

## Use of Spectral Reflectance Values for Determining Nitrogen, Phosphorus, and Potassium Contents of Rangeland Plants

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### ABSTRACT

This study was carried out to determine nitrogen, phosphorus, and potassium contents of rangeland plants using spectral reflectance value. The measurements were made in 1 m<sup>2</sup> area of different parts of a rangeland. A portable spectroradiometer capable of measuring the wavelength range of 325-1,075 nm of the electromagnetic spectrum was used to collect spectral data. Spectral measurements were made in a rangeland while nitrogen, phosphorus, and potassium content of the plants were determined in a laboratory. Stepwise linear regression was used to select wavelengths to investigate relationships between laboratory analysis results and spectral data. According to the result, significant relationships existed between predicted and measured nutrients, with  $R^2$  values of 0.85, 0.43, and 0.84 for nitrogen, phosphorus and potassium, respectively. While 11 wavelengths ( $R_{609}$ ,  $R_{647}$ ,  $R_{651}$ ,  $R_{654}$ ,  $R_{669}$ ,  $R_{675}$ ,  $R_{676}$ ,  $R_{680}$ ,  $R_{721}$ ,  $R_{727}$ ,  $R_{760}$ ) were used in the equation for estimating nitrogen level, 2 wavelengths ( $R_{675}$ ,  $R_{680}$ ) and fifteen wavelength ( $R_{410}$ ,  $R_{411}$ ,  $R_{417}$ ,  $R_{422}$ ,  $R_{460}$ ,  $R_{463}$ ,  $R_{468}$ ,  $R_{646}$ ,  $R_{651}$ ,  $R_{658}$ ,  $R_{669}$ ,  $R_{670}$ ,  $R_{674}$ ,  $R_{676}$ ,  $R_{682}$ ) were determined for phosphorus and potassium levels, respectively. The results indicated that the changes in nitrogen, phosphorus, and potassium content affected reflectance values of the visible region of spectrum (especially in the red regions) and, therefore, spectral reflectance data could be used to estimate the nitrogen, phosphorus and potassium contents in rangeland plants.

**Keywords:** Plant nutrient elements, Rangeland, Remote sensing, Spectral reflectance.

### INTRODUCTION

Rangelands are the most important roughage sources for ruminants. Forage quality of rangelands should be adequate during the grazing season. Plant nutrient level is one of the most important factors affecting forage quality. Balabanli *et al.* (2010) reported that increased nitrogen applications have positive effects on yield, dry matter (DM), and crude protein (CP). It was reported that the positive effect of nitrogen (N) increased with P (phosphorus) and K (potassium) applications in rangelands (Balabanli *et al.*, 2010). Similar results were reported by Carpici (2011) for nitrogen application.

Macro nutrient elements such as N, P and K account for plant growth and health and affect yield and quality of forage plants that are used in animal feeding (Salisbury and Ross, 1991). Determination of nutrient element levels provides information about nutrient conditions and quality in plants.

Conventional chemical analyses are usually made to determine nutrient element status of plants using laboratory techniques. Analysis of leaf samples in crop plants is usually undertaken with the objectives of diagnosing nutrient deficiencies and imbalances, and evaluating the effectiveness of the current nutrient management programs (Miles, 2010). But, conventional laboratory techniques are expensive, laborious, and time consuming.

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Furthermore, in many cases, the results of the laboratory analyses are sent to the livestock growers after the pastures have been grazed, hence, significantly reducing any benefit to the farmer in terms of feed management and budgeting for the grower's animals (Pullanagari *et al.*, 2011).

The reflectance spectrum of green leaves is considerably affected by their biochemical and biophysical properties. It is possible to extract biochemical information from a continuous vegetation spectrum produced using hyperspectral sensors (Mobasheri and Rahimzadegan, 2012). Investigation of the rangeland quality with remote sensing systems is an important method to improve its efficiency. Determination of leaf biochemical content by remote sensing could be used as an alternative method and could reduce the problems of laboratory analyses (Mutanga *et al.*, 2004). Remote sensing is the acquisition of information about an object without making physical contact with the object. In recent years, remote sensing systems have been used in a variety of applications including agriculture and forestry (Wright *et al.*, 2005). Remote sensing systems provide a measure of light energy reflectance in one or more wavelengths (Wilkerson, 2011).

Ground based systems play an important role in remote sensing. Levizou *et al.* (2004) reported that reflectance spectroscopy has become popular in ecophysiological studies depending on introduction of portable, sensitive, and reliable spectrometers. Because these systems have simplicity, rapidity, and nondestructive nature, they could be effectively used to determine spectral features of objects (Osborne *et al.*, 2002; Takahashi *et al.*, 2002; Bell *et al.*, 2002; Artigas and Yang, 2005; Genc *et al.*, 2008).

Vegetation status could be easily determined with the use of the remote sensing systems, which have several advantages over the conventional laboratory techniques. Ground based remote sensing systems could be used quickly and

inexpensively in determination of nutrient status of vegetations.

Objectives of the present study included: (i) determination of N, P, and K concentrations of rangeland plants and (ii) investigate which wavelengths are more important for N, P, and K levels of plants using the ground based remote sensing systems.

## MATERIALS AND METHODS

Rangeland experiments were carried out in Doyran rangeland located in Antalya, Turkey, between 1-15 May, 2007. During this season, grazing had not started yet. A total of 69 measurements were made in 23 different parts of the rangeland with 3 replications. Reflectance measurements were made using a portable spectroradiometer (FieldSpec® FR, Analytical Spectral Devices Inc., Boulder, Colorado, USA) capable of measuring the wavelengths of 325-1,075 nm of the electromagnetic spectrum (Castro-Esau *et al.*, 2006; Albayrak, 2008). However, due to the observed low signal to-noise ratio at wavelengths shorter than 400 nm and longer than 900 nm, measurements were evaluated for the wavelengths ranging from 400 nm to 900 nm (Han and Rundquist, 2003; Han, 2005; Lin and Liquan, 2006). Also, all obtained spectrums were visually evaluated using the RS3 software (Analytical Spectral Devices, Inc., Boulder, Colorado) (Darvishsefat *et al.*, 2011).

Canopy reflectance measurements were made during clear days between 10.00 am and 11.30 am (Starks *et al.*, 2006) in 69 distinct parts of the rangeland. During the measurements, fiber optic cable and portable computer were connected to the spectroradiometer and reflectance measurements were made for calibration with white reference panel (spectralon). Reference panel has reflective features of almost the entire light that come to the surface. The optical sensor of the spectroradiometer was mounted at 1.5 m

above vegetation surface in the plots and measurements were made with 10° field of view (Albayrak, 2008; Basayigit and Senol, 2009). Five independent measurements at each part of the rangeland were taken to obtain an averaged value. In order to decrease the measurement errors white reference panel measurement was made in each three measurements.

Plants in each measurement area were clipped after the reflectance measurements and samples were dried at 65°C for 48 hours (Brink *et al.*, 2003; Halgerson *et al.*, 2004). While total N content of the samples were analyzed according to a modified Kjeldahl method, dry ashing method was used for P and K analysis (Kacar and Inal, 2008).

ViewSpec<sup>TM</sup> Pro software (Analytical Spectral Devices Inc., Boulder, CO) was used to view and average the five reflectance. All statistical analyses were conducted using stepwise regression analysis implemented in MINITAB statistical program. In this method, wavelengths associated with plant N, P, and K levels were determined and regression equations were developed using the selected wavelengths (Ozyigit and Bilgen, 2011).

## RESULTS

According to the results, the concentrations of N, P, and K varied among

the measured areas of the rangeland. While the minimum, maximum, and mean values of N concentrations were determined as 0.616, 3.668, and 1.51%, respectively, the same values were determined for phosphorus as 0.039, 0.257, and 0.10%. Also, K concentrations of samples ranged between 0.355 and 2.214 %.

Regression equations composed of wavelengths related with the N, P, and K levels and the corresponding  $R^2$  values are shown in Table 1. As a result of stepwise regression analysis, 11 wavelengths ( $R_{609}$ ,  $R_{647}$ ,  $R_{651}$ ,  $R_{654}$ ,  $R_{669}$ ,  $R_{675}$ ,  $R_{676}$ ,  $R_{680}$ ,  $R_{721}$ ,  $R_{727}$ ,  $R_{760}$ ) for N, 2 wavelengths ( $R_{675}$ ,  $R_{680}$ ) for P, and 15 wavelengths ( $R_{410}$ ,  $R_{411}$ ,  $R_{417}$ ,  $R_{422}$ ,  $R_{460}$ ,  $R_{463}$ ,  $R_{468}$ ,  $R_{646}$ ,  $R_{651}$ ,  $R_{658}$ ,  $R_{669}$ ,  $R_{670}$ ,  $R_{674}$ ,  $R_{676}$ ,  $R_{682}$ ) for K levels were determined (Table 1).

Regression analyses showed that 11 wavelengths were associated with N level (Table 1). Among these wavelengths, 8 were in red regions [601-700 nm ( $R_{609}$ ,  $R_{647}$ ,  $R_{651}$ ,  $R_{654}$ ,  $R_{669}$ ,  $R_{675}$ ,  $R_{676}$ ,  $R_{680}$ )] of the spectrum, while 3 were in NIR (Near Infrared Reflectance) regions [701-900 nm ( $R_{721}$ ,  $R_{727}$ ,  $R_{760}$ )]. Coefficient of determination ( $R^2$ ) of the equation composed of these wavelengths was 0.85 and RMSE (root mean square error) was 0.0774. Also, 2 wavelengths (675 nm, 680 nm) from the red region of the spectrum were associated with P, with  $R^2$  of the calculated equation as 0.43

**Table 1.** Regression equations and  $R^2$  values for N, P, and K levels in rangeland conditions.

	Equations	$R^2$	RSME <sup>a</sup>
N <sup>b</sup>	$N = -0.426 + (-630xR_{647}) + (296xR_{680}) + (517xR_{651}) + (-569xR_{675}) + (244xR_{654}) + (66.4xR_{609}) + (44.5xR_{760}) + (-141xR_{727}) + (-280xR_{669}) + (86.4xR_{721}) + (381xR_{676})$	0.85**	0.0774
P <sup>c</sup>	$P = 0.0316 + (42.5xR_{680}) + (-42.6xR_{675})$	0.43**	0.0010
K <sup>d</sup>	$K = 0.759 + (-417xR_{646}) + (410xR_{651}) + (-458xR_{669}) + (147xR_{682}) + (140xR_{417}) + (-423xR_{410}) + (334xR_{670}) + (1059xR_{460}) + (-716xR_{468}) + (-410xR_{674}) + (225xR_{658}) + (-480xR_{463}) + (164xR_{422}) + (178xR_{676}) + (256xR_{411})$	0.84**	0.0550

\*\* $: P < 0.01$ , <sup>a</sup> Root Mean Square Errors. <sup>b</sup> Nitrogen, <sup>c</sup> Phosphorus, <sup>d</sup> Potassium



and *RMSE* of 0.0010 (Table 1).

Fifteen wavelengths were selected for potassium. While 7 of these wavelengths ( $R_{410}$ ,  $R_{411}$ ,  $R_{417}$ ,  $R_{422}$ ,  $R_{460}$ ,  $R_{463}$ ,  $R_{468}$ ) were in the blue region (400-500 nm) of spectrum, 8 wavelengths ( $R_{646}$ ,  $R_{651}$ ,  $R_{658}$ ,  $R_{669}$ ,  $R_{670}$ ,  $R_{674}$ ,  $R_{676}$ ,  $R_{682}$ ) were placed in red region.  $R^2$  and *RMSE* values of the equation were calculated as 0.84 and 0.0550, respectively. Regression analysis of canopy reflectance measurements is shown in Figures 1, for N, P, and K, respectively.

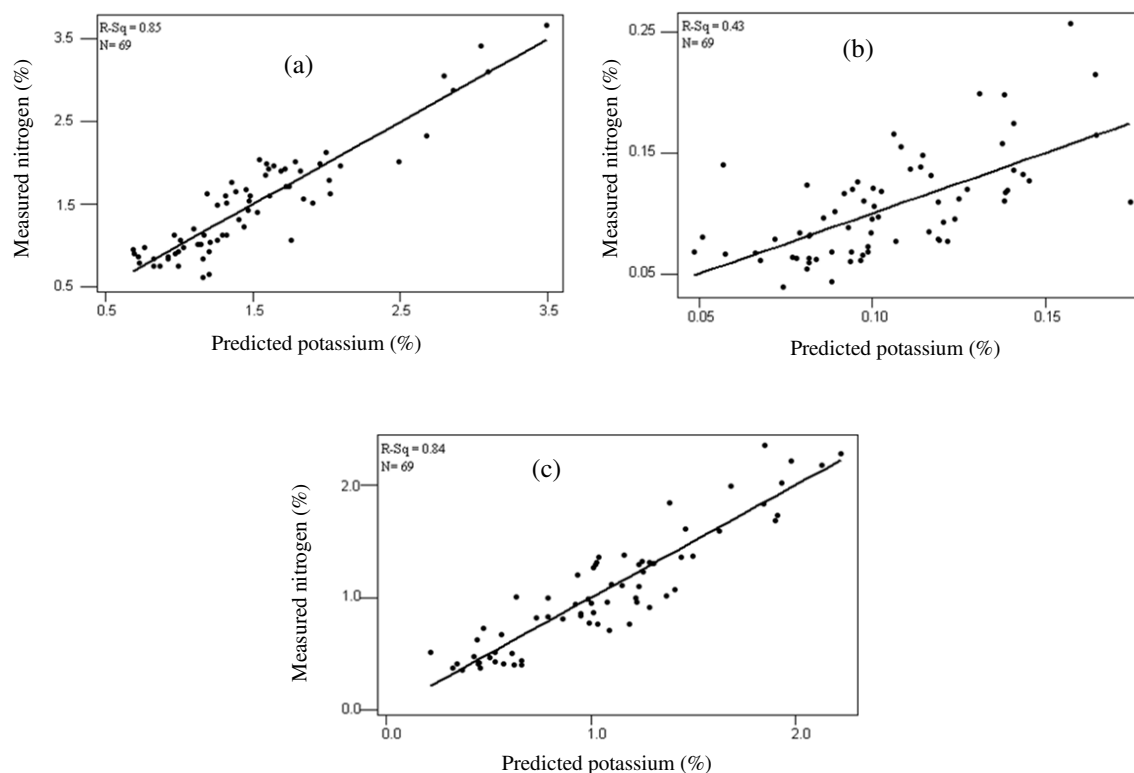
## DISCUSSION

Vegetation reflectance values are successfully used for different purposes such as determination of biomass productivity (Tarr *et al.*, 2005; Cho *et al.*, 2007), leaf area index (Gitelson *et al.*, 2003; Darvishzadeh *et al.*, 2008), vegetation cover and species identification (Zeng *et al.*, 2000; Ghasemloo *et al.*, 2011), plant pigment identification studies (Merzlyak *et al.*, 2003) and other plant components (Curran, 1989), health status of vegetation (Reeves *et al.*, 2001) and green herbage yield (Gianelle and Vescovo, 2007).

Also, in many studies, remote sensing systems have been used to determine nutrient content of plants and successful results have been acquired (Mutanga *et al.*, 2003; Porder *et al.*, 2005; Basayigit *et al.*, 2009; Winterhalter *et al.*, 2012). According to this study results, significant relationships were determined between NPK levels and reflectance values in spectral reflectance measurements of Doyran rangeland located in Antalya. Equations with high  $R^2$  value were found for nitrogen (0.85) and potassium (0.84). Also, red region wavelengths, which were included in the regression equations, had significant role in determination of nitrogen and potassium levels. According to results of this study, *RMSE* values of the models obtained for prediction of N, P, and K were close to zero. Low *RMSE* values verified reliability of the models (Liu *et al.*, 2003).

In this study, significant relationships were determined between nitrogen levels and reflectance values in red and NIR wavelength regions of the spectrum. Several studies on different plant species have indicated that two-waveband reflectance ratios of plant leaves correlated more closely with leaf chlorophyll and leaf N concentration, compared with leaf reflectance in a single narrow waveband. Plant canopy reflectances in the visible (400–700 nm) and NIR (700–900 nm) wavelengths of spectrum are primarily influenced by chlorophyll content and leaf cell structure, respectively (Starks *et al.*, 2006). Chlorophyll absorbs light in the red region of the visible spectrum, therefore, low reflection occurs in this region. Chlorophyll concentration decreases in the case of nitrogen deficiency while reflections increase in red region of spectrum (Daughtry *et al.*, 2000). In another study, Lamb *et al.* (2002) reported that leaf reflectance in red-edge range of wavelengths (690–740 nm) could be used to predict leaf nitrogen concentration and total nitrogen content of ryegrass (*Lolium multiflorum* Lam.). This shows that, red region reflections change due to the nitrogen content of plants. Starks *et al.* (2006) found that crude protein content (thereby nitrogen content) closely correlated with red and NIR region reflections in bermudagrass (*Cynodon dactylon* L. Pers.) pastures. In other study, significant relationships were determined between nitrogen ratio and reflectance values (Mutanga *et al.*, 2004). Mutanga *et al.* (2005) determined close relationships between reflectance values in 748 nm wavelength in NIR region of spectrum and nitrogen levels.

Limited studies exist in the literature on the relationships between phosphorus and potassium levels and reflectance values in rangeland vegetation. In the present study, it was found that significant relationships existed between phosphorus levels and red region wavelengths, while potassium levels correlated with blue and red region. Mutanga and Kumar (2007) showed that



**Figure 1.** Relationship between laboratory-measured N concentrations (a) P concentrations (b) K concentrations (c) and the predicted values based on the reflectance data obtained in rangeland conditions.

bands in the visible and SWIR (small wavelength infrared reflectance) regions were more sensitive to phosphorus levels ( $R^2 = 0.63$ ) in grass rich pasture. Also, in another study, Mutanga *et al.* (2004) reported that phosphorus and potassium elements, which are responsible for both the photosynthetic process and the tissue composition of plants, affected reflection and absorption in the visible region of spectrum. Bogrekci and Lee (2005) reported that spatial variation in actual and predicted maps of phosphorus variability could be represented using diffuse reflectance spectroscopy in the UV, VIS, and NIR regions.

Canopy light reflectance properties based mainly on the absorption of light at a specific wavelength are associated with specific plant characteristics. The spectral

reflectance in the visible (VIS) wavelengths (400–700 nm) depends on the absorption of light by leaf chlorophyll and associated pigments such as carotenoid and anthocyanins (Babar *et al.*, 2006). Using visible region of spectrum i.e. red (500–600 nm) to green (600–700 nm) reflectance ratio, Gamon and Surfus (1999) suggested that prediction of anthocyanins content was possible. Phosphorus level of plants is one of the most important factors that affect anthocyanin content. Phosphorus deficiency may lead to high anthocyanin levels and, consequently, purple discoloration appears in the leaf margins of plants (Osborne *et al.*, 2002). Salisbury and Ross (1991) reported that anthocyanin absorbs energy in the green region while reflecting in the red and blue regions of the spectrum.



Also, chlorophyll concentrations and content change due to potassium levels in plants. While Oosterhuis and Bednarz (1997) reported that potassium deficiency caused reduction in chlorophyll a and total chlorophyll concentration in cotton, Lamrani *et al.* (1996) demonstrated that different potassium nutrition levels have different effects on chlorophyll a and b in cucumber. Also Zhao *et al.* (2001) reported that the translocation of photosynthates from leaves to fruits was restricted by potassium deficiency during cotton development. As a result of this situation, while hexose, sucrose and starch contents increased in leaves, sucrose and starch contents of stems and floral buds decreased. This caused lower chlorophyll concentration and the poor development of leaf anatomy and chloroplast ultra structure. These alterations cause changes of reflectance features in plants.

### CONCLUSIONS

This study was conducted to investigate whether nitrogen, phosphorus and potassium content could be determined by the use of spectral reflectance values. According to the result of the study, significant relationships existed between red ( $R_{609}$ ,  $R_{647}$ ,  $R_{651}$ ,  $R_{654}$ ,  $R_{669}$ ,  $R_{675}$ ,  $R_{676}$ ,  $R_{680}$ ) and NIR ( $R_{721}$ ,  $R_{727}$ ,  $R_{760}$ ) regions and nitrogen level. Also results showed that spectral reflectance values in  $R_{675}$  and  $R_{680}$  wavelengths were associated with phosphorus levels and reflectance values in blue ( $R_{410}$ ,  $R_{411}$ ,  $R_{417}$ ,  $R_{422}$ ,  $R_{460}$ ,  $R_{463}$ ,  $R_{468}$ ) and red ( $R_{646}$ ,  $R_{651}$ ,  $R_{658}$ ,  $R_{669}$ ,  $R_{670}$ ,  $R_{674}$ ,  $R_{676}$ ,  $R_{682}$ ) region of spectrum were useful to identify potassium levels. The results indicated that differences in nutrient concentration largely influence the spectral reflectance and, thus, revealed that the remote sensing systems could be used to estimate the nitrogen, phosphorus, and potassium concentration in rangeland plants.

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## کاربرد اندازه طیف بازتابی برای تعیین مقدار نیتروژن، فسفر و پتاسیم در گیاهان مرتعی

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### چکیده

هدف این پژوهش تعیین مقدار نیتروژن، فسفر و پتاسیم گیاهان مرتعی با اندازه گیری طیف بازتابی بود. اندازه گیری ها در مساحت های یک متر مربعی در بخش های مختلف یک مرتع انجام شد. برای جمع آوری داده های طیفی، از یک دستگاه تابش سنج طیفی قابل حمل که قادر به اندازه گیری طول موج ۳۲۵-۱۰۷۵ نانو متر از طیف مغناطیسی بود استفاده شد. اندازه گیری های طیفی در مرتع انجام شد ولی مقدار نیتروژن، فسفر، و پتاسیم گیاه در آزمایشگاه تعیین شد. برای انتخاب طول موج ها و بررسی رابطه بین اندازه گیری های آزمایشگاهی و داده های طیفی، رگرسیون خطی گام به گام به کار رفت. نتایج نشان داد که رابطه بین اعداد پیش بینی شده و اندازه گیری شده عناصر غذایی از نظر آماری معنی دار بود و ضریب تبیین ( $R^2$ ) آن رابطه برای نیتروژن، فسفر، و پتاسیم به ترتیب ۰.۴۳، ۰.۸۵ و ۰.۸۴ بود. برای نیتروژن ۱۱ طول موج (R609, R647, R651, R654, R669, R675, R676, R680, R721, R727, R760) در معادله برآورد مقدار آن عنصر به کار رفت در حالی که برای فسفر ۲ طول موج (R675, R680) و برای پتاسیم ۱۵ طول موج (R410, R411, R417, R422, R460, R463, R468, R646, R651, R658, R669, R670, R674, R676, R682) در معادله بود. نتایج نشان داد که تغییرات مقدار نیتروژن، فسفر، و پتاسیم روی اندازه بازتاب در بخش قابل رویت طیف (به ویژه در بخش قرمز) تاثیر میگذارد و بنا بر این داده های طیف بازتابی را می توان برای برآورد مقدار نیتروژن، فسفر و پتاسیم در گیاهان مرتعی به کاربرد.