Using Color Infrared Imagery to Detect Sooty Mold and Fungal Pathogens of Glasshouse-propagated Plants

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Additional index words. spectroradiometer, color infrared (CIR) imagery, simple vegetation index, citrus (C. sinensis), sooty mold (C. fuliginea), greasy spot (Mycosphaerella citri), powdery mildew (Sphaerotheca fuliginea)

Abstract. Fungi are major biotic constraints for optimum production and quality of glasshouse plants. When plants are infested with sooty mold (C. fuliginea) or infected with pathogens, the reflected wavelengths of the electromagnetic spectrum are altered. Spectroradiometric measurements and color infrared (CIR) images of control, honeydew-coated, and sooty mold-infested saplings and individual leaves from trifoliate orange (P. trifoliata), sour orange (C. aurantium), ‘Valencia’ orange (C. sinensis), and ‘Bo’ tree (Ficus religiosa) were obtained. Grapefruit saplings and individual leaves infected with Mycosphaerella citri (greasy spot) were imaged under glasshouse conditions. Similarly, muskmelon foliage showing low and high levels of powdery mildew (Sphaerotheca fuliginea) disease severity were analyzed. When examining individual leaves, all fungal biotic stressors generally resulted in variable spectral reflectance data, especially in the blue (450 nm) and green (550 nm) wavelengths; however, values in the red (650 nm) tended to increase and values in the near-IR (850 nm) tended to decrease with stress. Near-IR/red image ratios were significantly reduced (P < 0.05) in stressed whole plant foliage and individual leaves relative to healthy controls. The accumulation of insect honeydew (which occurs before sooty mold infestation) significantly increased (P < 0.05) near-IR reflectance values and near-IR/red ratios in ‘Valencia’ orange and near-IR/ratios in ‘Bo’ tree foliage and individual leaves. Image acquisition and enhancement techniques may prove useful in large-scale production greenhouses where existing infrastructure and high plant populations require high throughput data analysis and identification of biotic stressors.

Remote sensing has been used for the last several decades to detect both abiotic and biotic stressors affecting agricultural crops. Studies have traditionally focused on aerial image acquisition using color infrared (CIR) photography of plant stress, including salinity problems (Everitt et al., 1981; Myers et al., 1963), nutrient deficiencies (Thomas et al., 1989), insect predation (Blazquez et al., 1988; Everitt et al., 1994), and pathogens (Brenchley, 1964; Colwell, 1956; Hart et al., 1973; Hart and Myers, 1968), and mealy bugs (Hemiptera: Pseudococcidae) (Drees and Jackman, 1998). All of these stressors are common in glasshouse environments. In addition, because CIR images can be subdivided into three separate wavebands [green (500 to 600 nm), red (600 to 700 nm), and near-IR (NIR; 700 to 1100 nm)], the use of image enhancement techniques that rely on these, such as unsupervised/supervised image classification and image ratios [such as the simple vegetation index (NIR/Red)], have proven to be reliable approaches for improving the data acquired from CIR images and their subsequent interpretation (Little et al., 2005; Summer and Little, 2005; Summy et al., 2003, 2004, 2005; Yousef et al., 2005). Sooty mold (Figs. 1B and 1E) is a common problem in which excessive insect feeding activities occur. The condition results from honeydew deposition (Fig. 1D) by insects such as whiteflies (Hemiptera: Aleyrodidae), aphids (Hemiptera: Aphididae), and mealy bugs (Hemiptera: Pseudococcidae) (Drees and Jackman, 1998). All of these insect groups are common in the glasshouse; however, specific examples differ depending on plant host and geographic location. Ultimately, all of these insects are capable of producing large amounts of honeydew excretions. These excretions are composed of numerous sugars capable of supporting sooty mold growth, which consists primarily of Sphaerotheca and related fungal species (Farr et al., 1989; Reynolds, 1999).

In addition to sooty mold accumulation, there are a myriad of fungal leaf spots that occur on glasshouse plant foliage and it would be impossible to investigate all of them here. For reviews of leaf spot diseases of ornamentals, flowering potted plants, tomatoes, citrus, and cucurbits, see Chase (1987), Daughtrey et al. (1995), Jones et al. (1991), Timmer et al. (2000), and Zitter et al. (1996), respectively. Most of the common leaf spots will cause significant tissue chlorosis that either surrounds the necrotic lesions or extends throughout the affected foliage. In the present study, citrus plants that were propagated under glasshouse conditions were infected by Mycosphaerella citri, an ascocytetous causing “greasy spot” (Fig. 1G) (Farr et al., 1989; Timmer et al., 2000). This disease produces characteristic black fruiting structures (pseudothecia) in leaf tissues, whole leaves become chlorotic, and defoliation of saplings or plant grafts can be common.

Powdery mildews are caused by cleistothecial ascomycetes and are characterized by the orange production of conidia, conidio- phores, and hyphae that appear white on the upper surface of plant leaves (Fig. 1I). The primary examples of fungal species causing powdery mildew of dicot hosts in greenhouses include Erysiphe cichoracearum (cucurbits and many flowers), E. polygoni (numerous vegetables and hydrangeas), and Sphaerotheca fuliginea (cucurbits; discussed in this study) (Farr et al., 1989; Texas Agricultural Experiment Station, 1988).

The purpose of this study was to acquire spectral reflectance data and CIR imagery from plants produced under glasshouse conditions and affected by the following fungal stressors: 1) sooty mold infestation, 2) a foliar leaf spot disease accompanied by extensive tissue chlorosis, and 3) a powdery mildew disease. Additionally, CIR image wavebands were used to construct NIR/red derivative image ratios to facilitate interpretation of treatment differences.

Materials and Methods

Plant propagation and fungal stressors. Trifoliate orange (P. trifoliata), ‘Valencia’ orange (C. sinensis), sour orange (C. aurantium), grapefruit (C. paradisi), and ‘Bo’ tree (Ficus religiosa) plants
were grown at the Texas A&M University–Kingsville Citrus Center (Weslaco, TX). Muskmelon (*Cucumis melo*) plants were grown at the Texas Agricultural Experiment Station (Weslaco, TX). “Control” cohorts were not infested with sooty mold, insects, or infected with pathogens. “Treatment” groups were naturally infested with sooty mold (*Capnodium citri*), insects, and infected with pathogens (*Sphaerotheca fuliginea* or *Mycosphaerella citri*) under routine glasshouse-growing conditions.

**Acquisition of spectral reflectance data and color infrared imagery.** Spectral reflectance data were acquired from incident radiation reflected from foliage in a whole plant context and from individual leaves (see below) using a FieldSpec® VNIR spectroradiometer equipped with a remote cosine receptor (Analytical Spectral Devices [ASD], Boulder, CO). At the time of data collection, the instrument was optimized for ambient lighting conditions and a reflectance measurement was obtained from a white Spectralon® reference plate (ASD) to facilitate the conversion of radiance measurements (watts/m²) to percent reflectance. Reflectance measurements were obtained using a target probe fitted with an 18° instantaneous field of view adapter and data were processed using ViewSpecPro® software (ASD). CIR images of whole plant foliage and individual leaves were acquired using a conventional 35-mm SLR camera (Nikon FE) (Nikon, Melville, NY) employing Kodak Ektachrome Infrared EIR film (Eastman Kodak, Rochester, NY) fitted with a 55-mm lens and a Wratten 15 yellow filter (Tiffen, Hauppauge, NY). Conventional color (RGB) photographs were obtained using a digital camera (Sony Mavica® 400) (Sony Electronics, San Diego, CA).

Color RGB and CIR images of whole plant foliage were obtained in situ from trifoliate orange, sour orange, ‘Valencia’ orange, and ‘Bo’ tree saplings within a glasshouse composed of yellow fiberglass (Figs. 1A–E). In situ reflectance data and images of muskmelon foliage were obtained under conventional frosted glass panels and grapefruit foliage was obtained under a plastic mesh screenhouse (Figs. 1F–I). For whole plant foliage data, a representative plant for each treatment was selected from a larger cohort of identically stressed plants for imaging.

Individual leaves were excised from saplings of each tree and immediately mounted between glass plates. Representative leaves exhibiting a range of infestations or symptoms were selected for comparison with healthy (control) leaves of the species examined in this study with the exception of *C. melo* (below). Spectral samples from each treatment consisted of multiple spectroradiometer readings (five) of 10 measurements each to obtain a mean of means. For *C. melo*, comparison was only made between healthy (control) and powdery mildew infected leaves in situ only. All photographs for a particular species were completed on the same day (26 Apr. 2005), under sunny conditions, between 1100 and 1700 HR.

For imaging, individual leaves were pressed between two glass plates (0.3 × 22 × 30 cm) to reduce the Gaussian curvature that is often associated with normal leaf structure and accentuated during stress (Nath et al., 2003). This was done to normalize reflectance values over the entire surface of the leaf so that measured differences are due to “treatment” and not leaf structure (Little et al., 2005). Leaves pressed between glass plates were mounted on a black, plastic plant growth flat filled with distilled water to absorb incident light and prevent background reflection in CIR images. These are subsequently referred to as “template images” and were obtained within a greenhouse composed of yellow fiberglass. For more information concerning the effect of different greenhouse materials on spectral reflectance data and...
CIR image acquisition, see Summy et al. (2004).

Analysis of spectral reflectance data and color infrared imagery. Reflectance measurements obtained from spectral curves of individual leaves [see previous use of the spectroradiometer (above)] were displayed and percent reflectance values at four wavelengths [blue (450 nm), green (550 nm), red (650 nm), and NIR (850 nm)] were recorded. The resulting reflectance data were analyzed with analysis of variance (ANOVA) and comparisons made, if appropriate, with Tukey’s least significant difference (LSD) ($P < 0.05$) (Systat v.10; SPSS, 2000) (Table 1).

After processing, CIR images were converted to tagged image format (.TIF) files from slides; separated into green, red, and NIR wavebands; and imported into IDRISI32® (Clark Laboratories, Worcester, MA). A simple vegetative index (NIR/red) was used as an “image ratio” to analyze CIR imagery. Approximately 20% of unrepresented wavelengths was eliminated from derivative ratio images using contrast stretching. Contrast stretched derivative ratio images were layered with a stratified spatial sample of random points to compare healthy (control) and infected or infested regions of whole plant foliage or individual leaves. “Points” derived from derivative images each represent a ratio of NIR to red at a single pixel, which are generated by dividing the entire NIR band of a CIR image by its red band.

Twenty random points were chosen from the top seven internodes (if possible) of citrus plants examined for sooty mold development (Figs. 2-I, 2-II, and 2-III). Image subsamples were obtained from ‘Bo’ tree (Fig. 2-IV) and grapefruit (Fig. 2-V) images; 20 random points were selected from the outlined image subsamples in Figure 2-IV-g–i and Fig. 2-V-j and k (see also Table 2). Additionally, 20 random points were obtained from mature muskmelon leaf ratio images (Fig. 2-VI-I and m; Table 2). Therefore, $n = 20$ for each individual plant within an image (Fig. 2-I, 2-II, and 2-III). Table 1. Mean spectroradiometer reflectance data derived from individual leaves of greenhouse-reared plants infested with sooty mold, common insect pests, or infected with fungal pathogens.

| Treatment                        | Figure | Mean blue (450 nm) | Mean green (550 nm) | Mean red (650 nm) | Mean near-infrared (850 nm) |
|----------------------------------|--------|--------------------|---------------------|-------------------|-----------------------------|
| Trifoliolate orange (Poncirus trifoliata) | 3-I-a  | 2.8 c*             | 6.3 a               | 2.9 c             | 25.9 a                      |
| Control (20 d)                   | 3-I-b  | 2.7 c              | 5.5 a               | 3.0 c             | 24.6 ab                     |
| Sooty mold* (intermediate)       | 3-I-c  | 2.7 c              | 5.1 a               | 3.7 bc            | 21.0 ab                     |
| Sooty mold (heavy)               | 3-I-d  | 4.0 a              | 6.5 a               | 5.5 a             | 19.3 ab                     |
| Sooty mold + mealy bug*-infested | 3-I-e  | 3.1 bc             | 5.0 a               | 3.9 bc            | 17.5 b                      |
| Leaf miner*-infested             | 3-I-f  | 3.4 abc            | 5.5 a               | 3.4 bc            | 17.5 b                      |
| Citrus mite*-infested            | 3-I-g  | 3.6 ab             | 6.1 a               | 4.0 b             | 18.1 ab                     |
| Sour orange (Citrus aurantium)   |        |                    |                     |                   |                             |
| Control (10 d)                   | 3-II-h | 2.5 c              | 12.7 a              | 3.7 b             | 29.0 a                      |
| Control (20 d)                   | 3-II-i | 2.8 bc             | 6.9 b               | 3.0 c             | 32.1 a                      |
| Sooty mold (light)               | 3-II-j | 3.0 c              | 5.6 b               | 3.3 bc            | 24.9 ab                     |
| Sooty mold (intermediate)        | 3-II-k | 3.3 b              | 5.4 b               | 4.2 b             | 19.9 bc                     |
| Sooty mold (heavy, dark)         | 3-II-l | 3.2 bc             | 5.1 bc              | 4.5 b             | 14.3 ed                     |
| Sooty mold (heavy, tan)          | 3-II-m | 2.4 c              | 3.6 c               | 3.6 bc            | 9.0 d                       |
| Valencia orange (C. sinensis)    |        |                    |                     |                   |                             |
| Control (20 d)                   | 3-III-o| 3.2 c              | 11.0 a              | 4.0 b             | 33.9 b                      |
| Honeydew                         | 3-III-p| 3.9 b              | 8.6 b               | 4.5 b             | 40.8 c                      |
| Sooty mold                       | 3-III-q| 6.2 a              | 9.4 b               | 10.0 a            | 16.7 a                      |
| Bo tree (Ficus religiosa)        |        |                    |                     |                   |                             |
| Control                          | 3-IV-r | 3.5 a              | 6.5 a               | 3.6 a             | 33.9 a                      |
| Honeydew                         | 3-IV-s | 3.6 a              | 6.1 a               | 3.3 b             | 34.5 a                      |
| Sooty mold                       | 3-IV-t | 3.8 a              | 5.6 a               | 4.1 a             | 26.1 b                      |
| Grapefruit (C. paradisi)         |        |                    |                     |                   |                             |
| Healthy control                  | 3-V-u  | 2.7 c              | 15.6 c              | 4.4 d             | 41.3 ab                     |
| Low greasy spot severity         | 3-V-v  | 4.1 bc             | 21.5 b              | 9.2 e             | 38.3 b                      |
| Intermediate greasy spot severity| 3-V-w  | 5.5 b              | 22.5 b              | 11.5 b            | 41.3 ab                     |
| High greasy spot severity        | 3-V-x  | 10.3 a             | 33.0 a              | 29.9 a            | 43.2 a                      |
| Muskmelon (Cucumis melo)         |        |                    |                     |                   |                             |
| Control                          | 2-VI-l | 4.4                 | 11.7                | 4.0               | 52.1                        |
| Powdery mildew*                  | 2-VI-m | 5.6*               | 10.5                | 5.5*              | 38.9*                       |

*Leaf age (in days).
*Each mean value is the average of spectral samples from each treatment consisted of multiple spectroradiometer readings (five) of 10 measurements each to obtain a mean of means. Values that are followed by different letters (within the same column for each plant species) differ significantly according to Tukey’s least significant difference ($P < 0.05$); values followed by an asterisk differ significantly ($P < 0.05$) from their respective control according to the Student’s $t$ test.
*Sooty mold [Capnodium spp. of fungi (and others spp.)].
*Mealy bug [Planococcus citri].
*Leaf miner [Phyllocnistis citrella].
*Citrus mite [Eutetranychus citri].
*Greasy spot [Myzopodia phycidaphila].
*Powdery mildew [Sphaerotheca fuliginea].

Results and Discussion

The last Census of Horticultural Specialties (National Agricultural Statistics Service, 1998) indicated that $\approx 31.6$ million ft$^2$ of greenhouse space was used to produce food crops with a wholesale value of $184.2$ million and an investment into the agricultural labor force of $3.6$ billion in the United States alone. In addition to this, much of the production of horticultural and landscaping transplants and vegetable and flower seeds occurs in large-scale greenhouse facilities.

Many studies have evaluated the acquisition of multispectral imagery as a tool to estimate yield responses and plant damage in the field (Yang and Everitt, 2002; Yang et al., 2005). However, very few have demonstrated the technical feasibility of using multispectral CIR imagery for detection of plant stress in the glasshouse (Summy et al., 2003, 2004; Summy and Little, 2005).

In this study, whole plant foliage and individual leaves that were infested with sooty mold and infected with fungal pathogens were imaged using conventional color [RGB; Figs. 1 and 3 (upper rows)] and CIR photography [CIR; Figs. 1 and 3 (middle rows)]. The resulting images were processed so that the NIR waveband of each CIR image was divided by its red waveband to obtain a simple vegetation index derivative image where each pixel represents a ratio of NIR to red [Figs. 2 and 3 (lower rows)]. High NIR and low red reflectance values are typical of healthy vegetation; thus, a ratio of these two values can be used as an indicator of plant status (Everitt et al., 1999). In addition, differences between healthy and stressed foliage can be accentuated and subtle differences more easily identified and analyzed using image ratios (Yousef et al., 2005).

Leaf age influences NIR/red ratio differences and this confounding factor may influence interpretation of whole plant foliage data acquired at close distances. To test the effect of leaf age on acquisition of NIR/red ratio data from CIR images, leaves of varying ages were compared using the template images (described in “Materials and Methods”). Twenty- and 35-d leaves from trifoliolate orange did not differ in NIR (850 nm), red (650 nm), blue (450 nm), or green (550 nm) reflectance values. In addition, there was no difference between NIR/red ratios or observable differences in the derivative ratio images (Table 2; Fig. 3-I). However, sour
orange leaves that differed in ages from 10 to 35 d did not exhibit significantly different red or blue reflectance values, but the NIR/red ratio values and the resultant derivative images were significantly different (Table 2; Fig. 3-II).

This study showed that honeydew accumulation on ‘Valencia’ and ‘Bo’ tree leaves resulted in significant increases in NIR/red ratios (Table 2). In addition, the increased overall reflectance patterns are observable in derivative images (Figs. 3-III and 3-IV). ‘Valencia’ orange leaves were coated very evenly with insect honeydew (Fig. 3-III-p), whereas honeydew deposits on ‘Bo’ leaves were very spotty (Fig. 3-IV-s). This difference in distribution along the leaf surface may have contributed to differences in

Fig. 2. Near-infrared/red ratio derivative images of whole trifoliate orange (I; Poncirus trifoliata), sour orange (II; Citrus aurantium), ‘Valencia’ orange (III; C. sinensis), ‘Bo’ tree (IV; F. religiosa), grapefruit (V; C. paradisi), and muskmelon (VI; Cucumis melo) plants. (I) Trifoliate orange saplings: uninfested control (a), infested with sooty mold (Capnodium citri spp. and other fungi), mealy bugs (Planococcus citri), leaf miners (Phylloncistis citrella), and citrus mite (Eutetranuchus citri) (b); (II) sour orange saplings: uninfested control (c) and sooty mold infected (d); (III) ‘Valencia’ orange saplings: uninfested control (e) and sooty mold infected (f); (IV) ‘Bo’ saplings: uninfested leaf subsample (g), honeydew coated leaf subsample (h), and sooty mold infested leaf subsample (i) (see also Table 1 for data); (V) grapefruit saplings: subsample of noninfected leaves (j; see also Fig. 1F) and a subsample of leaves showing various levels of greasy spot (Mycosphaerella citri) severity (k; see also Fig. 1G); cantaloupe foliage: healthy mature leaf (l; see also Fig. 1H) and mature leaf infected with powdery mildew (Sphaerotheca fuliginea) hyphae and conidia (m; see also Fig. 1I). Images were contrast stretched (see lower right hand corner of each image) where necessary to eliminate unnecessary wavelengths. See also Tables 1 and 2 for spectrophotometric data and near-infrared/red ratios derived from derivative images of foliage in a whole plant context.
Table 2. Mean near-infrared (NIR)/red ratios derived from color infrared derivative images from whole plant foliage and individual leaves of greenhouse-reared plants infested with sooty mold, common insect pests, or infected with fungal pathogens.

| Treatment                        | Figure | Mean NIR/red* |
|----------------------------------|--------|---------------|
| **Trifoliate orange (Poncirus trifoliata)** |        |               |
| Whole plant                      |        |               |
| Control                          | 2-I-a  | 5.78          |
| Sooty mold + insect infestations | 2-I-b  | 4.21*         |
| Individual leaves                |        |               |
| Control (20 d)                   | 3-I-a  | 10.20 a       |
| Control (35 d)                   | 3-I-b  | 9.50 ab       |
| Sooty mold (intermediate)        | 3-I-c  | 7.79 b        |
| Sooty mold (heavy)               | 3-I-d  | 4.64 d        |
| Sooty mold + mealy bug-infested  | 3-I-e  | 6.20 cd       |
| Leaf miner-infested              | 3-I-f  | 7.58 c        |
| Citrus mite-infested             | 3-I-g  | 5.74 d        |
| **Sour orange (Citrus aurantium)** |        |               |
| Whole plant                      |        |               |
| Control                          | 2-II-c | 4.63          |
| Sooty mold                       | 2-II-d | 4.19*         |
| Individual leaves                |        |               |
| Control (10 d)                   | 3-II-h | 6.44 c        |
| Control (20 d)                   | 3-II-i | 10.40 a       |
| Control (35 d)                   | 3-II-j | 8.85 b        |
| Sooty mold (light)               | 3-II-k | 7.54 bc       |
| Sooty mold (intermediate)        | 3-II-l | 6.11 c        |
| Sooty mold (heavy, dark)         | 3-II-m | 3.05 d        |
| Sooty mold (heavy, tan)          | 3-II-n | 2.69 e        |
| **Valencia orange (C. sinensis)** |        |               |
| Whole plant                      |        |               |
| Control                          | 2-III-e| 6.27          |
| Sooty mold                       | 2-III-f| 3.49*         |
| Individual leaves                |        |               |
| Control (20 d)                   | 3-III-o| 7.22 b        |
| Honeydew                         | 3-III-p| 8.72 c        |
| Sooty mold                       | 3-III-q| 2.40 a        |
| **Bo tree (Ficus religiosa)**    |        |               |
| Whole plant                      |        |               |
| Control                          | 2-IV-g | 4.56 b        |
| Honeydew                         | 2-IV-h | 5.54 a        |
| Sooty mold                       | 2-IV-i | 2.76 c        |
| Individual leaves                |        |               |
| Control                          | 3-IV-r | 6.26 b        |
| Honeydew                         | 3-IV-s | 7.90 a        |
| Sooty mold                       | 3-IV-t | 5.21 b        |
| **Grapefruit (C. paradisi)**     |        |               |
| Whole plant                      |        |               |
| Control                          | 2-V-j  | 6.38          |
| Greasy spot                      | 2-V-k  | 2.70*         |
| Individual leaves                |        |               |
| Healthy control                  | 3-V-u  | 6.43 a        |
| Low greasy spot severity         | 3-V-v  | 4.01 b        |
| Intermediate greasy spot severity| 3-V-w  | 2.89 e        |
| High greasy spot severity        | 3-V-x  | 1.56 d        |
| **Cantaloupe (Cucumis melo)**    |        |               |
| Whole plant                      |        |               |
| Control                          | 2-V-1  | 5.34          |
| Powdery mildew                   | 2-V-m  | 3.90*         |

*Contrast stretched near-infrared/red ratio derivative images were layered with a stratified spatial sample of random points to compare “control” and “treatment” regions of plant foliage or individual leaves. For whole plant foliage near-infrared/red ratio data, 20 random points were chosen from the top seven internodes (if possible) of P. trifoliata, C. aurantium, and C. sinensis plants examined for sooty mold development; image subsamples were used for F. religiosa (Fig. 2-IV-g–j) and C. paradisi (Fig. 2-V and k). For C. melo, “control” and “treatment” regions (Fig. 2-V-1 and 2-V-m, respectively) of plant foliage or individual leaves were compared. Values in columns followed by an asterisk differ significantly (P < 0.05) from their respective control according to the Student’s t test. For individual leaves (see Fig. 3), 10 points were randomly chosen from the stratified spatial sample. Values that are followed by different letters (within the same column for each plant species) differ significantly at P < 0.05 using Tukey’s least significant difference.

*Sooty mold [Cnapsodium spp. of fungi (and others spp.)].

*Leaf age (in days).

*Mealy bug (Planococcus citri).

*Leaf miner (Phylloniscis citrella).

*Citrus mite (Eutetranychus citri).

*Greasy spot (Mycophaeella citri).

*Powdery mildew (Sphaerotheca fuliginea).
wavelength ranges of 300 to 400 nm (ultraviolet), 600 to 700 nm (red), and 700 to 900 nm (NIR).

In this study, symptoms and signs of two foliar fungal pathogens were imaged under glasshouse conditions, including a common powdery mildew of cucurbits (Sphaerotheca fuliginea) and the greasy spot pathogen of citrus (Mycosphaerella citri). M. citri is a foliar pathogen of citrus that causes significant amounts of chlorosis in leaves as part of the disease process. Chlorosis discolors citrus leaves as large numbers of fungal pseudothecia are produced (Mondal and Timmer, 2006). In this disease, as severity increases, red reflectance from the chlorotic portions of affected leaves increases appreciably, but NIR reflectance does not change significantly (Table 1). Because of the relatively steady NIR values, there is a concomitant decrease in the NIR/red ratios (Table 2). Thus, in Fig. 3-V-x, the average NIR/red ratio (1.56), for high greasy spot severity, was coded as black in the derivative image (less than 2.26) to remove background noise (e.g., unnecessary wavelengths; see “Materials and Methods”).

In the case of the powdery mildew fungus, S. fuliginea, leaves are not discolored by chlorosis, but the adaxial surfaces are covered with hyphal growth and numerous conidia. This gives the surface of the leaf a “powdery” appearance (Little, 2004). In some ways, this situation is comparable to the sooty mold examples discussed earlier with the exception that the fungal growth on the surface of the leaf appears white instead of tan, brown, or black. In the case of the cucurbit powdery mildew, NIR/red ratios were significantly reduced as a result of decreased levels of NIR reflectance and increased red reflectance (Tables 1 and 2; Fig. 2-VI). It is hypothesized that incident radiation coming into contact with the hyaline fungal structures on the surface of the leaf is both scattered and absorbed. The random growth of conidiophores on the leaf surface produces an infinite number of angles, which reflect light, thus causing scattering. In addition, the numerous water droplets that are trapped between hyphae and conidiophores covering the leaf will act to absorb light.

In this study, changes in NIR and red reflectance, and a CIR image enhancement technique, the NIR/red ratio (simple vegetation index), have provided a mechanism to differentiate between healthy and stressed plants. Although similar trends have been observed using field-level airborne remote sensing, this is the first study, to our knowledge, which differentiates plant stress resulting from sooty mold and fungal pathogens using such image analysis techniques under a glasshouse setting. Additionally, the comparison of whole plant foliage with individual leaves mounted on a template has proven to be a valuable experimental tool to differentiate component reflectances that contribute to the identification of overall differences between healthy and stressed hosts.

Practical uses and the adoption of such image analysis technology in the large-scale greenhouse setting will depend on several factors. The application must be: 1) effective
(the technology must accurately differentiate between healthy and stressed plants under diverse conditions), 2) inexpensive (acquisition of CIR images must be performed with inexpensive CIR cameras with digital charge coupled devices), and (3) user-friendly (on-site software processing with an easily interpreted interface). Our results suggest a considerable potential for identification of honeydew, sooty mold (and concurrent feeding insect infestations) as well as fungal foliar diseases.

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