Phenotyping in Protected Cropping

Subjects: Horticulture | Plant Sciences | Environmental Sciences

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Protected cropping produces more food per land area than field-grown crops. Protected cropping includes low-tech polytunnels utilizing protective coverings, medium-tech facilities with some environmental control, and high-tech facilities such as fully automated glasshouses and indoor vertical farms. High crop productivity and quality are maintained by using environmental control systems and advanced precision phenotyping sensor technologies that were first developed for broadacre agricultural and can now be utilized for protected-cropping applications. The adoption of climate monitoring and control technologies and precision phenotyping methodologies in protected cropping is required for sustaining future food security and enhancing nutritional quality.

precision phenotyping

protected cropping

sensor technology

1. Introduction

By 2050, the United Nations (UN) projects that that the world's human population will reach 9.7 billion, and an estimated two-thirds of the population will live in urban environments ^[1]. An increased population with the increased frequency of extreme weather events has made food security a critical topic of global concern ^[2]. The UN Food and Agriculture Organization (FAO) has identified three key areas to increase agricultural food security. Two of these key areas are to increase the intensity of farming and to increase innovative technology to make farming more efficient ^[3].

Protected cropping is a cultivation method that greatly improves yields by more intensively producing crops on less land area and is defined as a horticultural growing process that utilizes a structure to protect crops from abiotic and biotic stressors. Protected cropping is also known by other names such as protected cultivation and protected agriculture ^[4]. Protected cropping usually involves various degrees of environmental control (e.g., temperature, humidity, CO₂, and nutrition) to reach the highest yields possible and includes a range of facilities from low-tech shade houses to completely climate-controlled high-tech glasshouses ^{[5][6]} Compared to broadacre agriculture, protected cropping improves crop yields and quality, and resource efficiency; increases resiliency to extreme weather events; and, thus, increases national food security ^[7]. Protected cropping also has the ability to provide food supply year-round independent of the seasonal changes in environmental parameters that drive crop productivity.

Technology across the production chain in the protected-cropping industry is identified as a main influential factor in improving product quality and outcome and is regarded as essential for future food security ^[7]^[8]. These include phenotyping technologies that monitor genotypic expression of plant traits for a suite of goals, such as to inform

breeding programs and plant-health assessments. Phenotyping technology has been developed primarily for field and broadacre agriculture through precision phenotyping ^{[9][10]}. However, these methods can be adapted and adopted for use in protected-cropping facilities. The uptake of these technologies is not yet widespread due to capital investment, added maintenance costs, education about using new technologies, and technology transfer from traditional cropping to the protected-cropping industry ^{[2][11]}. The uptake of climate monitoring and control technology will increase productivity within protected-cropping structures already in existence, and this is part of the key objectives to expand the protected cropping industry ^[2]. The application of precision phenotype monitoring in protected cropping will further increase yields and crop quality across the full range of protected-cropping-facility types.

2. Using Phenotyping to Advance the Protected-Cropping Industry

A crop phenotype is the observable characteristics that are expressed as a result of a genotype's interaction with the environment. Phenotyping is the science of the characterization of the crop's traits such as growth, development, architecture, physiology, health, nutrition, and yield. While many plant phenotyping techniques have been developed for the purpose of breeding, disease detection, and improving stress tolerance for field crops ^[12] ^{[13][14]}, phenotyping is also used to monitor plant health, development, disease manifestation, and fruit quality so that growers can take early action to change abiotic conditions and maximize the yield of crops. Crop phenotyping can optimize breeding programs for new varieties that will produce food of high nutritional and aesthetic quality ^[15]. With appropriate phenotype monitoring, resource-use efficiency can be maximized ^{[16][17]}.

Crop phenotyping is an essential process in vegetable and fruit production and is particularly important to mitigate the impacts of climate change, as new varieties of heat- and drought-tolerant fruit and vegetables will have to be developed in the future to maintain crop yield and quality. Currently, assessment of phenotype characteristics relies largely on visual scoring methods by experts, which is time-consuming and can introduce bias via human error ^[15]. Thus, plant phenotyping has become a field for innovation to hasten breeding programs and quality assessment, pre- and post-harvest. High-throughput phenotyping (HTP) platforms have been developed and are being used to collect data for quantitative studies of complex traits related to the growth, yield, and adaptation to biotic (e.g., disease and insects) and abiotic (e.g., drought and salinity) stress. There are a number of techniques available or in development to assess these traits; however, to properly assess phenotypic expression, it is also important to measure and control the environmental parameters to which a crop is exposed, as phenotyping were developed for broadacre crops, these technologies are being transitioned to protected-cropping facilities in various ways ^[10]. Once systems are developed to streamline precision phenotyping in protected cropping, these data-collection systems can be further integrated into automated control systems that will be able to autonomously make cropmanagement decisions based on the observed plant expression ^{[4][18]}.

3. Environmental Monitoring Is a Prerequisite for Plant Phenotyping

3.1. Phenotypic Plasticity in Response to Environmental Parameters

In addition to crop-related traits, effective phenotyping requires continuous assessment of environmental parameters. The environmental conditions in which a crop is grown must be known and, ideally, controlled for consistency across a crop cycle. The primary environmental conditions that impact crop growth are root-zone temperature, moisture and electrical conductivity (EC), light quality and quantity, air temperature, and relative humidity (RH). While some impacts of these environmental parameters on external and internal phenotypic expression are known ^[5], the field is being explored further as better methods of environmental and phenotypic monitoring become accessible.

3.2. Root-Zone Temperature, Moisture Content, and Electrical Conductivity

Root-zone temperature, moisture and EC play key roles in crop performance. While controlling root-zone temperature is a by-product of maintaining optimal air temperature, substrate moisture and EC are controlled by irrigation and fertilization regimes. Irrigation timing impacts root development in soils and soilless substrates. A moisture-saturated root zone results in poor root development, and if this development is hindered early in the growth of the plant, that plant's ability to produce and bear fruit is reduced because the root structure is not there to maintain it. In the case of hydroponic cultivation, EC is an indirect measure of nutrient availability and is crucial for nutrient uptake, and optimal EC is highly specific to the crop. EC that is higher than necessary results in stagnation of nutrient ions. High EC has been related to blossom end rot in tomatoes, as, with increasing EC, mass flow is reduced due to less water entering the plant roots ^[19]. Very low EC, in the case of hydroponic crops, can lead to root cell rupture due to the large imbalance of osmotic pressure across the root membrane. An example of sensor technology that measures root-zone EC, moisture, and temperature can be found in **Table 1**. Some root-zone temperature, moisture, and EC sensors can be integrated into sophisticated greenhouse and glasshouse control systems to dictate irrigation cycles to optimize plant access to water and nutrients throughout the crop's growth cycle ^[20].

Table 1. Environmental parameters and crop impact, listed with available sensor technology and control mechanisms to maintain optimal conditions. Photo credits: (1) <u>www.grodan.com</u> (accessed on 14 September 2022), (2) <u>www.vaisala.com</u> (accessed on 14 September 2022), (3) <u>www.apogeeinstruments.com</u> (accessed on 14 September 2022), and (4) <u>www.directindustry.com</u> (accessed on 14 September 2022).

Environmental Parameter	Impact on Crop	Sensor	Control Mechanism	Example
Electrical conductivity (EC)	High: Blossom-end rot, nutrient deficiency, and reduced yield. Low: Cell rupture.	Slab or soil EC sensors (usually include temperature and moisture measurements)	Irrigation regimes, pH modification, and EC modification of stock solution	

Environmental Parameter	Impact on Crop	Sensor	Control Mechanism	Example
Root-zone moisture	High: Roots do not develop enough to support a full-grown producing plant. Low: Root die-off and plant dehydration.	Soil-moisture probes or slab or soil EC sensors (usually include temperature and moisture measurements)	Properly timed irrigation and proper landscaping to prevent pooling (slope)	
Root-zone temperature	High: >25 °C, NH ₄ toxification, leading to cell death. Low: 3–11 °C, NH ₄ uptake stimulates plant growth.	Soil-temperature and moisture probes that include EC measurements	Shade cloth, irrigation solution temperature, heating pad, and heating cables	
Air temperature	High: Leaf dehydration and earlier stomatal shutdown. Metabolic shutdown due to inability to dissipate heat. Low: Delayed blooming and stunted or slow growth. Large day–night temperature differentials impact fruit set.	Dual air-temperature and relative-humidity probes	Pad and fan cooling, cold-coil fan cooling, shade cloth to reduce radiant heat, hot-water pipes, and hot air via external heat source	2
Relative humidity	High: Low stomatal conductance, reducing nutrient distribution to plant and fruit. Low: Early stomatal shutdown, resulting in reduced photosynthesis.	Dual air-temperature and relative-humidity probes	Misting system, condensing system, and dehumidification	
Light quality	280 nm: Reduces quantum yield and rate of photosynthesis. 315–400 nm: Promotes pigmentation and thickens plant leaves. 400–440 nm: Promotes vegetative growth. 640–660 nm: Vital for flowering.	Spectroradiometer or a combination of PAR and net radiometer	Colored shade cloth, fluorescent films, and light supplementation	

Environmental Parameter	Impact on Crop	Sensor	Control Mechanism	Example
	740 nm: Increases photosynthesis ^[21] .			
Light quantity	High: Leaf dehydration, sunscald, photodamage, and lowered photosynthetic rates. Low: Stem elongation, lower photosynthetic rate, reduced yield, misshapen fruit, and reduced shelf life.	PAR sensors	Shade cloth and light supplementation with light-emitting diodes	4

harvested ^[23]. Misshapen, swollen, and hollow tomato formations have also been documented as a result of low light conditions ^[22]. In contrast, a higher irradiance from the sun results in leaf dehydration and premature shutdown, leading to a reduced photosynthetic capacity and lower crop yield. Excess light can lead to sunscald in a wide range of crops, including tomatoes and capsicum ^[22]. In lettuce, high light environments can lead to the increased presence of tip burn ^[24]. Light quality also influences internal and external phenotypic expression. Plants primarily use photosynthetically active radiation (PAR), which consists of wavelengths from 300 nm to 700 nm. Red light exposure has been shown to reduce the bitter flavor in lettuce leaves ^[25], while UV-B exposure causes the accumulation of secondary metabolites, which influence a variety of plant physiological processes and internal quality of some vegetables ^[22]. In protected cropping, PAR sensors or global radiation sensors are used both outside and inside the growth compartments to monitor light levels (**Table 1**). These measurements can be and often are integrated into shade curtain control systems to ensure that crops are receiving appropriate light conditions throughout the day ^[26].

Light quality also plays a key role in taste and aroma profiles for different fruiting or vegetative plants. Light quality, intensity, and duration can be manipulated to elicit desired attributes of leafy greens, including leaf shape, leaf color, nutrition, flavor, texture, and aroma ^[27].

3.4. Temperature and Relative Humidity (RH)

Temperature and RH are important environmental variables that must be maintained for proper crop development and to achieve maximum yield. With an increase in temperature, RH decreases, as RH is a measure of the percentage of moisture the air can hold as vapor at that particular temperature. As temperature increases, the air's ability to hold more moisture increases. Crops also have different microclimate needs at different stages of development; thus, continuous measurement is necessary to maintain these parameters, which can be performed with a variety of sensors, an example of which can be found in **Table 1**. While these temperature and RH sensors are used to continuously monitor the microclimate, they are also the main trigger for misting/dehumidifying and heating/cooling systems to maintain optimal RH and temperatures, and they are essential tools for maintaining high crop yield and quality ^[28].

While optimal temperatures for day and night will vary with crop type and variety, the majority of horticultural crops are warm-weather plant species, with their main growing seasons occurring over the summer months. Typically, temperatures between 20 and 30 °C and with an RH of 50 to 80% are desirable; however, each crop variety has specific requirements. Cucumbers, for instance, have been shown to taste sweeter when grown in moderate RH as opposed to high RH and, in general, produce better fruit with respect to quality attributes and external appeal ^[29]. While air temperature is an important parameter, studies have shown that canopy (leaf) temperature may be far more important. Leaf temperature is directly related to transpiration rates and measurement of this parameter provides information about leaf temperature, as well as plant water status ^[30].

3.5. Sensor Technology to Monitor Environmental Parameters

Consistent environmental conditions are essential for reproducible phenotypic responses from different genotypes. Therefore, if environmental conditions are tightly controlled, ideal plant growth and development can be achieved; thus, developmental stages can be identified and targeted to identify specific traits. Moreover, in tightly controlled conditions, deviations from typical phenotypic expression become more obvious and make assessing disease, plant health, and fruit quality more straightforward. There are various sensors available to monitor air temperature, RH, root-zone temperature, moisture, and EC, as well as light quantity and quality (**Table 1**). While all of these parameters, such as shade curtains or irrigation, can be monitored individually via computer or smartphone and the control mechanism changed manually, there are integrated hardware and software systems available that can automatically manage these environmental controls. Due to the variability of microclimates within protected-cropping facilities, high-resolution measurements are useful for correctly informing the grower on the overall environment that the crop is experiencing; thus, they can make more appropriate management decisions ^{[31][32]}.

3.6. Environmental Monitoring Is a Prerequisite for Plant Phenotyping

In summary, plants have plasticity to alter their phenotype, exhibiting different external and internal expression depending on the environment. Environmental control and data capture are important throughout the phenotyping process, as antecedent events can be inherited by the progeny, which is known as the 'memory effect' ^[33]. Frequent high-throughput phenotyping will promote faster phenotypic data acquisition for correlation with genomic information ^[34]. In protected cropping, phenotypic expression dictates fruit aesthetic and nutritional quality pre- and post-harvest, suggesting that two important subjects need to be considered:

- Precise control over crop microclimate to maintain desired phenotypic expression across crop cycles;
- Frequent phenotypic surveys of plants and fruit, throughout the cropping cycle and during post-harvest sorting, storage, and distribution.

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