# Diagnoses of Abiotic Stress in Cucumber Plant with Non-destructive Physiological Instruments

J. H. Sung, S. R. Suh, G. C. Chung, K. H. Lee

**Abstract**: This paper describes methods to diagnose abiotic stresses such as low root temperature, low light intensity and high salinity in cucumber plants with several physiological instruments. The stresses could be detected by measuring and analyzing the differences in chlorophyll content, temperature difference between leaf and atmosphere and light absorptance at wavelengths of 480, 560, 710, 1420 and 1650nm. It was concluded that the stresses could be first diagnosed from the 3rd to 10th day after treatment and the overall accuracy of diagnosis was estimated between 25 and 75%. Near-infrared spectrometer showed better and earlier detection than the other instruments investigated.

Keywords: Low root temperature, Low light intensity, High salinity, Stress diagnosis, Cucumber plant

## Introduction

To improve agronomic and horticultural plant production, elimination of physiological disorder in plants is one of the decisive task for farmers, especially for winter cultivation of plants in greenhouses. Physiological disorder in plant has been well defined by Lichtenthaler (1996) as "dis-stress" which is a severe and a real stress that causes damage, and thus has a negative effect on the plant and its development. The physiological disorder of plant is developed by various sources - diseases, insects, nutritional and abiotic environmental stresses. Abiotic environmental stresses such as low root temperature, low light intensity and high salinity in soil are not only act as sources of the disorder but also may induce other disorder like nutritional deficiences.

To eliminate the disorder, it will be helpful for farmers to detect the stress in advance. However, the

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Corresponding author: Sang Ryong Suh, Professor, Dept. of Bio Systems & Agricultural Engineering. Institute of Ag. Sci. and Tech., Chonnam National University, Korea: e-mail: srsuh@chonnam.ac.kr detection is not easy, especially for the abiotic stresses. Much effort has been focused in developing sensors to detect various plant stresses. Sophisticated instruments to measure chlorophyll content, leaf temperature, chlorophyll fluorescence, stomatal resistance, photosynthetic efficiency, light reflectance and xylemsap flow have been developed and widely used (Schurer et al., 1991).

The sensors are classified as destructive and nondestructive types. Non-destructive sensors are preferable and recommended for the detection (Pearcy et al., 1989; Buschmann et al., 1994). Among the plant sensors of non-destructive type, Gausman and Quisenberry (1990) and Milton et al. (1991) emphasized the importance of spectrophotometric measurement of reflectance, transmittance and absorptance of a single leaf can often be used to detect plant stresses or damages caused by nutritional deficiency, diseases, growth regulators and soil salinity. They noted that stressed leaves usually exhibit higher reflectance (less absorptance) than non-stressed leaves. Carter (1994) and Elvidge and Chen (1995) suggested various ratios of leaf reflectance (vegetation index) which are effective to detect plant stresses with the passive reflectance. Lichtenthaler and Rinderle (1988) and Buschmann et al. (1994) noted the importance of active laser induced chlorophyll fluorescence measurement in stress detection in plants along with passive reflectance (sunlight reflectance). Sung et al. (1999) and Suh et al. (2000, 2001) tried to detect nutritional deficiencies of N, P, and Ca of cucumber and tomato plants using various physiological instruments.

The goals of this study are to select suitable physiological instruments for non-destructively detecting abiotic stresses caused by low root temperature, low light intensity and high salinity of soil in cucumber plant cultivation and to estimate the possibility of diagnosing the stresses using the instruments.

### Materials and Methods

#### 1. Materials

Cucumber (*Cucumis sativus* L.) seeds were sown in the carbonized chaff. Cucumber seedlings at two leafy stage were transferred into 4-L pots containing Cooper's nutrient solution (1975). Cultivation of cucumber plants for the experiments were described elsewhere (Sung et al., 1999; Suh et al, 2000).

#### 2. Instruments

The same physiological instruments used by Suh et measure al. (2001) were used to physiological parameters of cucumber plants such as chlorophyll content (SPAD-502, Minolta Co. Ltd., Japan); photochemical efficiency (Fv/Fm, Fv: variable fluorescence, Fm: maximal fluorescence) by chlorophyll fluorescence measurement system (CF-1000, Morgan Scientific Inc. USA); the temperature difference between atmosphere and leaf temperatures (DT) (510B, Everest InterScience Inc. USA); and the spectrum data of light absorptance in the range of 400 to 2500 nm in 2 nm intervals (NIRS 6500, Perstorp Analytical Inc.).

# 3. Methods

The experiments were performed by comparing instrumental bio-information collected from cucumber plants treated with abiotic stresses such as low root temperature, low light intensity and high salinity with that of control cucumber plants. A shading curtain (blocking 60% of light) was installed in the upper part of plants for low light intensity stress. The temperature of nutrient solution was maintained at 10°C for low root temperature stress. For the high salinity stress, additional NaCl was supplied in Cooper's nutrient solution.

Each two leaves from plants for 10~15 days were selected at 3 plants per pot. Then, the bio-information of the leaves was collected using the physiological instruments. The data were acquired for 2 hours from 10:00 to 12:00 for 12~20 days.

Experiments for the three abiotic stresses were

repeated 4 times. In each replication four physiological instruments were used to collect the bio-information except chlorophyll fluorescence measurement system with two replications and near-infrared spectrometer with three replications for the stress of low root temperature.

All experiments were performed in a plastic greenhouse and growth chambers at Chonnam National University, Kwangju from April 1997 to June 2000.

# **Results and Discussion**

Significant difference between the stressed and control plants were tested using a two-sample T-test. For the test, we presumed the plant as the stressed plant when the T-test resulted a difference at the significant level of less than 5% during over continuous 3-days within the 12th day after treatment.

Most of the previous studies on the physiological instruments to detect plant stresses have been mainly concerned to find sensitive physical or physiological factors occurred due to the stress imposed. However, the stress could not always be detected because of irregular response of plants possibly due to the variation of adaptation or acclimation to stress. Hence it is desirable to have information on possibility, rather than accuracy, to detect the stresses in order to apply the diagnosis technique to practical uses. In the stressed plant, we tried to find out the first day of any physiological changes in leaves possibly to detect stresses along with the diagnostic rate (ratio of number of experiments that were possible to detect stresses within significant level out of total number of experiments). The diagnostic results of the instruments are as follows.

# 1. Chlorophyll meter

To confirm the possibility to diagnose the effect of low root temperature on chlorophyll content, daily changes of the content of treated and control plants were measured and their differences indicated by the significance level resulted from the T-test were investigated. Chlorophyll content of treated plants showed an obscure trend of lower values than that of control in all 4 replications. The results were expected because of gradually lowered physiological activity of the treated plants. However, the trend was not regarded as an indication to detect the stressed plant until the result was proved by the statistical approach to

distinguish the stressed plant as noted. Among the 4 replications, only one was proved as the stress detectable, and Fig. 1 shows the daily changes of chlorophyll content and significant level of the difference between treated and control plants. As a result, overall diagnostic rate of effect of low root temperature was estimated at 25%. The first day to detect the stress was the 8th day after treatment.

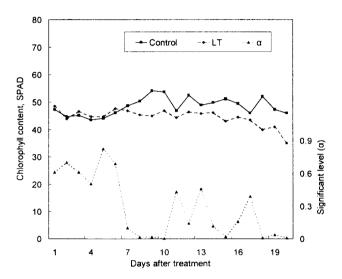


Fig. 1 Comparison of chlorophyll content in leaves of low root temperature treated and control cucumber plants (the stress was clearly diagnosed from 8th day after treatment).

Possibilities to detect the other stresses were checked by the similar manner described above. Chlorophyll content of plants under low light intensity showed a trend of lower values than that of control in all 4 replications as expected. The stress could be detected in 2 replications at 5th and 9th day after treatment, respectively. Fig. 2 shows an example of the experimental data that the stress was detected. Overall diagnostic rate for the low light intensity stress was approximately 50%.

To diagnose high salinity, daily variation of chlorophyll contents of highly salinized and control plants is shown in Fig. 3. This is a case the stress was detectable in which chlorophyll content of treated plants showed higher values than that of control. Statistical analysis showed that the instrument was able to detect significant difference in chlorophyll content on the 6th day and the overall diagnosis rate of chlorophyll meter was about 50%.

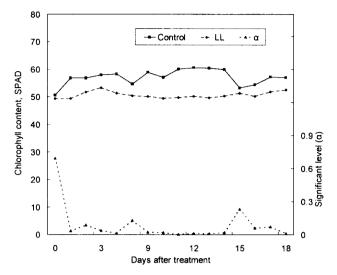


Fig. 2 Comparison of chlorophyll content in leaves of low light treated and control cucumber plants (the stress was clearly diagnosed from 9th day after treatment).

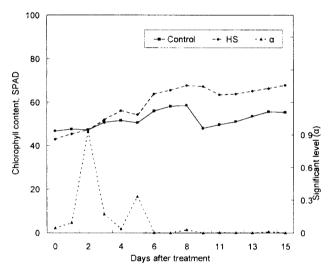


Fig. 3 Comparison of chlorophyll content in leaves of highly salinized and control cucumber plants (the stress was clearly diagnosed from 6th day after treatment).

# 2. Chlorophyll fluorescence measurement system

Lichtenthaler and Rinderle(1988) reported physiological activity of plant leaf could be evaluated by photochemical efficiency (Fv/Fm) measured by the chlorophyll fluorescence measurement system. system was used to detect the abiotic stresses and was proved as an ineffective instrument for diagnosing the effect of low root temperature, low light intensity and salinity because any consistent differentiate the treated from control plants was not obtained as shown in Fig. 4, 5, and 6, respectively.

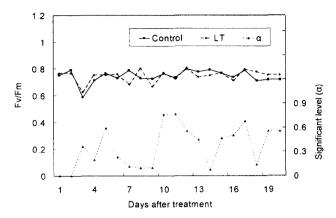


Fig. 4 Comparison of photochemical efficiency (Fv/Fm) of leaves of low root temperature treated and control cucumber plants (the stress could not be diagnosed).

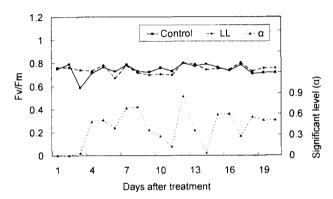


Fig. 5 Comparison of photochemical efficiency (Fv/Fm) of leaves of low light treated and control cucumber plants (the stress could not be diagnosed).

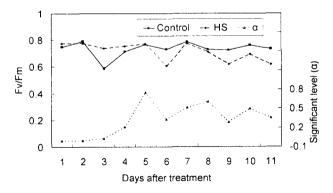


Fig. 6 Comparison of photochemical efficiency (Fv/Fm) of leaves of low light treated and control cucumber plants (the stress could not be diagnosed).

## 3. Infrared thermometer

The leaf temperature did not showed any consistent result to distinguish the control plants from the plants

treated by low root temperature and low light intensity as shown in the experiments of nutritional deficient (Suh et al., 2001).

Possibility to detect high salinity stress was checked. Leaf temperature of plants under high salinity stress showed higher value than that of control only once out of 4 replications from 5th after treatment as shown in Fig 7. Overall diagnostic rate for the high salinity stress was evaluated as 25%.

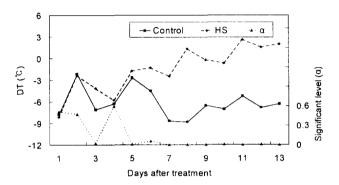


Fig. 7 Comparison of temperature difference between leaf and atmosphere (DT) of highly salinized and control cucumber plants (the stress was clearly diagnosed from 5th day after treatment).

### 4. Near-Infrared spectrometer

Appropriate wavelengths to detect the stresses were investigated by analyzing light absorptance difference between treated and control plants as in the previous studies (Sung et al., 1999; Suh et al., 2000 and 2001).

For low root temperature stress, sensitive wavelength ranges were 450~1,100nm and 1,350~2,000nm but the appropriate wavelengths could not be specified because of the wavelength ranges sensitive to the stress were very wide. Within the ranges, wavelengths of 560 and 1,420nm were selected and checked the diagnostic rate. The stress could be detected on 1st to 7th day, relatively earlier than the other instruments tested, and the overall diagnostic rates for detecting the stress were 75%. Fig. 8 shows an example of daily variations of the light absorptance of treated and control plants and significant level of their difference at a wavelength of 560nm.

The low light intensity stress could be detected at the wavelength ranges of 450~520, 540~690 and 1,330~2,000nm and appropriate wavelengths were selected as 480, 560 and 1,650nm. The stress could be detected at 3rd to 10th day in 480 and 560nm with the overall diagnostic rate of about 25%. One of

unexpected experimental results in the visible sensitive range was a tendency of higher values of light absorptance in treated plants than in control plants as shown in Fig. 9. The tendency could be explained as a result of acclimation to the low light intensity stress of treated plants. At 1,650nm, the stress could be detected on the 6th or 7th day with the diagnostic rate of 50%. At this wavelength, light absorptance from leaves of treated plants was lower than that from leaves of control plants.

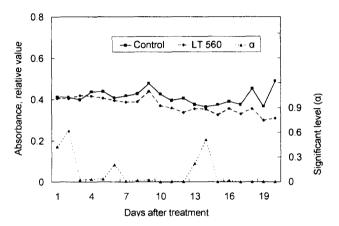


Fig. 8 Comparison of light absorbance at a wavelength of 560 nm from leaves of low root temperature treated and control cucumber plants (the stress was clearly diagnosed from 7th day after treatment).

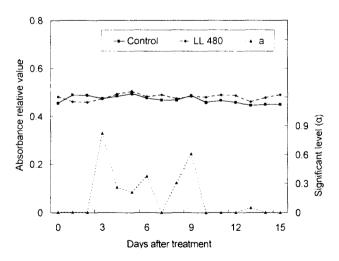


Fig. 9 Comparison of light absorbance at a wavelength of 480 nm from leaves of low light treated and control cucumber plants (the stress was clearly diagnosed from 10th day after treatment).

To detect high salinity stress, sensitive wavelength ranges were 520~650nm and 690~760nm and the

appropriate wavelengths were 560 and 710nm. Higher light absorptance from treated plants than the control was apprehended as shown in Fig. 10. The result was expected because treated plants showed higher chlorophyll content as shown by chlorophyll meter. The salinity stress could be detected at the 6th day and at least in the 10th day by the appropriate wavelengths. The overall diagnostic rates of the spectrometer at wavelengths of 560 and 710nm were estimated at 50 and 75%, respectively.

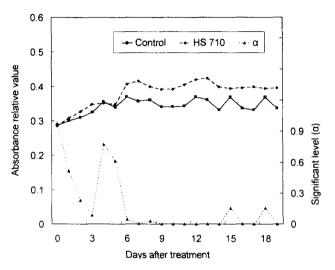


Fig. 10 Comparison of light absorbance at a wavelength of 710 nm from leaves of highly salinized and control cucumber plants (the stress was clearly diagnosed from 6th day after treatment).

The spectrophotometric analysis of cucumber leaves resulted in higher diagnostic rates for the abiotic stresses investigated among the 4 different instruments as described above. The result was similar to the experimental results to detect nutritional deficient stresses in cucumber and tomato plants (Suh et al., 2000 and 2001). In particular, the spectrophotometric analysis showed an superior ability of early detection compared to the other instruments investigated.

The present study on abiotic stresses and the previous studies on nutritional deficient stresses in cucumber plants were mainly concerned to check the possibility to diagnose the various stresses applied separately. It would be worthwhile to consider the possibility to apply the results to complex stresses because farmers would confront two or more stresses at a same time in practical farming. However, it is expected that answer for this problem can be given

only after physiological reaction of plants to the stresses are clarified.

#### **Conclusions**

The results of experiments to diagnose abiotic stresses such as low root temperature, low light intensity and high salinity on cucumber plants using different measuring instruments are as follows:

Low root temperature stress could be diagnosed by chlorophyll meter and near-infrared spectrometer, and the diagnostic rate of the instruments were estimated at 25 and 75%, respectively. Low light intensity stress could be detected by chlorophyll meter and near-infrared spectrometer, and their diagnostic rates were 50 and 25~50%, respectively. High salinity stress was detected by chlorophyll meter, leaf thermometer and near-infrared spectrometer at the diagnostic rates of 50, 25 and 25~75%, respectively.

The first day to detect low root temperature stress after treatment was in a range of 1st to 8th day by chlorophyll meter and near-infrared spectrometer. Low light intensity stress was detected by chlorophyll meter and near-infrared spectrometer within a range of 3rd to 10th day after treatment with the earliest detection ability. High salinity stress was detected by chlorophyll meter, leaf thermometer and near-infrared spectrometer from 5th to 10th day after treatment. Generally near-infrared spectrometer resulted in a superior ability of early detection compared to the other instruments investigated.

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