

## **MONITORING SALINITY STRESSED SPINACH VIA SPECTRAL REFLECTANCE MEASUREMENTS**

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### **ABSTRACT**

Extensive research has been carried out using hand-held radiometers, aerial platforms and satellites to observe water and nitrogen status of vegetation. Salinity detection using remote sensing attracted scientist's attention in the past three decades, however, their work often focused on detection of salt affected soils. Real time detection of salt affected crops is important as well. It is considered an uncharted research area especially for vegetables given that most experiments focus on major crops (cotton, wheat, etc.). The main objective for this study was to investigate the potential usefulness of growing-season spectral measurements to detect and monitor salinity stress effects on spinach. Spinach plants were planted in twelve 1x1 meter plots that had 4 salinity treatments (equivalent to yield reductions of 0, 10, 25 and 50%), three replicates each. A hand-held radiometer with filters in the visible and infrared parts of the electromagnetic spectrum was used to monitor salt affected spinach plants. Spectral indices; salinity stress index (SSI) and normalized difference vegetation index (NDVI), as well as spectral reflectance at different wavelengths ranging between 510 nm and 1480 nm were used to monitor salinity stress effects on spinach. SSI indicated salinity very well only in the middle of the growing season. Although water and nitrogen were adequately applied, the independence of SSI from the effects of other growth retardation sources can not be confirmed. Reflectance at 750 nm was superior to all the tested reflectance factors and indices in monitoring salinity. Reflectance at 1000 nm comes in the second place among all the tested reflectance factors in differentiating salinity treatments. There are advantages of using reflectance at 750 nm and 1000 nm for monitoring salinity stressed spinach. Also, they may be integrated with 660 nm, 760 nm and 1480 nm bands to develop new indices.

### **INTRODUCTION**

Practically, saline soils cannot be reclaimed by chemical amendments, conditioners or fertilizers. A field can only be reclaimed by removing salts from the plant root zone. Excessive soil salinity reduces the yield of many crops. This ranges from a slight crop loss to complete crop failure, depending on the sensitivity of crop and the severity of the salinity problem. Although several treatments and management practices can reduce salt levels in the soil (Cardon et al., 2006), there are some situations where it is either impossible or too costly to attain desirably low soil salinity levels. In some cases, the only viable management option is to plant salt-tolerant crops.

A possible alternative to measuring soil salinity is to detect plant responses to saline environments as an indicator of soil salinity. Monitoring salinity stressed plants using remote sensing can be less expensive and much easier if compared to field measurements and laboratory tests. Plant

responses may provide a more comprehensive assessment of salinity because plant roots generally penetrate deep in the soil profile. Remote sensing of plant canopy is particularly useful in this respect. Tremendous research has been achieved on the use of remote sensing for detection of salt affected soil (Tricatsoula, 1988, Abdel-Hamid and Shrestha, 1992, Karavanova and Orlov, 1993, and Myers et al., 1996). Gausman and Quisenberry (1990) and Milton et al. (1991) emphasized the importance of spectrophotometric measurement of reflectance; transmittance and absorbance of a single leaf, which 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 absorbance) than non-stressed leaves. Yet, using remote sensing of salinity stressed vegetation in canopy level is relatively unexplored. Although leaf reflectance has been studied in response to salinity stress, the spectral regions (or wavelengths) at which leaf reflectance is most responsive to stress remain largely undefined. Hand-held radiometers can be used for small fields; alternatively, reasonably priced platforms such as the unmanned aerial vehicles (UAV) can be beneficial for monitoring larger fields (Simpson et al., 2003). The objectives of this study were: (1) to test the ability of hand held radiometer with filters at thirteen different wavelengths ranging between 510nm, and 1480nm of the electromagnetic spectrum to detect and monitor salinity stressed spinach; one of the vegetables that are considered highly tolerant to salinity is spinach (FAO, 1985). (2) to choose the most suitable wavelengths that could be used for salt stress detection, and (3), to test the potential of remotely sensed spectral indices; salinity stress index (SSI) and normalized difference vegetation index (NDVI), in differentiating salinity treatments.

## **MATERIALS AND METHODS**

### **2.1. Experimental setup**

The experiment was achieved in Tucson, Arizona, USA (Lat. 32° 16' N, Long. 110° 56' W and Elev. 713m). Spinach (*Spinacia oleracea* L., var. Rushmore) was planted on 28 Oct. (day of year [DOY] 301), 2005 in an alluvial, Silty Clay Loam soil. Table (1) shows some chemical and physical properties of this soil. Planting of spinach in Arizona starts the beginning of October and continues until the middle of February. Rushmore is a popular variety because of its resistance to blue mold, which eliminated the need for pesticides. Nitrogen fertilizer was required at a rate of 127 lb N/acre (60 kg/Fed.) (Thompson and Doerge 1995). After considering soil N, half of the required amount was broadcasted at planting in the form of ammonium sulfate and the other half was injected with water in the form of UAN-32, concentrated mixtures of urea and ammonium nitrate (32%), in two equal doses at DOY 335 and DOY 351.

**Table1: Some chemical and physical characteristics of soil before applying treatments.**

Chemical characteristics							Physical characteristics		
	pH*	EC <sub>e</sub> ****	ESP	NO <sub>3</sub> -N**	PO <sub>4</sub> -P**	K***	Field Capacity	Wilting point	Texture
Unit		dS m <sup>-1</sup>	%	ppm	ppm	ppm	%	%	
	7.4	1.4	7.4	23	14	225	19.5	11.4	Silty Clay Loam

\* pH is obtained from 1:1 water extract.

\*\* NO<sub>3</sub>-N and PO<sub>4</sub>-P are from Olsen bicarbonates extracts.

\*\*\* K was attained using neutral molar ammonium acetate.

\*\*\*\* EC<sub>e</sub> soil paste extract

## 2.2. Experimental design and treatments

Twelve 1 x 1 m plots were formed, after soil was manually tilled, which represent a completely randomized block design with four salinity treatments (0, 10, 25 and 50% yield reduction), each replicated 3 times. The 0, 10, 25 and 50% spinach yield reduction were achieved by maintaining 2, 3.3, 5.3 and 8.6 dS m<sup>-1</sup> (EC<sub>e</sub>), respectively (Cardon et al., 2006). The electrical conductivity of irrigation water (EC<sub>iw</sub>) in relation to the soil water extract (EC<sub>e</sub>) is difficult to predict because of the influences of texture, drainage, duration of saline irrigation and leaching fraction. However, assuming a leaching fraction of 15%, a reasonable estimate can be calculated through equation 1 (Cuenca, 1989).

$$EC_{iw} = (0.65) * EC_e \rightarrow (1)$$

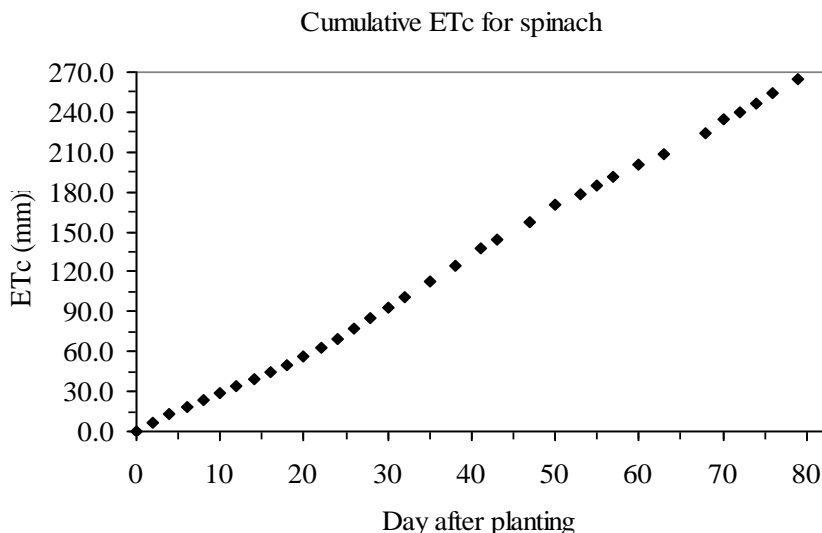
Based on this relationship, the 0, 10, 25 and 50% spinach yield reduction could be achieved by maintaining irrigation water at 1.3, 2.2, 3.5 and 5.6 dS m<sup>-1</sup> (EC<sub>iw</sub>), respectively.

Equivalent irrigation salt content (EC<sub>w</sub>) was calculated for each treatment. Sodium chloride was mixed with water and kept in a 100 liter barrel, which were connected to pumps. The pumps would be adjusted for each treatment to inject the salt into the irrigation system at the time of irrigation. Soil samples were collected three times during the season (day of year [DOY] 354, 2005; 6, 2006; and 23, 2006) for soil electrical conductivity measurements (EC<sub>eM</sub>) to be compared to target electrical conductivity (EC<sub>eT</sub>). A simple excel work sheet was developed to give the speed of the pump that would result in the specified salt concentration using inputs such as salt concentration in the barrel (EC), desired concentration, and watering time.

## 2.3. Irrigation system and salinity injector

Four rows of spinach were planted in each plot, 10 cm apart in each row, at about 1 cm depth. Irrigation water amount was determined based on reference evapotranspiration data provided by the Arizona Meteorological network (AZMET). Single crop coefficient procedures were used. Though, due to the deviation from the standard climatic conditions, the tabulated crop coefficient values were adjusted to account for interval between irrigations and climatic changes using meteorological data provided by a nearby AZMET weather station, 150 m from the study area, and equations provided in Allen et al. (1998).

Bubbler irrigation systems were installed in the field (one bubbler head per plot). Bubblers were calibrated using liter graduated bucket. The average flow rate of the bubblers was 7.56 liter per minute. A 1-inch PVC main pipe was used for the distribution of water to bubblers with pressure gauge at the beginning of the main line. For steady pressure, irrigations took place at early morning two to three times a week from planting until DOY 16 (2006). The maximum allowable depletion did not exceed 0.35 of the total available soil water in the root zone. The calculated cumulative crop evapotranspiration ( $ET_c$ ) for days after planting is presented in figure 1.



**Figure 1: Cumulative crop evapotranspiration ( $ET_c$ ) in mm vs. days after planting for spinach.**

A valve was used to control the water flow for each treatment. All the pipes were buried under the soil and the bubblers were raised above the soil by about 7-cm. A variable rate positive displacement pump was calibrated and used on an external power source (120 volt electricity) for injection of salts. The injector was calibrated using a graduated cylinder, in milliliters, and the injector inlet (suction) was put in the cylinder that is filled to a known volume with the salt that would actually be injected. The injector was equipped with a switch and a graduation from 1 to 10 with 1 being the smallest rate and 10 the largest. The injector was timed for a 1 minute at each speed. Then the 1 minute rates were converted to per hour rates. Most crop plants are more susceptible to salt injury during germination or in the early seedling stages. Therefore, during early-season good water ( $EC_w$  of about  $0.5-1.0 \text{ dS m}^{-1}$ ) was applied to provide good conditions for the crop to grow through its most injury-prone stages. Therefore salinity treatments did not start until 4 Dec. 2005 (DOY 338).

#### **2.4. Reflectance measurements using the multispectral radiometer**

A CROPSCAN, Inc. multispectral radiometer (MSR) system was used with narrow band interference filters to selectively sense specific bands in the visible, near infrared (NIR), and short wavelength infrared (SWIR) regions of the electromagnetic spectrum. It has 16 upward looking and 16 downward looking photodiode sensor (model MSR16R) filtered at the following wavelengths: 510, 550, 560, 610, 660, 710, 750, 760, 810, 900, 1000, 1050, 1240, 1480, 1640 and 1680 nm. Only the first 14 bands were under investigation in this study. The design of the radiometer data acquisition system allowed for near simultaneous inputs of voltages representing incident and reflected irradiation, which helped improving measurement of reflectance from the spinach when sunlight conditions were not ideal.

The radiometer was hand held by a telescopic stick that can be adjusted at different heights. The diameter of the field of view (FOV) was one-half the height of the radiometer above the spinach plants. The radiometer was held a height of 2-m above the spinach plants such that the FOV was 1-m. The CROPSCAN included data acquisition program that digitize the voltages and record the percent reflectance at each of the selected wavelengths, as well as correcting for the sun angle and temperature effects. The radiometer sensor millivolt readings were logged in the data logger controller (DLC). The CROPSCAN software for PC included programs to interface to the DLC to retrieve the data and process the retrieved data to calculate the percent reflectance. The output reflectance data files were ASCII text files, comma delimited for easy import into spreadsheet programs.

Scans were taken at two to three times a week and sometimes every week at 12:30 pm starting at DOY 338 (2005) to DOY 23 (2006). No data were available from DOY 2 to DOY 16 due to clouds and inappropriate conditions. Reflectance along with the normalized difference vegetation index (NDVI - Rouse et al., 1973) and salinity stress index (SSI - Stong, 2003) were plotted versus day of year (DOY) in figures 2 to 6.

#### **2.5. Vegetation and Salinity Indices**

The normalized difference vegetation index, NDVI (Rouse et al., 1973) is an index that is highly correlated to vegetation density. It incorporates the red and near infrared reflectance mostly to measure density of green vegetation. NDVI is the difference between the near infrared (NIR) band and the red band divided by the sum of the NIR and red bands, and its values typically range from 0 (bare soils) to 1.0 (full canopy) over agricultural covers (Equation 2). If both water and salinity stresses take place, NDVI is more likely to be affected by salinity than water stress (El-Shikha, 2005). The second index is the salinity stress index (SSI) that was created to assess salinity stress in crops (Stong, 2003). The SSI respond to salinity treatments was inconsistent until middle of the season for cantaloupe and squash (El-Shikha, 2005). It is calculated by dividing the summation of the reflectance at 1640 and 1480 nm, both shortwave infrared (SWIR), by the reflectance at 1000 nm (NIR), Equation 3. The limits of the salinity stress index changes

from one crop to another and they are not defined for spinach. However, higher values of SSI reflect more salinity.

$$NDVI = \frac{\rho_{NIR} - \rho_{RED}}{\rho_{NIR} + \rho_{RED}} \rightarrow (2)$$

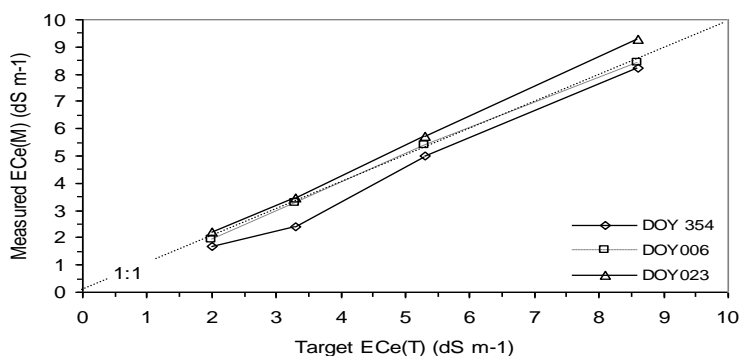
$$SSI = \frac{\rho_{1640} - \rho_{1480}}{\rho_{1000}} \rightarrow (3)$$

where  $\rho_{NIR}$  = reflectance in the near infrared (810-nm),  $\rho_{RED}$  = reflectance in the red (660-nm).  $\rho_{1640}$ ,  $\rho_{1480}$ , and  $\rho_{1000}$  are reflectance at 1640-nm, 1480-nm, and 1000-nm, respectively.

A Student's paired t-Test, with a two-tailed distribution and equal variances assumption was used to test the significance of the difference between treatments at 95 % significance level.

## RESULTS AND DISCUSSION

Soil salinity measured as electrical conductivity of saturated soil paste ( $EC_{eM}$ ) versus the target soil salinity ( $EC_{eT}$ ) was illustrated in figure 2. Generally, the curves of the measured to target salinity were close to the 1:1 line; however, the measured soil salinity tended to be lower in DOY 354 and higher in DOY 23 than the target soil salinity. Differences between target and measured salinity were mostly insignificant, excluding two incidents (one for the 10% yield reduction treatment, DOY 354, and another for the 50% yield reduction treatment, DOY 23). The soil salinity in DOY 6 was very close to the 1:1 line.

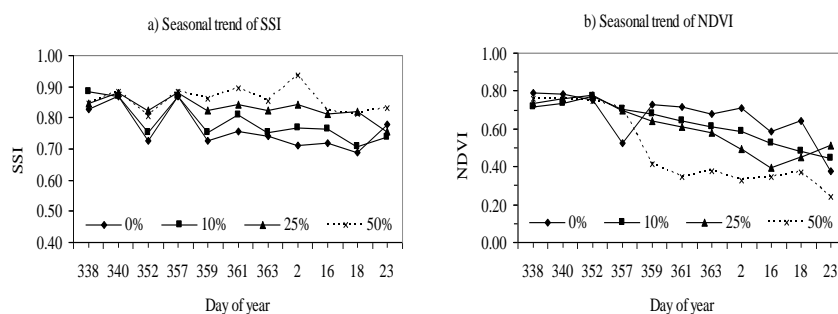


**Figure 2: Soil salinity measured as electrical conductivity [ $EC_e(M)$ ] in response to targeted salinity treatments [ $EC_e(T)$ ] in DOY 364, 2005; 6, 2006; and 23, 2006).**

Salinity stress index (SSI) and normalized difference vegetation index (NDVI) for salinity treatments (0, 10, 25 and 50% yield reduction) are plotted in figure 3. It includes data from day of year DOY 338 (2005) until DOY 23 (2006). SSI (Fig. 3a) was reasonable at DOY 352 and worked very well in the

middle of the season from DOY 259 until DOY 16. It significantly separated the four salinity treatments where higher SSI values were associated with the higher salinity treatments and vice versa, lower SSI values with the low salinity treatments. No significant differences were observed between the 0% and 10% or 25% and 50% treatments before DOY 359. Differences became insignificant between the 25% and 50% treatments after DOY 2. On the other hand, the difference between the 0% and 10% treatments became insignificant later in the season (DOY 16). The question whether other source of growth retardation (i.e. water, nitrogen, etc.) might give a bogus SSI signal of salinity stress could not be answered in this paper and requires further study.

NDVI (Fig. 3b) indicates that salinity treatment started at close to full cover (NDVI $\approx$ 0.8) which eliminated the effect of soil background. In other words, soil background was not the main reason that SSI did not work before DOY 359. Similar to the SSI, NDVI separated the four treatments starting at DOY 359; however, it kept separating them until DOY 18. Generally, NDVI had a decreasing trend during the season which might be attributed to the change in leaf structure and color due to salinity. Nevertheless, the decrease was relatively rapid for the (50%) salinity treatment. Theoretically, SSI is resistant to other sources of stress such as water and nitrogen deficiencies (Stong, 2003). Therefore it would be superior to NDVI in detecting salinity without interference from water or nitrogen.

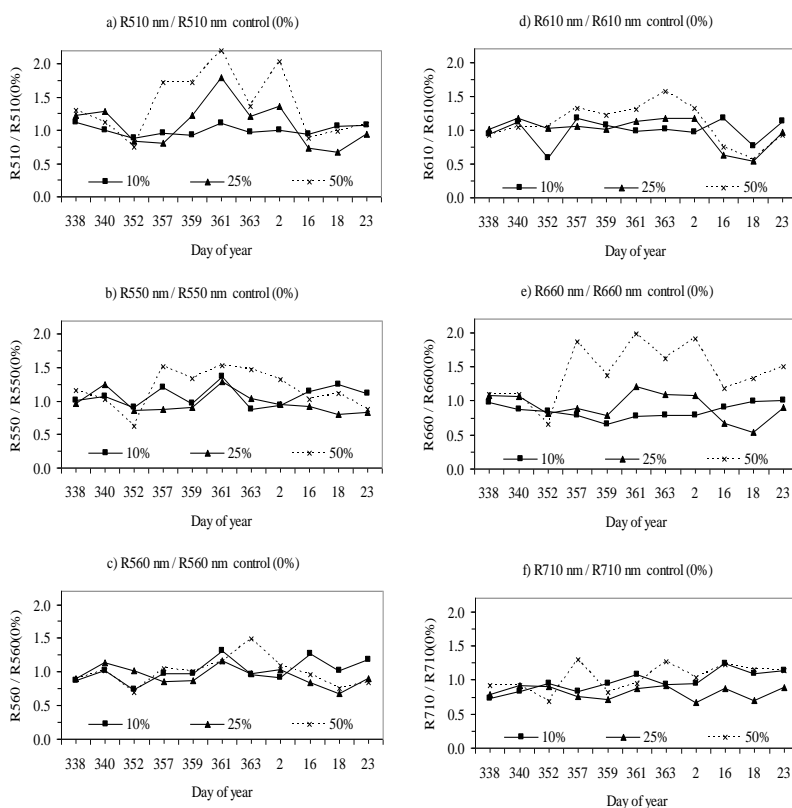


**Figure 3: Seasonal trend of spinach a) SSI and b) NDVI for the four salinity treatments (0, 10, 25 and 50% yield reductions).**

The sudden decrease of NDVI (below 0.4) and the equivalent increase of SSI at DOY 23 (2006) could be due to plant aging. If we assume that water and nitrogen applications were the right amounts; differences in NDVI between treatments could result from the severe effect of salinity on leaves structure and color.

The reflectance at 510 nm 550 nm and 560 nm for salinity treatments (10, 25 and 50% yield reduction) relative to the reflectance of the control treatment (0% yield reduction) at the same wavelengths are plotted against the day of year (DOY) in figure 4. Reflectance at 510 nm (Fig. 4a) was not as good as the SSI or the NDVI in separating the salinity treatments. Significant differences were seen from DOY 359 until DOY 2. The reflectance at 550 nm (Fig. 4b) separated the 50% treatment from the other two (10% and 25%);

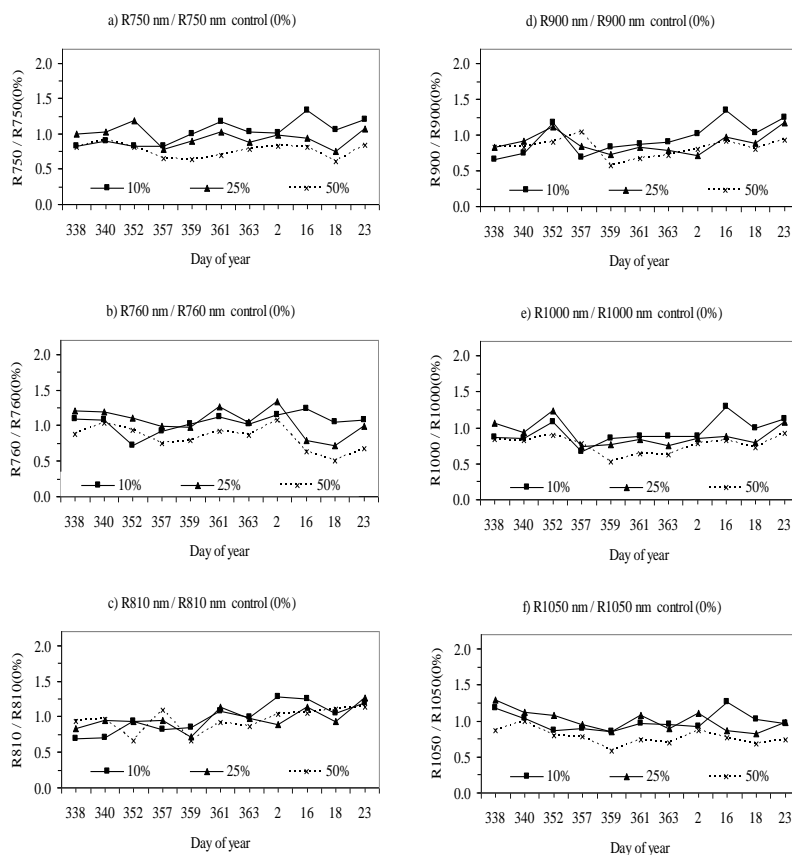
however, differences were insignificant between the 10% and 25% salinity treatments. Salinity treatments were mixed when the reflectance at 560 nm (Fig. 4c) was used. Reflectance at 610 nm, 660 nm and 710 nm for salinity treatments (10, 25 and 50% yield reduction) relative to control treatment (0% yield reduction) demonstrates the relatively higher potential of the 660 nm band (Fig. 4e) for salinity detection compared to the 610 nm (Fig. 4d) and 710 nm (Fig. 4f) bands. It separated salinity treatments very well from DOY 357 until DOY 2; however, treatments (10% and 25%) were mixed before and after that period. The high salinity treatment (50%) was separated from the 10% and 25% treatments as early as DOY 357 until the end. The higher the stress the higher the reflectance at the 660 nm (red) band, which agrees with the fact that healthier and non-stressed plants would show less reflectance in the red and high in the near-infrared. Reflectance at the 710 nm band was not very helpful in differentiating salinity treatments, where reflectance at this band (red edge) is more affected by nutrient stresses. Fluctuation of reflectance at the 710 nm band around 1 with no major spikes could be an indication that no major nutrition stress happened during the experiment.



**50 nm**  
**and c) 560 nm, d) 610 nm, e) 660 nm and f) 710 nm for salinity**  
**treatments 10, 25 and 50% yield reductions relative to control**  
**(0% yield reduction).**



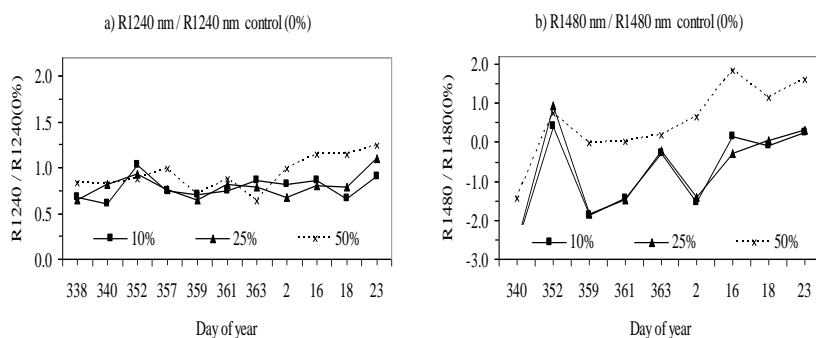
Figure (5) indicated the reflectance at 750, 760, 810, 900, 1000 and 1050 nm for salinity treatments (10, 25 and 50% yield reduction) relative to control treatment (0% yield reduction). Surprisingly, the reflectance at 750 nm (Fig. 5a) was superior to the tested salinity stress index (SSI) itself. It evidently separated the 10, 25 and 50% treatments from DOY 357 until the last day of data measurement; however, differences between treatments were insignificant sometimes. It is a promising part of the spectrum that can be utilized in developing new salinity stress indices. None of the other two bands (760 nm [Fig. 5b] or 810 nm [Fig. 5c]) was capable of showing salinity stress consistently during the season.



**Figure 5: Seasonal trend for reflectance at bands: a) 750 nm, b) 760 nm and c) 810 nm, d) 900 nm, e) 1000 nm and f) 1050 nm for salinity treatments 10, 25 and 50% yield reductions relative to control (0% yield reduction).**

Reflectance at 1000 nm (Fig. 5e) showed similar trend as of the 750 nm band (Fig. 5a). It separated the salinity treatments from DOY 359 until end of season with more insignificant differences between the 25% and 50% treatments later in the season. Again, more thoughts have to be given to that band as well as the 750 nm band. Band 900 nm (Fig. 5d) separated the 10% treatment from the 25% and 50% treatments but it mixed the 25% and 50% treatments. The reflectance at the 1050 nm (Fig. 5f) was not as consistent as the 900 nm or the 1000 nm bands.

Seasonal trends of reflectance at 1240 nm and 1480 nm for salinity treatments (10, 25 and 50% yield reduction) relative to control treatment (0% yield reduction) are shown in figure 6. The 1240 nm band (Fig. 6a) separated the 50% treatment from both the 10% and 25% treatments from DOY 2 until DOY 23 but discrepancy and insignificant differences were evident before DOY 2. On the other hand, the reflectance at 1480 nm (Fig. 6b) indicated clear difference between the 50% treatment and both 10% and 25% treatments during almost the entire season. However, negative values were apparent after starting the salinity treatments. No significant difference was manifested between the 10% and 25% treatments. Being in the water absorption (high absorption and low reflectance) region of the electromagnetic spectrum could confound the ability of the 1480 nm band to detect salinity stress.



**Figure 6: Seasonal trend for reflectance at bands: a) 1240 nm and b) 1480 nm for salinity treatments 10, 25 and 50% yield reductions relative to control (0% yield reduction).**

### SUMMARY AND CONCLUSIONS

The experiment was achieved in Tucson, Arizona, USA (Lat. 32° 16' N, Long. 110° 56' W and Elev. 713m). Spinach (*Spinacia oleracea* L., var. Rushmore) was planted on 28 Oct. (day of year [DOY] 301), 2005 in an alluvial, Silty Clay Loam soil. Authors concluded the following:

- Normalized difference vegetation index gave a good indication of salinity stress but being affected by water and nitrogen deficiencies could limit its use.
- Salinity stress index worked fairly well in the middle of the season when the salinity treatments had significant differences in the NDVI.
- Reflectance at 750 nm had clearer and earlier separation of treatments than the salinity stress index. Similar trend was observed with the 1000 nm band.
- Bands at 660 nm, 760 nm and 1480 nm were more sensitive to the highest salinity treatment (50% yield reduction).
- The reflectance at 750 nm and 1000 nm along with 660 nm, 760 nm and 1480 nm can be integrated to develop new salinity indices.
- Further research to study the effect of other growth retardation sources (water stress, nutrient stress, etc.) on the SSI performance with spinach is required.

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**مراقبة تأثير السبائح بالأجهاد الملحي عن طريق قياس الانعكاس الطيفي  
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لقد أجريت العديد من الأبحاث لتقدير إجهاد النبات المائي أو الإجهاد نتيجة نقص العناصر الغذائية مثل النيتروجين باستخدام النظم المختلفة للإستشعار عن بعد مثل الستلايت، الطائرات، و النظم المحمولة باليد أو علي أنظمة الري المتحركة. ركزت معظم أبحاث الإستشعار عن بعد بصفة عامة علي المحاصيل الرئيسية مثل القطن و القمح في حين لم تحظى محاصيل الخضر بنفس الإهتمام من الباحثين. كما تمت دراسات وافية علي إستخدام الإستشعار عن بعد لتقدير تأثير التربة بالملوحة في حين مازال مجال إستخدام الإستشعار عن بعد لقياس إجهاد المحاصيل لتأثيرها بالأملاح جديد و بدأ في جذب إنتباه الباحثين. لذلك تم إجراء هذا البحث لدراسة إستخدام الإشعاع المنعكس من السبائح في المنطقة المرئية و غير المرئية عند أطوال موجية: 550، 510، 610، 660، 710، 750، 760، 810، 900، 1000، 1050، 1240 و 1480 نانوميتر من الفيض الكهرومغناطيسي بالإضافة إلي معيار الإجهاد الملحي و معيار الخضرة ذو الفرق العياري. و إشملت التجربة أربع معاملات ملوحة بما يكافئ الحصول علي تخفيض للمحصول بنسبة مئوية مقدارها 10% ، 25% و 50%. أظهر معيار الملوحة نتائج مرضية في منتصف الموسم فقط. كما بين معيار الخضرة ذو الفرق العياري فروق بين معاملات الملوحة و قد يكون ذلك لتأثير الملوحة السلبي علي شكل الأوراق و لونها أو لتأثير النباتات بعوامل أخرى غير مقاسة مثل الإجهاد المائي. صعوبة التأكد من عدم تأثير معيار الملوحة بعوامل أخرى غير الملوحة مثل نقص المياه و النيتروجين يستلزم إجراء تجارب مستفيضة في هذا المجال. و بينت النتائج أيضاً أن الانعكاس عند أطوال موجية 750 و 1000 نانوميتر قد تفوق علي المعيار المستخدم في إيضاح الإجهاد الملحي. من الممكن إستخدام الأطوال الموجية السابقة بمفردها أو بالإضافة للأطوال الموجية عند 660، 760 و 1480 نانوميتر لعمل معايير ملوحة جديدة. و يمكن القول ان نتائج هذه التجربة تدعم إمكانية إستخدام الإستشعار عن بعد لتقدير الإجهاد الملحي للسبائح.