Response of Vegetation Indices to Changes in Three Measures of Leaf Water Stress

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ABSTRACT: The responses of vegetation indices to changes in water stress were evaluated in two separate laboratory experiments. In one experiment the normalized difference vegetation index (NDVI), the near-IR to red ratio (near-IR/red), the Infrared Index (II), and the Moisture Stress Index (MSI) were more highly correlated to leaf water potential in lodgepole pine branches than were the Leaf Water Content Index (LWCI), the mid-IR to red ratio (Mid-IR/red), or any of the single Thematic Mapper (TM) bands. In the other experiment, these six indices and the TM Tasseled Cap brightness, greenness, and wetness indices responded to changes in leaf relative water content (RWC) differently than they responded to changes in leaf water content (WC) of three plant species, and the responses were dependent on how experimental replicates were pooled. With no pooling, the LWCI was the most highly correlated index to both RWC and WC among replicates, followed by the II, MSI, and wetness. Only the LWCI was highly correlated to RWC and WC when replicates were pooled within species. With among species pooling the LWCI was the only index highly correlated with RWC, while the II, MSI, Mid-IR, and wetness were most highly correlated with WC.

INTRODUCTION

REMOTE SENSING OF WATER STRESS is of great interest to plant scientists and natural resource managers (Olson, 1977; Wiegand et al., 1983). Digital spectral reflectance data have been used for assessments of stress in agricultural crops (Sinclair et al., 1971; Ripple, 1986; Collier, 1989), grassland (Tucker, 1977), rangeland (Everitt and Nixon, 1986), and forestland (Rhode and Olson, 1970; Cohen, 1989). Linear transformations of reflectance data, or vegetation indices, are commonly used for such assessments. These indices fall into two broad categories, termed ratio-based and n-spaced (Elvidge and Lyon, 1985). As discussed in this paper, vegetation indices are relative to the six Landsat Thematic Mapper (TM) bands that detect reflected energy. The wavelength intervals for the TM bands in µm units are as follows: band 1 (TM1), 0.45 to 0.52; TM2, 0.52 to 0.60; TM3, 0.63 to 0.69; TM4, 0.76 to 0.90; TM5, 1.55 to 1.75; and TM7, 2.08 to 2.35.

Although few vegetation indices have been developed specifically for plant water stress evaluation, several ratio-based indices have received this application. Birth and McVey (1968) developed the near-infrared to red reflectance ratio (near-IR/red),

\[ \frac{\text{TM4}}{\text{TM3}} \]  

and Rouse et al. (1974) developed what is now called the normalized difference vegetation index (NDVI),

\[ \frac{\text{TM4} - \text{TM3}}{\text{TM4} + \text{TM3}} \]  

These two indices respond to changes in amount of green biomass (Deering et al., 1975; Tucker, 1979), chlorophyll content, and leaf water stress (Tucker et al., 1980; Arai et al., 1983). Hardisky et al. (1983) found that the infrared index (II),

\[ \frac{\text{TM4} - \text{TM5}}{\text{TM4} + \text{TM5}} \]  

more closely tracked changes in plant biomass and water stress than did the NDVI. Rock et al. (1985) found that the moisture stress index (MSI),

\[ \frac{\text{TM5}}{\text{TM4}} \]  

closely followed changes in plant water stress. Hunt et al. (1987) demonstrated a responsiveness to changes in water stress of the leaf relative water content index (LWCI),

\[ -\log\left[1 - \frac{\text{TM4} - \text{TM5}}{\text{TM4} - \text{TM6}}\right] - \log\left[1 - \frac{\text{TM4} - \text{TM5}}{\text{TM4} - \text{TM6}}\right] \]

where \( f \) represents reflectance in the specified bands when leaves are at their maximum relative water content (RWC). RWC is defined as

\[ \frac{\text{field weight-ovendry weight}}{\text{turgid weight-ovendry weight}} \times 100 \]

Musick and Pelletier (1988) demonstrated a strong correlation between the ratio

\[ \frac{\text{TM5}}{\text{TM7}} \]

and soil moisture. In this paper Equation 6 is called the Mid-IR index.

The Tasseled Cap transformation (Kauth and Thomas, 1976) is an n-spaced vegetation index (Jackson, 1983). The first two axes of the Tasseled Cap are separate vegetation indices called brightness and greenness, respectively. Brightness and greenness typically account for 95 percent of the spectral variation in Landsat Multispectral Scanner (MSS) scenes of agricultural areas (Crist et al., 1986). Sensitivity of greenness to plant water stress has been evaluated. McDaniel and Haas (1982) found that greenness is highly correlated with plant moisture content. Jackson et al. (1983) found that greenness was not a good stress indicator.

Crist and Cicone (1984) adapted the Tasseled Cap to TM data. The first two indices of the TM Tasseled Cap are similar to those for MSS data. The TM Tasseled Cap, however, has a third useful vegetation index which contrasts reflectance in the visible and near-infrared bands (TM bands 1 to 4) with reflectance in the water absorption bands (TM bands 5 and 7). Crist et al. (1986) refer to this index as wetness. Crist and Cicone (1984) and Musick and Pelletier (1988) demonstrated that wetness responds to changes in soil moisture, but no reference to its sensitivity to plant water stress was found.

Although some of the vegetation indices discussed above have been compared for their sensitivity to changes in plant water stress (Tucker, 1979; Jackson et al., 1983; Hardisky et al., 1983), a comparison of all of them has not been reported. Such a comparison should be useful for examining their utility for drought stress evaluation.

The objective of this study was to compare the response of the ratio-based and n-spaced vegetation indices discussed to...
changes in leaf water stress under laboratory conditions. The data used to accomplish these objectives came from two different experiments. In Experiment One, the measure of water stress evaluated was leaf water potential ($\psi$). Leaf water potential is defined as the free energy status of water in a leaf and is a commonly used, direct measure of leaf water stress (Ritchie and Hinkley, 1975). In Experiment Two, leaf RWC and leaf water content (WC)—grams of water per gram of oven-dry leaves—were evaluated.

**MATERIALS AND METHODS**

**EXPERIMENT ONE**

For this experiment, foliated lodgepole pine (*Pinus contorta* Doug. ex Loud.) branches from 1 to 1.25 m long were collected from 10 to 15 year old trees in the Fraser Experimental Forest, in the Rocky Mountains near Fraser, Colorado. Upon collection, they were immediately placed in airtight plastic bags and transported to a laboratory. Ten cm was excised from their cut ends and they were placed in a vessel of water 15 cm deep. The branches remained in the vessels for a 24-hour period. Throughout this period, $\psi$ of needle fascicles was measured with a pressure chamber using a method described by Scholander et al. (1965).

After 24 hours, samples of $\psi$ were near zero and it was assumed that the branch foliage had come to full turgor. At this time, the branches were excised at the water line to remove wetted foliage and stems, and randomly stacked on the floor within the field of view of the radiometer over a piece of 60- by 80-cm cardboard painted flat black. The branches were stacked four layers deep to an overall height of approximately 12 cm and the 4- to 6-cm long needles overlapped freely. The branches remained as stacked for a three-day period under fluorescent room lighting at a room relative humidity of between 30 and 40 percent. During this three day period 40 measurement sets of spectral reflectance and $\psi$ were recorded as they dried. A measurement set consisted of reflectance through each of the TM bands 3, 4, 5, and 7.

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The transformed data were subjected to simple linear regressions, using TM band reflectance and vegetation indices as dependent variables and $\psi$ as the independent variable. An assumption in regression analysis is that the independent variable is measured without error. The standard deviation of nine $\psi$ measurement sets was less than 0.1 MPa, eleven were between 0.1 and 0.2 MPa, five were between 0.2 and 0.3 MPa, six were between 0.3 and 0.4 MPa, and only five were over 0.4 MPa. Thus, errors existed in measuring $\psi$, but they were not large relative to the range of $\psi$ values examined (2.25 MPa).

To determine the relative value of the indices examined for estimating $\psi$, a technique called inverse regression was used to compute 95 percent confidence interval half-widths (CIs) on predictions of $\psi$ from each of the indices. This technique is described by Eisenhart (1939) and Nathrela (1963) and was suggested for this data set by Dr. Harirhan Iyer. The procedure often is used to calibrate an instrument and permits one to compare the usefulness of various instruments for making a given measurement. CIs were computed for five levels of $\psi$: -0.5, -1.0, -1.5, -2.0, and -2.5 MPa. To permit comparisons between individual bands and the various vegetation indices, CIs also were calculated for predictions of $\psi$ from reflectance in individual bands.

**EXPERIMENT TWO**

For this experiment, a Beckman UV-5240 spectrophotometer equipped with an integrating sphere and halogen reference standard was used to measure hemispheric diffuse reflectance from leaves over the wavelength range from 0.4 to 2.5 $\mu$m. The instrument was located at the Jet Propulsion Laboratory in Pasadena, California, and measured and recorded reflectance every 0.001 $\mu$m in the region from 0.4 to 0.8 $\mu$m and every 0.004 $\mu$m in the 0.8 to 2.5 $\mu$m region. Bandwidths ranged from 0.001 to 0.028 $\mu$m. Leaves were stacked between seven and ten layers deep for each reflectance spectrum obtained.

Collected for this experiment were foliated branches one-half metre long of *Pinus contorta* D. Don (coulter pine), *Ceanothus crassifolius* Torr. (ceanothus), and *Salvia melifera* Greene. (black

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*Department of Statistics, Colorado State University, Fort Collins, CO. Personal communication, October, 1987.
sage). Branches were collected at the San Dimas Experimental Forest, San Gabriel Mountains, Los Angeles County, California. On a given day, branches from only one species were collected and branches were collected on three different days for each species, yielding nine branch sets. Immediately after collection of a given branch set, the following procedures were used:

- Within minutes the branches were immersed in water, and several centimetres were excised from their cut ends before placing them in test tube water baths for a 24-hour period. In all cases for which subsequent measurements were made, \( \phi_s \) of the branches leveled off at near zero by the end of this period. At that time it was assumed that the foliage had come to full turgor.
- The current year’s foliage was then plucked from the branches and a sample from the plucked leaves was selected to be used for spectral data collection. The sample’s turgor weight was measured, and then the sample was placed in the spectrophotometer to obtain a reflectance spectrum. The leaves in the instrument’s sample compartment were not disturbed for subsequent spectral reflectance measurements, and they dried more slowly than if exposed to air circulation.
- While the spectrophotometer was obtaining a spectrum, the remainder of the leaves were weighed for turgor weight and then placed in a cardboard box. The box was covered with a piece of paper to prevent rapid leaf drying.
- Five more times during the following 24-hour period, a reflectance spectrum of the leaves in the spectrophotometer sample compartment was collected and the leaves in the cardboard box were weighed.
- After the sixth and final measurement, the leaves were removed from the spectrophotometer sample compartment and weighed. Then both them and the leaves from the box were dried for two days at 65°C before weighing each one final time for its oven dry weight.
- RWC and WC were calculated for the leaves from the box for each of the six times a reflectance spectrum was obtained. RWCs and WCs of the sample from the spectrophotometer at the beginning and end of the 24-hour measurement period were calculated to determine if these measurements were similar to those obtained on the foliage in the box. In all cases, the two measurements of RWC and WC for the spectrophotometer leaves were within a few percent of the corresponding measurements for the leaves in the cardboard box. Thus, the six RWC and WC measurements obtained from the leaves in the cardboard box can be used to describe the RWC and WC of the foliage in the spectrophotometer each time a reflectance spectrum was obtained.

Using the data from the nine branch sets, brightness, greenness, and wetness values were calculated for the spectral measurements associated with each of the six levels of RWC and WC observed. This was done using measured reflectance at the midpoint of the six reflectance TM bands and the TM Tasseled Cap equivalent transformation coefficients for reflectance factor data given by Crist (1985) and shown in Table 2. Values for the six vegetation indices of Equations 1 to 6 also were calculated using midpoint reflectance values. Because band-midpoint reflectance values were used, results may be somewhat different than if band-integrated values had been used. Differences should be slight, however.

For each of the nine replications, Pearson product moment coefficients of correlation (Mendenhall, 1975) were calculated for the relationships between RWC, WC, the TM bands, and the nine vegetation indices evaluated. Correlation coefficients also were calculated for these relationships after pooling replications within species, and after pooling replications among species.

### RESULTS AND DISCUSSION

#### Experiment One

Leaf water potentials observed during this experiment (Table 1) were representative of those exhibited by lodgepole pine during an annual growth cycle. The greatest changes in reflectance occurred in bands 5 and 7, and were 3.4 and 2.2 percent, respectively. Reflectance in TM3, TM4, and TM7 was inversely related to \( \phi_s \) while TM4 reflectance was directly related to \( \phi_s \) (Table 3). TM4 had the highest standard error and smallest regression line slope and coefficient of determination \( (r^2) \) of any of the bands. Thus, it was the least responsive changes in \( \phi_s \). TM5 had the highest \( r^2 \) and greatest slope, and was the most responsive of the bands to changes in \( \phi_s \). TM5 had a slightly lower \( r^2 \) and a somewhat lesser slope than TM5, but also had a lower standard error. The lower standard error implies that TM5 may provide more reliable estimates of \( \phi_s \), but the lesser sensitivity (slope) means measurements must be more precise for practical applications. All vegetation indices, except the LWCI and Mid-IR, were better correlated to \( \phi_s \) (higher \( r^2 \)) than were TM5 and TM7 (Table 3), each accounting for approximately the same amount of variation associated with changes in \( \phi_s \).

That only 59 percent of the variation in the LWCI was explained by changes in \( \phi_s \) has a probable explanation. The LWCI was developed using theory based on absorption coefficients for water at varying thicknesses (Hunt et al., 1987). As such, this vegetation index responds to changes in water volume in leaf tissue. Although a change in \( \phi_s \) over the range observed should be associated with a change in water volume in leaf tissue, the relationship is expected to be nonlinear and the change in actual volume was probably quite small.

Water absorbs more strongly in TM7 than in TM5 (Cuccio and Petty, 1951). Thus, there is a physical basis for expecting a response of the Mid-IR to changes in \( \phi_s \) and experimental evidence exists that this ratio responds to changes in soil moisture (Musick and Pelletier, 1988). The lower \( r^2 \) resulting from this experiment does not support this however.

Confidence interval half-widths calculated to permit further comparisons among the vegetation indices and individual wavebands appear in Table 4. In terms of CIs, the relative ranking of the bands and indices is the same for all levels of \( \phi_s \). The MSI has the lowest CI, followed by the IL, near-IR/red, NDVI, TM5, TM7, TM3, LWCI, Mid-IR, and TM4. An appropriate interpretation is that all indices examined, except the LWCI and Mid-IR, appear better than individual bands for evaluating \( \phi_s \) in foliated lodgepole pine branches. However, even for the MSI, the lowest CI is 0.61 MPa. Thus, the range between the lower and upper confidence limits (1.22 MPa) spans a range of \( \phi_s \) values that is large relative to the full range examined (2.25 MPa).

#### Experiment Two

On an individual replication basis, the relationship between RWC and WC for this experiment was quite strong. For each decrease in RWC, there was a corresponding decrease in WC (Table 5), and correlation coefficients between the two stress measures were greater than 0.99 for all nine replications. Thus, when examining the correlation between water stress and band index responses on this basis, both of these measures of stress can be presented together.

With ceanothus, TM5 and TM7 were the most responsive bands to changes in RWC and WC and the relationships were consistent in sign (i.e., all inversely related) among replications (Figure 1a). For both bands, the relationships were strong, with TM7...
being slightly more responsive for all three replications. For the other bands, the relationships were mixed among replications. As an example, TM4 had a strong positive correlation with RWC and WC for Replication A, a strong negative correlation for Replication B, and a weak negative correlation for Replication C. Among the indices evaluated, the II and wetness were strongly correlated with RWC and WC and the relationships were consistently positive. The MSI exhibited relationships with RWC and WC that were essentially the inverse of those of the II. The LWCI also had generally strong and consistent relationships with RWC and WC. For the other indices, relationships were mixed in sign and/or weak.

For coulter pine, with a few minor exceptions, essentially the same results were observed as for ceanothus (Figure 1b). Bands 5 and 7 exhibited the strongest and most consistent relationships with RWC and WC, although the relationships for TM5 were slightly stronger here. Of the indices evaluated, the II and MSI were the most closely correlated to RWC and WC, and again, these indices exhibited an inverse relationship with each other. The relationships for the LWCI with RWC and WC were consistently negative, and for this species they were stronger than for ceanothus. Wetness was strongly and positively correlated to RWC and WC for two replications only and was weakly negative for the third. For coulter pine, brightness was more closely and consistently correlated with RWC and WC among replications than was wetness.

The relationships of RWC and WC with band and index responses for black sage were unlike those of the other two species (Figure 1c). For sage, no single band was consistently well correlated to water stress. Bands 3, 4, 5, and 7 were highly correlated to RWC and WC for Replications B and C, but the coefficient had a different sign and/or the relationships were weakly correlated for Replication A. Band 3 exhibited positive correlations with RWC and WC for Replication A. This was unexpected—being contrary to what theory would suggest (Tucker, 1980)—and apparently affected the relationships of the II and MSI with RWC and WC, making them weak. For this species, the Mid-IR had generally strong and consistent relationships with water stress, while wetness had very strong relationships.
From the above discussion it is apparent that, even for a single species, a given TM band varied in its response to water loss depending on the leaf set that was evaluated. This is probably because leaf structure and the proportions of the various cell constituents are variable among leaves. Thomas et al. (1971) noted that this situation may inhibit the ability to predict leaf water stress from reflectance measurements. Similarly, this topic is the focus of another paper using the band reflectance data from this experiment (Cohen, 1990). Analysis of the data here reveals that variation among leaf sets also was enough to inhibit the ability to predict water stress from vegetation indices.

To illustrate this, recall that wetness was strongly correlated with RWC and WC among the three replications for ceanothus and that the relationships were consistently positive (Figure 1a). The slopes of simple linear regression lines drawn through the data for each replication are different however (Figure 2a). Furthermore, while wetness values changed by less than 2.0 from high to observed low RWC for each replication, intercepts among replications ranged from -4.38 to -15.66. For still other indices, such as the near-IR/red for black sage, even though correlation coefficients were relatively high among replications (Figure 1c), differences in slope direction among replications for two replications and a weakly strong one for the other. The LWCI was the index most strongly and consistently correlated with RWC and WC for black sage.

FIG. 1. Correlation coefficients for the relationships of relative water content and water content with Thematic Mapper (TM) bands and vegetation indices for nine experimental replications using leaves of ceanothus (A), coulter pine (B), and black sage (C). See Equations 1 to 6 in text for index formulations.

FIG. 2. (A) Wetness as a function of ceanothus leaf relative water content for three experimental replications. (B) The near-IR to red ratio as a function of black sage leaf relative water content for three experimental replications. See Equations 1 to 6 in text for index formulations.
were observed (Figure 2b). Similar observations were made with each replication for all band and index responses.

Given the above observations, it seems prudent to ask "what are the relationships of band and index responses to RWC and WC when data from replications are pooled by species and are pooled among species?" This question is pertinent to situations where remote sensing data are collected over vegetation communities by imaging sensors, where ground resolution cells are large enough to view several plant canopies.

The correlation coefficients for the relationships between RWC and WC when data were pooled among replications for ceanothus, coulter pine, and black sage were 0.88, 0.67, and 0.69, respectively. This means the relationships for RWC and WC with band and index responses must now be considered separately. With RWC, only the LWCI exhibited a relatively strong relationship for each species and had a consistent sign among species (Figure 3a). The same is true for the relationship of WC with band and index responses (Figure 3b). Note that for black sage, positive relationships for WC and bands 5 and 7 were observed. Again, this is unexpected, being contrary to what theory would suggest.

When all nine replications were pooled, RWC and WC had a correlation coefficient of only 0.06. Under these conditions, the LWCI was again the only band or index exhibiting relatively strong correlation with RWC (Figure 4). However, the II, MSI, Mid-IR, and wetness were all highly correlated with WC. Because the relationships of TM5 and TM7 with WC were strongly positive, correlation coefficients for the II and MSI had opposite signs than those observed with no pooling and with within species pooling. The correlation coefficient for wetness also had an opposite sign from that expected.

**SUMMARY AND CONCLUSIONS**

In Experiment One, six ratio-based indices were examined for their relationships with leaf water potential (ψᵣ). The normalized difference vegetation index (NDVI), the near-IR to red ratio (near-IR/red), the Infrared Index (II), and the Moisture Stress Index (MSI) were all more strongly related to ψᵣ than were the Leaf Water Content Index (LWCI) and the mid-IR index (Mid-IR) or the individual Thematic Mapper (TM) bands used in the index formulations. Because the coefficients of determination and confidence intervals for predicting ψᵣ from all of the indices except the LWCI and Mid-IR were similar, any of those four indices appear equally well-suited for ψᵣ evaluation in lodgepole pine foliage. However, because even the MSI, II, NDVI, and near-IR/red had confidence limits for predicting ψᵣ from index values that enclosed large ranges relative to the range of ψᵣ values examined, their utility in practical application may be limited.

Results from Experiment Two revealed considerable inconsistency in the responses of TM bands and vegetation indices to changes in leaf relative water content (RWC) and water content (WC). That is, differences in the sign and magnitude of correlation coefficients for all TM bands and several indices were noted among the nine experimental replications—three replications for each of three species. Even for a given species, when signs were consistent and correlation coefficients high among replications, variations in band and index responses among replications exceeded changes in band and index responses for a given replication as RWC and WC changed. In general, however, correlation coefficients were high and consistent in sign for TM5, TM7, the MSI, and II for all replications except Replication A of black sage; and for the LWCI in all replications except Replication B of ceanothus, where the correlation coefficient was consistent in sign but only mildly strong. TM Tasseled Cap wetness was
highly correlated with RWC and WC and consistent in sign among replications except for Replication A of black sage and Replication A of cottle pine.

When data for replications within species were pooled, only the LWCI was relatively well correlated with RWC and WC for each species. Pooling of the data for all nine replications revealed that only the LWCI was well correlated with RWC and that TM5, TM7, the II, MSI, Mid-IR, and wetness were strongly correlated with RWC. Bands 5 and 7 correlation coefficients were positive, however, causing the II, MSI, and wetness coefficients to have signs opposite to that which was generally observed (and to that which leaf optical theory would suggest) with no pooling and with pooling within species. That the response of TM bands and vegetation indices varied depending on how the data were pooled is undoubtedly due to their inconsistent responses to changes in RWC and WC among replications.

The poor response of the LWCI to changes in φᵢ in Experiment One, when contrasted against its superior response to changes in RWC and WC in Experiment Two, can be explained. The general nature of the relationship between φᵢ and RWC is nonlinear and the LWCI is a linear model developed using theory based on absorption coefficients for water. Thus, the LWCI responds to changes in water volume and not to changes in the free energy status of water in leaf tissue.

This study revealed that TM band and vegetation index responses to changes in leaf water stress are a function of the measure of stress examined and the sample of leaves used. As a result, the use of vegetation indices to predict water stress in leaves appears limited. Whether vegetation indices respond more definitively to changes in water stress in plant canopies and whole vegetation ecosystems cannot be determined from these results. Probably, however, for indices to be useful for canopy or ecosystem stress assessments, changes in live to the dead biomass ratio, canopy architecture, and other stress-related effects are necessary.

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