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Spectral Reflectance and Digital Image Relations Among Five Aquatic Weeds

James H. Everitt[1], K. Rodney Summy[2], and Chenghai Yang[1]

[1]USDA-ARS
Integrated Farming and Natural Resources Research
2413 E. Highway 83
Weslaco, TX 78596
[2]Department of Biology
University of Texas-Pan American
Edinburg, TX 78539

ABSTRACT

This study reports on the use of an artificial quartz halogen lighting source to facilitate the acquisition of spectral light reflectance measurements and digital multi-spectral imagery of invasive aquatic weeds. Spectral leaf or leaf/stem reflectance measurements were made on five aquatic weeds: Eurasian watermilfoil (*Myriophyllum spicatum* L.), hydrilla [*Hydrilla verticillata* (L. F.) Royle], parrotfeather [*Myriophyllum aquaticum* (J. M. da Conceicao) Vellozo], waterhyacinth [*Eichhornia crassipes* (Mart.) Solms], and waterlettuce (*Pistia stratiotes* L.). Reflectance measurements were studied at five wavelengths of the electromagnetic spectrum: 450 nm (visible blue), 550 nm (visible green), 650 nm (visible red), 680 nm (visible red edge), and 850 nm (near-infrared). Reflectance values differed significantly (P= 0.05) among the species at all five wavelengths. However, more distinct separations among species occurred at the 550 nm, 650 nm, 680 nm, and 850 nm wavelengths. Reflectance differences among species were attributed to variable foliage coloration and vegetative density. Close-range conventional color and color-infrared digital images of leaves or leaves/stems of the five species showed they differed in image tonal response. Reflectance measurements were related to the image tonal response of the plant species on both types of imagery. Supervised image classifications performed on both conventional color and color-infrared images showed the computer generally did an adequate job in identifying the image tonal responses of the weed species.

Additional Index Words: light reflectance, quartz halogen lighting, conventional color digital imagery, color-infrared digital imagery, image analysis, *Myriophyllum spicatum*, *Hydrilla verticillata*, *Myriophyllum aquaticum*, *Eichhornia crassipes*, *Pistia stratiotes*.

Trade names are mentioned for the benefit of the reader and do not imply endorsement of or a preference for the product listed by the United States Department of Agriculture.

Invasive, non-indigenous plant species are a problem in the United States where they have displaced many native plant species. Compared with approximately 17,000 native species, an estimated 5,000 introduced plant species have escaped and now exist in ecosystems of the United States (Pimentel et al., 2005). Nowhere are these biological invasions more evident than in rivers, lakes, and reservoirs. Over the past century numerous weed species have invaded the world's waterways (Barrett, 1989). In 2005, it was estimated that over $110 million was spent annually in the United States to control aquatic weeds (Pimentel et al., 2005).

Spectral light reflectance measurements have been used to characterize the spectral signatures and discriminate among crop, weed, wetland, and aquatic plant species (Gausman et al., 1981; Best et al., 1981; Gausman, 1985; Ullah et al., 2000). Reflectance measurements have also been used to distinguish among woody plant and aquatic plant species and related to their image responses on conventional color and color-infrared (CIR) aerial photographs (Gausman et al., 1977; Everitt et al., 2000; Escobar et al., 2002). The majority of studies using spectroradiometric measurements and acquisition of CIR imagery have been conducted outdoors under natural lighting conditions. The major limitation of this approach is that spectral reflectance measurements and CIR imagery generally cannot be obtained without optimum weather conditions.

Summy et al. (2004) and Jensen (2007) demonstrated the feasibility and use of an artificial light source to facilitate the acquisition of spectral light reflectance measurements and close-range CIR digital imagery under all weather conditions. Recently, Summy and Little (2008) demonstrated the application of this technique for distinguishing a variety of fungal pathogens on several crops under glasshouse conditions. Little information is available on spectral properties of aquatic vegetation using artificial light sources and the use of close-range digital imaging for distinguishing aquatic plant species. Spectral measurements acquired under suitable artificial lighting conditions in a laboratory environment could be particularly useful in evaluating spectral properties of aquatic plants, as such procedures tend to minimize or eliminate atmospheric effects and other types of “noise” which may confound interpretation of spectral data (Jensen, 2007). The objectives of this study were to use artificial lighting to (1) measure the visible and near-infrared (NIR) spectral leaf or leaf/stem reflectance of five exotic aquatic weeds and (2) to evaluate close-range CIR and conventional color digital imagery for distinguishing these weeds and relate their spectral characteristics to their image responses.

MATERIALS AND METHODS

The five aquatic weeds studied in this experiment included: Eurasian watermilfoil (*Myriophyllum spicatum* L.), parrotfeather [*Myriophyllum aquaticum* (J. M. da Conceicao) Vellozo], hydrilla [*Hydrilla verticillata* (L. F.) Royle], waterhyacinth [*Eichhornia crassipes* (Mart.) Solms], and waterlettuce (*Pistia stratiotes* L.). All five species are common introduced weeds found in Texas waterways and are widely distributed in other parts of the United States and the world (USDA, NRCS, 2007). Eurasian watermilfoil and hydrilla are submersed species, parrotfeather is an emergent plant species and waterhaycinth and waterlettuce are floating plant species.

This study was conducted under laboratory conditions. Spectroradiometric reflectance and image acquisition were acquired under artificial light conditions. The lighting source consisted of two heavy-duty 500 W ‘Commercial Electric’ quartz halogen lamps with rectangular reflector housing mounted at a 45° angle approximately 1 m above the floor. The lamps emitted relatively high levels of both visible and near-infrared radiation and produced a rectangular lighting pattern in which luminosity ranged from 114.2 +/- 0.12 fc in the central portion of the light source to 49.9 +/- 0.25 fc at a distance of 1.0 m from the center of the light source (Summy et al., 2004). Reflectance measurements and digital images were obtained in the center of the area illuminated by the light source. A sheet of plywood with flat gray paint was used as the background.

Spectral reflectance of leaves or leaves/stems of each plant species were measured using a FieldSpec dual VNIR spectroradiometer sensitive in wavelengths extending from 350 to 1100 nm and Viewspace Pro software (Analytical Spectral Devices, Inc., Boulder, CO). Each wavelength had a 10 nm bandwidth. For calibration, a remote cosine receptor was used to measure incident irradiation. Reference measurements were taken on a Spectrolon (Analytical Spectral Devices, Inc., Boulder, CO) plate at the time of measurements and converted to % reflectance. Measurements were made on plants from two collection periods: November 12-13, 2008 and December 9-10, 2008. Measurements were made on five randomly selected leaves of waterhyacinth and waterlettuce, and leaves/stems of parrotfeather, hydrilla, and Eurasian watermilfoil. Only mature plant material was measured from each species. The spectroradiometer sensor had an 18° field-of-view and measurements were acquired by holding the sensor probe vertically above the plant material. No shadows occurred in the illuminated area. Measurements of waterhyacinth and waterlettuce were acquired at 5 to 7.5 cm above the leaves, whereas measurements of parrotfeather, hydrilla, and Eurasian watermilfoil leaves/stems were obtained at 2.5 to 5 cm above the foliage due to their smaller surface area. For hydrilla and Eurasian watermilfoil, only plant material occurring on the top of beds at the water surface was used. The surfaces of hydrilla and Eurasian watermilfoil were moist when spectral measurements were acquired. Waterlettuce plants were collected in the Rio Grande near Brownsville, TX, whereas waterhyacinth was collected from an irrigation canal near Weslaco, TX. Hydrilla was collected from Choke Canyon Reservoir near Three Rivers, TX, while Eurasian watermilfoil was collected from Coleto Creek Reservoir near Goliad, TX. Parrotfeather was collected from the Atascosa River in Pleasanton, TX. Five whole waterhyacinth and waterlettuce plants were placed in buckets containing river water, whereas five clumps of leaves/stems of hydrilla, Eurasian watermilfoil, and parrotfeather were placed in ziplock plastic bags on ice and transported back to the laboratory for spectral measurements within 24 hours.

Spectral measurements were studied only from 400 to 900 nm because this range covers the sensitivity of conventional color and CIR digital imagery used for this study. Reflectance measurements were extracted from five wavelengths of the electromagnetic spectrum: 450 nm (visible blue), 550 nm (visible green), 650 nm (visible red), 680 nm (visible red edge), and 850 nm [near-infrared (NIR)]. Data from the five wavelengths were analyzed using the 1-way analysis of variance. Spectral reflectance was the dependent variable and plant species were the independent variable for the analysis. Duncan's multiple range test was used to test statistical significance at the 0.05 probability level among means (Steel and Torrie, 1980).

Conventional color digital imagery of leaves or leaves/stems of the five species was acquired with a Konica Minolta Model A200 digital camera (Konica Minolta Corp., Osaka, Japan). The spectral sensitivity was 400 to 700 nm comprised of the blue (400 to 500 nm), green (500 to 600 nm), and red (600 to 700 nm) spectral bands. Digital CIR imagery of the plant material was obtained with a DuncanTech MS-3100 CIR camera system (DuncanTech, Auburn, CA) equipped with a NI 1424 frame-grabber (National Instruments, Austin, TX). The spectral sensitivity of the camera was 500 to 900 nm. This was comprised of the green (500 to 600 nm), red (600 to 750 nm), and NIR (750 to 900 nm) spectral bands. Images acquired with the conventional color camera were separated into blue, green, and red waveband images, while images obtained with the CIR camera were separated into green, red, and NIR waveband images. Adobe Photoshop 6 (Adobe Systems, Inc., San Jose, CA) was used to separate the wavebands, which were imported as TIFF files into IDRISI 32 v.2 (Clark University, Worcester, MA) for processing. The quality of imagery of the plant material acquired under artificial light was comparable to that of imagery of plants acquired under natural (clear and sunny) lighting conditions (Summy et al., 2004; Summy and Little, 2008).

The conventional color and CIR digital images of the five plant species collected in November 2008 were subjected to a supervised image analysis technique. Four subsamples were selected from each species (same for both images) to be used as training samples. In addition, four subsamples were also selected from background and plant shadows in each image. The Maximum Likelihood classifier was used to classify the two images (IDRISI, Inc., Clark University. Worcester, MA). Background and plant shadows classes were reclassified to form a single class: background/plant shadows. Thus, there were six classes for each image that included: waterhyacinth, waterlettuce, parrotfeather, Eurasian watermilfoil, hydrilla, and background/plant shadows.

RESULTS AND DISCUSSION

Fig. 1 shows mean light reflectance measurements over the 400 to 900 nm wavelength interval for the five weed species for the November 2008 sampling dates. Distinct differences occurred among the species at several wavelengths. Table 1 shows mean light reflectance data for the five species at five wavelengths. At the 450 nm visible blue wavelength, Eurasian watermilfoil had higher reflectance than parrotfeather, hydrilla, and waterhyacinth, but its reflectance value could not be separated from that of waterlettuce. Parrotfeather, hydrilla and waterhyacinth had similar reflectance values at the 450 nm wavelength. For the 550 nm visible green wavelength, waterlettuce had higher reflectance than the other species, whereas hydrilla and waterhyacinth had lower reflectance. Waterlettuce and Eurasian watermilfoil had higher visible red (650 nm) reflectance than the other species. At the 680 nm visible red edge, Eurasian watermilfoil had higher reflectance than the other species while waterhyacinth had lower reflectance. The red edge reflectance value of waterlettuce differed from those of the other species. For the 850 nm NIR wavelength, waterlettuce had higher reflectance than waterhyacinth, Eurasian watermilfoil, and hydrilla, but its reflectance value was similar to that of parrotfeather. Hydrilla and Eurasian watermilfoil had lower NIR reflectance than the other species.

Mean light reflectance measurements over the 400 to 900 nm wavelength interval for the five weed species in December 2008 are shown in Fig. 2. The December 2008 spectral curves for the five species followed a similar trend to the November 2008 curves. Mean reflectance values among the five species in December 2008 differed significantly at the five wavelengths studied (Table 2). At the 450 nm wavelength, Eurasian watermilfoil had higher reflectance than the other species. Waterlettuce had higher reflectance than the other species at the 550 nm wavelength. Hydrilla had lower reflectance than Eurasian watermilfoil, parrotfeather, and waterlettuce at the 550 nm wavelength, but its reflectance value was similar to that of waterhyacinth. At the 650 nm wavelength, waterlettuce and Eurasian watermilfoil had higher reflectance than the other species, while waterhyacinth had lower reflectance. Eurasian watermilfoil had higher reflectance than the other species at the 680 nm wavelength, whereas waterhyacinth had lower reflectance. For the 850 nm

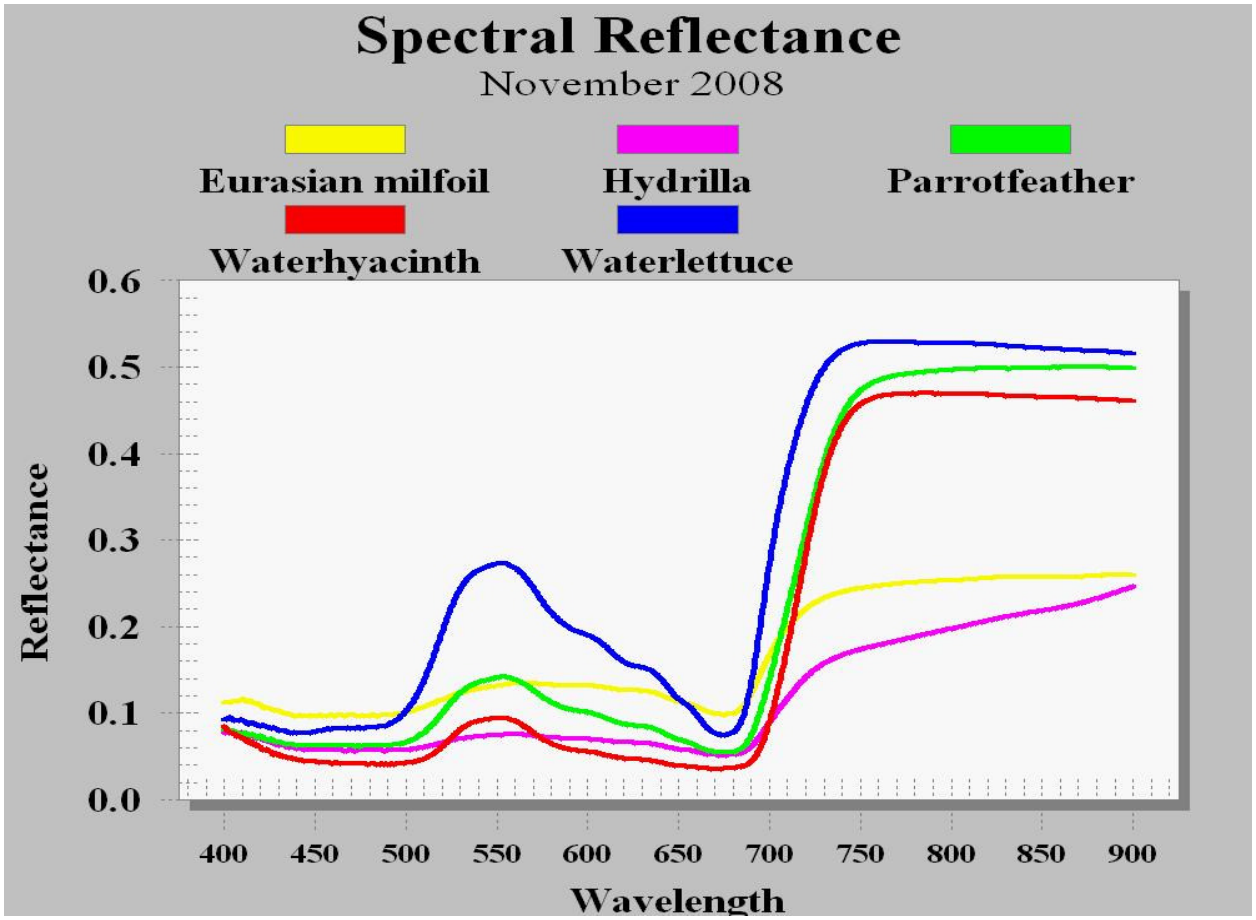

Fig. 1. Mean spectroradiometric laboratory reflectance measurements for leaves or leaves/stems of five aquatic weed species in November 2008.

Table 1. Mean light percent reflectance measurements at five wavelengths for five aquatic weed species for the November sampling dates.

Species	Reflectance values[1] for five wavelengths				
	450	550	650	680	850
Hydrilla	5.7c	7.4c	5.8b	5.2c	25.3c
Milfoil[2]	9.6a	13.2b	11.4a	9.9a	21.8c
Parrotfeather	6.2bc	13.9b	6.9b	5.5c	49.9ab
Waterhyacinth	4.4c	9.4c	3.9c	3.6d	46.4b
Waterlettuce	7.9ab	27.1a	11.6a	7.8b	52.1a

[1]Values within a column followed by the same letter do not differ significantly at the 0.05 probability level, according to Duncan's multiple range test.
[2]Milfoil=Eurasian watermilfoil.

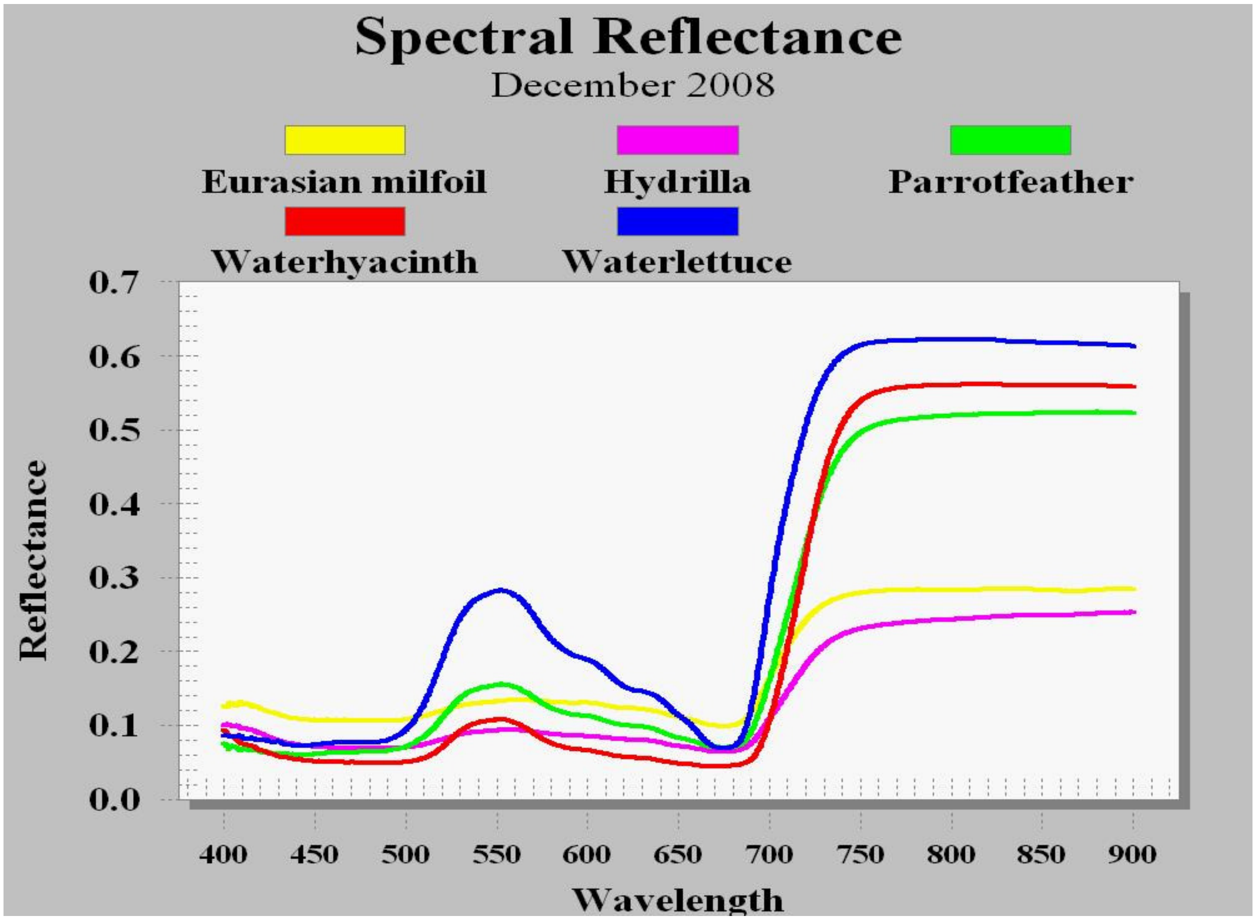

Fig. 2. Mean spectroradiometric laboratory reflectance measurements for leaves or leaves/stems of five aquatic weed species in December 2008.

Table 2. Mean light percent reflectance measurements at five wavelengths for five aquatic weed species for the December sampling dates.

Species	Reflectance values[1] for five wavelengths				
	450	550	650	680	850
Hydrilla	7.1b	9.3d	7.2b	6.5b	24.9c
Milfoil[2]	10.7a	13.1b	11.1a	10.0a	28.3c
Parrotfeather	6.0bc	15.4b	8.2b	7.0b	52.2b
Waterhyacinth	5.1c	10.7cd	4.8c	4.5c	56.0b
Waterlettuce	7.3b	28.0a	11.5a	7.1b	61.8a

[1]Values within a column followed by the same letter do not differ significantly at the 0.05 probability level, according to Duncan's multiple range test.
[2]Milfoil=Eurasian watermilfoil.

wavelength, waterlettuce had higher reflectance than the other species while hydrilla and Eurasian watermilfoil had lower reflectance.

Visible reflectance in vegetation is primarily affected by plant pigments and carotenoids (Myers et al., 1983; Gausman, 1985). Foliage colors varied from the very light green of waterlettuce, to bright green of parrotfeather, to dull gray-green of Eurasian watermilfoil, to dull darker green of hydrilla, to deep dark green of waterhyacinth (Fig. 3A). The darker green foliage (higher chlorophyll concentration) of waterhyacinth and hydrilla reflected less of the green light and absorbed more of the blue, red, and red edge light than the various lighter green foliage of waterlettuce, parrotfeather, and Eurasian watermilfoil (lower chlorophyll concentration) (Myers et al., 1983; Gausman, 1985; Campbell, 1996).

Differences in NIR reflectance among the species were primarily attributed to differences in their vegetative density (Tucker, 1979; Gausman, 1985; Campbell, 1996). Waterhyacinth and waterlettuce had much larger and thicker leaves than the other species, while parrotfeather had a densely leafed stem. In contrast, Eurasian watermilfoil and hydrilla had sparsely leafed stems with gaps and breaks among the leaves. Internal leaf structure measurements were not made, but this could also contribute to the differences in NIR reflectance among the species (Gausman, 1985; Campbell, 1996).

The high green and high NIR reflectance of waterlettuce is in agreement with field canopy reflectance data reported for this species (Everitt et al., 2003). The low red and high NIR reflectance for waterhyacinth, and relatively low red and low NIR reflectance of hydrilla are in general agreement with canopy reflectance data reported for these species (Everitt et al., 1999, 2000). The low NIR reflectance of Eurasian watermilfoil agrees with canopy reflectance data for this species (Everitt et al., 2007). However, Everitt et al. (2007) reported moderate visible reflectance for Eurasian watermilfoil, whereas in the current study this species had high reflectance at the four wavelengths studied. The lower visible canopy reflectance may be related to measurements made of Eurasian watermilfoil beds at the water surface that integrated the plant material and water. Variation in plant phenology could also contribute to these differences.

Fig. 3A shows the conventional color digital image of the leaves or leaves/stems of the five weed species from the November 2008 sampling dates. Conventional color imagery has visible (400 to 700 nm) spectral sensitivity. Therefore, spectral measurements at the 450 nm blue, 550 nm green, 650 nm red, and 680 nm red edge wavelengths are useful for interpreting the conventional color image responses of the weed species (Table 1). The deep dark green image of waterhyacinth (1) was attributed to its low reflectance at the 450 nm, 650 nm, and 680 nm wavelengths. The light green color of waterlettuce (2) was primarily due to it very high reflectance at the 550 nm wavelength. The bright green color of parrotfeather (3) was primarily due to its relatively high reflectance at the 550 nm wavelength. The dull gray-green color of Eurasian watermilfoil (4) was due to its high reflectance at all four visible wavelengths. The dull darker green foliage color of hydrilla (5) was attributed to its relatively low reflectance at the four visible wavelengths.

Few studies have been conducted using conventional color imagery for distinguishing the weeds in this study. However, Jakubauskas et al. (2002) used aerial conventional color videography for mapping waterhyacinth. They reported that waterhycinth had a conspicuous dark green response similar to that shown in the current study. Vittor (2004) used aerial conventional color photography to map submerged Eurasian watermilfoil, but since the plants were under water they had a very dark gray to black signature.

Fig. 3B shows the supervised classification of the conventional color digital image of the five weed species from the November 2008 sampling dates. Color codes for classes in the scene are given in the Fig. 3 caption. The classification did an adequate job in identifying most of the classes, but there were some errors. This was apparent in parrotfeather where the left leaf/stem margins were misclassified as waterhyacinth. There were also minor errors in waterhyacinth and waterlettuce where portions of the leaf margin were identified as parrotfeather. There was some minor confusion between Earasian watermilfoil and hydrilla.

Fig. 4A shows the CIR digital image of leaves or leaves/stems of the five weeds from the November 2008 sampling dates. Since the CIR image has visible/ NIR spectral sensitivity (500 to 900 nm), spectral measurements made at the 550 nm green, 650 nm red, 680 nm red edge, and 850 nm NIR wavelengths can be used to interpret the image responses of the weed species (Table 1). The bright red image tone of waterhyacinth (1) was attributed to its low reflectance values at the 650 nm and 680 nm wavelengths and high reflectance at the 850 nm wavelength. The distinct light pink color of waterlettuce (2) was primarily attributed to its exceptionally high reflectance at the 550 nm wavelength, but its high NIR (850 nm) reflectance also contributed to its image response. The dark pink image of parrotfeather (3) was attributed to its relatively high reflectance at the 550

Fig. 3. Conventional color digital image (A) of leaves or leaves/stems of waterhyacinth (1), waterlettuce (2), parrotfeather (3), Eurasian watermilfoil (4), and hydrilla (5). Plants were collected in November 2008. Supervised computer classification (B) of the leaves or leaves/stems of the five plant species. Color codes for the classification are: red = waterhyacinth, pink = waterlettuce, light green = parrotfeather, orange = Eurasian watermilfoil, dark green = hydrilla, and gray = background/plant shadows.

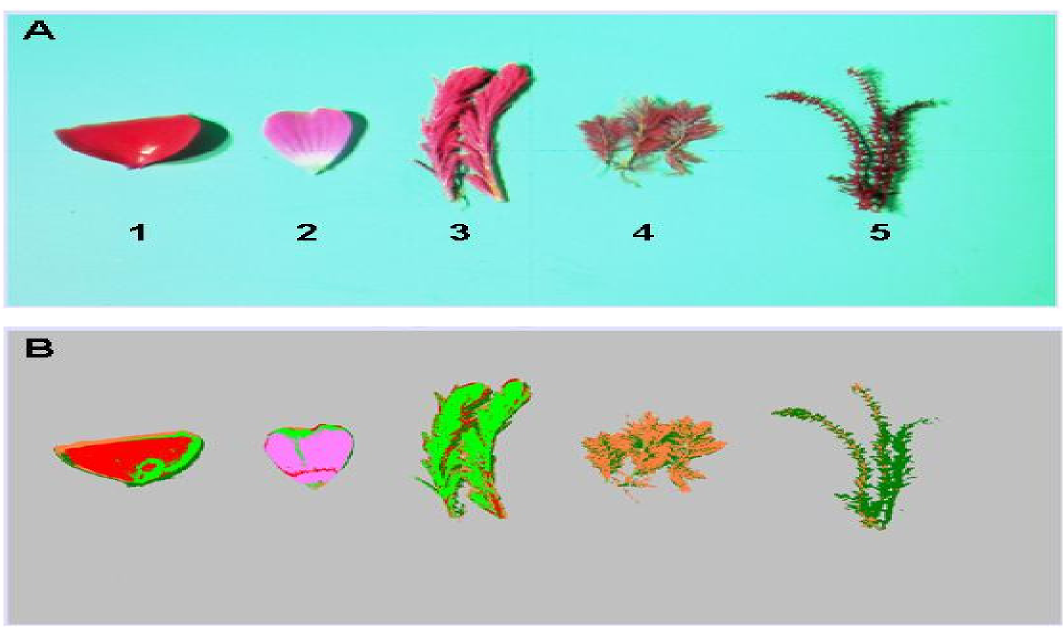

Fig.4. Color-infrared digital image (A) of leaves or leaves/stems of waterhyacinth (1), waterlettuce (2), parrotfeather (3), Eurasian watermilfoil (4), and hydrilla (5). Plants were collected in November 2008. Supervised computer classification (B) of the leaves or leaves/stems of the five plant species. Color codes for the classification are: red = waterhyacinth, pink = waterlettuce, light green = parrotfeather, orange = Eurasian watermilfoil, dark green = hydrilla, and gray = background/plant shadows.

nm wavelength and high reflectance at the 850 nm wavelength. The dull pink image of Eurasian watermilfoil (4) was due to its high reflectance at the 550, 650, and 680 nm wavelengths, and low reflectance at the 850 nm wavelength. The dark reddish-brown tone of hydrilla (5) was primarily due to its low NIR (850 nm) reflectance, but its relatively low reflectance values at the 550 nm, 650 nm, and 680 nm wavelengths also contributed to its image color.

The bright red CIR digital image response for waterhyacinth is in close agreement to that reported for this species in both aerial CIR photography and videography (Everitt et al., 1999 and 2000), and multispectral satellite imagery (Venugopal, 1998; Albright et al., 2004). The reddish-brown CIR image of hydrilla is similar to that reported in aerial CIR photographic and videographic studies (Martyn et al., 1986; Everitt et al., 1999 and 2000). The distinct light pink digital image for waterlettuce concurs with the findings of Everitt et al. (2003), who reported a similar image response for this species in aerial CIR photographic and videographic images. The dull pink digital image of Eurasian watermilfoil is in general agreement to the image response reported for this species in aerial CIR photographic and videographic images (Everitt et al., 2007).

The supervised image classification of the CIR digital image of the five weeds from the November 2008 sampling date is shown in Fig. 4B. Color codes for the classes in the scene are given in the Fig. 4 caption. A qualitative comparison of the classified image to the CIR image suggests that the supervised classification generally identified most of the classes. However, there were some misclassified pixels in each class. This was most evident in waterhyacinth and waterlettuce where portions of the leaf margins and background/plant shadows were misclassified as parrotfeather. There were also some errors in parrotfeather where some of the leaf/stem margins and background/plant shadows were misclassified as waterhyacinth. Some of the background/plant shadows in Eurasian watermilfoil were misclassified as hydrilla. Conversely, some of the background/plant shadows in hydrilla were confused with Eurasian watermilfoil.

CONCLUSIONS

Results from this study showed that an artificial quartz halogen lighting source can be used successfully to acquire all weather spectral light reflectance measurements and close-range CIR and conventional color digital imagery for distinguishing among five invasive aquatic weed species. Reflectance measurements on leaves or leaves/stems of the five species at four visible wavelengths and one NIR wavelength differed significantly. Differences in measured reflectance among the species were related to variable foliage colors and vegetative density. The species had distinct image tonal responses in both CIR and conventional color digital imagery. Spectral measurements could be related to the image tonal responses of the species. A supervised image classification performed on both the CIR and conventional color images showed that the computer did an adequate job in separating most of the image tonal responses of the species. Plant species spectral measurements and their digital image tonal responses obtained under artificial lighting in this study were similar to those reported for the species in field reflectance experiments and studies using conventional aerial photographic, videographic, and satellite imagery. The use of artificial lighting under laboratory conditions may be particularly useful in studies designed to explain the physiological basis of reflectance by different plant species, or temporal changes in reflectance by members of the same species characterized by various levels of stress, senescence, and other factors. The weed species in this study are widely distributed in both the United States and other areas of the world, thus these findings should provide insight into using remote sensing techniques for their discrimination and that of other species.

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