΛΛΗΝΙΚΗ
ΔΗΜΟΚΡΑΤΙΑ
IKY
ΙΔΡΥΜΑ ΚΡΑΤΙΚΩΝ
ΥΠΟΤΡΟΦΙΩΝ
STATE SCHOLARSHIPS
FOUNDATION

powerhouse