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USING CANOPY INDICES TO QUANTIFY THE ECONOMIC OPTIMUM NITROGEN RATE IN SPRING WHEAT

Nahuel I. Reussi Calvo,* H. Sainz Rozas, H. Echeverría, and N. Diovisalvi

N.I. Reussi Calvo and N. Diovisalvi, Laboratorio de suelos FERTILAB, Moreno 4524, CP 7600, Mar del Plata, Buenos Aires, Argentina; H. Sainz Rozas and H. Echeverría, Unidad Integrada Balcarce, Route 226 km 73.5, CC 276, CP 7620, Balcarce, Argentina. *Corresponding author (nreussicalvo@laboratoriofertilab.com.ar).

ABSTRACT

In-season nitrogen (N) applications to spring wheat (*Triticum aestivum L.*) may increase profits and improve N fertilizer accuracy. The objectives were to develop a calibration tool employing Green Seeker Green 506 (NDVI) and SPAD 502 chlorophyll meter (SPAD) measurements for calculating the differential from economic optimum N rate (dEONR) at growth stages (Z22, Z24, Z31 to Z39), and provide N rate algorithms for use in applying variable rate N fertilizer. Sensing was conducted in N rate trials over 3 year encompassing 10 site-years across Southeastern Buenos Aires Province (Argentina). The relationship between sensor indices and dEONR was evaluated by fitting quadratic-plateau (QP) regression models. Statistically significant QP models were determined at Z24, Z31 and Z39 growth stage. Relative canopy index models [rSPAD and normalized difference vegetative index (rNDVI)] reduced variation and improved the calibration of measured N stress with the dEONR. For Z31 and Z39, the rSPAD had the best goodness of fit statistics when compared to rNDVI (adjR^2 = 0.67 and 0.57 at Z31, and 0.68 and 0.52 at Z39, respectively). However, adjustment at Z24 was higher for rNDVI (adjR^2 = 0.53 and 0.61 for rSPAD and rNDVI, respectively). A single QP model to estimate with 58% confidence the dEONR was adjusted for the Z31 and Z39 growth stages. This indicates the same calibration for N rate determination based on rSPAD or rNDVI values can be used during stem elongation the spring wheat. This model can be used as an N rate algorithm for applying N fertilizer in-season.

Key words: chlorophyll meter; remote sensor; diagnose; nitrogen, wheat.

Abbreviations: adjR^2, adjusted R^2; dEONR, nitrogen rate differential from the economic optimum nitrogen rate; EONR, economic optimum nitrogen rate; LCL, 95% lower QP model confidence limit; N, nitrogen; $N0_3$-N, nitrate-nitrogen; NH_4^+-N, amonio-nitrogen; Nan, anaerobically incubated N; NDVI, normalized difference vegetative index; NNI, nitrogen nutrition index; NUE, nitrogen use efficiency; QP, quadratic-plateau; rNDVI, relative NDVI; rSPAD, relative SPAD; SEB, Southeastern Buenos Aires; SOM, soil organic matter; UCL, 95% upper QP model confidence limit.

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INTRODUCTION

Nitrogen (N) is the nutrient that most often limits crop production. The worldwide fertilizer NUE for cereal production is approximately 33%, and the fertilizer N lost represents a value of $15.9 billion (USD) (Raun and Johnson, 1999). In Mollisols of the Pampas Argentina, the most extended methodology to diagnose N fertility for spring wheat (*Triticum aestivum* L.) is mainly based on the determination of soil nitrate N (NO_3^--N) content (0-60 cm depth) at sowing (Calviño et al., 2002; Barbieri et al., 2012; Reussi Calvo et al., 2013). To use it, different N availability thresholds (soil + fertilizer) have been suggested, which varies according to the area, farming systems, and target yield (Barbieri et al., 2012; Garcia et al., 2010). However, the application of this diagnosis method does not consider the impact of input costs and their relation to the price of the product (input-output ratio). Alvarez (2008) reported that the use of fixed thresholds N allowed achieve positive net margins of investment in fertilization only in years with favorable price relationships. Barbieri et al. (2009a) found that fertilization at tillering in spring wheat decreased N optimum economic dose (EONR) compared with fertilization at sowing, being lower for French than for traditional varieties. Furthermore, excess NO_3-N effect generated by the application of N above the optimal level not only represents an economic loss due to lower EUN but also a potential source of contamination of both aquatic ecosystems and groundwater (Ladha et al., 2005).

The Pampas region in Argentina (30° to 40º S and 57º to 66º O) is known as one of the most important world grain productive areas, with wheat, corn (*Zea mays* L.) and soybean (*Glycine max* (L.) Merr.) as its main crops (Satorre and Slafer, 1999). Southeastern Buenos Aires Province (SEB) is one of the major wheat production sub-regions with a cropped area of 0.7 million ha, and a production of 2.5 million tons (Figure 1). This region is characterized by a very low probability of water deficit (less than 5%) from sowing to heading stage of wheat (Reussi Calvo and Echeverría, 2006). Thus, split N application can maximize yield and NUE (Barbieri et al., 2008, Velasco et al., 2012). The analysis of soil samples enables to estimate the EONR at tillering (Barbieri et al., 2009a), but the sampling and analytical procedures can be time-consuming compared to the use on methodologies based of indices on the canopy.

(Please, place Figure 1 here)

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Chlorophyll meter Minolta SPAD® 502 (SPAD) readings have also been proposed to diagnose N nutrition. The method relies on the strong relationship between SPAD readings and leaf N concentration, which has often been reported in many species (Schepers et al., 1992; Waskom, 1996) as well as for spring wheat (Gandrup et al., 2004). However, SPAD readings are affected by numerous factors unrelated to N availability (Blackmer and Schepers, 1995; Gandrup et al., 2004), suggesting that normalization procedures are needed to standardize reading in order for the SPAD to be a practical N management tool. This normalization produces a relative SPAD value (rSPAD), sometimes called a sufficiency index (Schepers et al., 1992). This normalization has been suggested for various sensors and crops by different researchers (Gandrup et al., 2004; Scharf et al., 2006; Ziadi et al., 2008). For SEB conditions, Gandrup et al. (2004) reported a high association between SPAD or rSPAD and crop yield during the stem elongation stage. Scharf et al. (2006) determined that SPAD values correlated well to the economic optimum N rate (EONR) and yield response to N in corn. Similar response was documented by Hawkins et al. (2007) from V_{15} to R_1 growth stages for corn crop (Ritchie et al., 1993).

Another alternative for monitoring the N status of the crop is the use of multispectral remote sensing, which one of the most widespread worldwide is Green Seeker Green 506 (GS-506). The index most commonly used is normalized difference vegetative index (NDVI). Green Seeker calculates NDVI using red (650 ± 10 nm) and NIR (770 ± 15 nm) light. This index has proven to be a useful tool to indirectly obtain values of photosynthetic efficiency, productivity potential and potential crop yields (Raun et al., 2001; Bonfil et al., 2005; Melchiori et al., 2007; Shiratsuchi et al., 2011). As for the SPAD, some authors recommend the normalization of NDVI readings (Barker and Sawyer, 2010; Clay et al., 2012). For corn, Dellinger et al., (2008) determined a higher association between NDVI and EONR values at V_6-V_7 growth stage. Although numerous studies indicate the ability of SPAD and NDVI for monitoring the N status of wheat, there is little information on the use of these tools to define EONR in spring wheat.

Different models of response to applied N have been used to define EONR, with no consensus among researchers on which are the most convenient (Nelson et al. 1985, Barreto and Westerman, 1987, Blackmer and Meisinger, 1990). However, recent studies indicated that the use of quadratic-plateau model produce the most rational results from the agronomic standpoint (Barbieri et al., 2009a; Barker and Sawyer, 2010).

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The objective of this study was to develop a calibration tool employing SPAD and NDVI measurements for calculating the EONR at diverse growth stages for spring wheat at SEB.

MATERIALS AND METHODS

A total of 10 experiments were conducted under no-tillage in 2010, 2011 and 2012 in fields with different farming histories in Southeastern Buenos Aires Province, Argentina (from 34°41´ S, 58°27´W to 38°23´ S, 58°40´W). This area has an average annual temperature of 13.8°C; and an average rainfall of 870 mm, 45% of which occurs during the wheat growing seasons. Early-season rain (June-September) is lower than potential evapotranspiration in 3 of 30 yr. Late-season rain (October-December) is lower than potential evapotranspiration in 26 out of 30 yr (Calviño and Sadras, 2002). The experimental sites were located in Maipú, General Madariaga, Balcarce, Miramar, Pieres and Lobería counties (Figure 1). Predominant soils type were Typic Argiudolls with loam surface texture, clay loam texture in the underlying horizon, and sandy loam texture a sandy-loam texture below 110-cm depth (C horizon). Table 1 shows some soil characteristics of the experimental sites.

(Please, place Table 1 here)

At each site, the experimental design was a randomized complete block (RCB) with three replications and five treatment levels. Treatments were six N rate levels (0, 50, 100, 150, 200 and 250 or 300 kg N ha^{-1}). At planting time, applications of 25 kg P ha^{-1} and 20 kg S ha^{-1} were performed for avoiding phosphorus (P) and sulfur (S) nutrient deficiencies. The P and S fertilizers were applied at planting. Nitrogen fertilizer was surface broadcasted urea (46% N) at 2- or 3-leave stage. The experimental unit size was 30 m^2 (3 m wide by 10 m long). Wheat planting date ranged from 20 May through 20 June, which is considered optimal for this area. Plant populations at harvest ranged from 220 to 260 plants m^{-2}. Weeds, pests and fungal diseases were controlled using appropriate pesticides at recommended rates.

Soil samples were taken at sowing at 0-20, 20-40 and 40-60 cm depths. At the topsoil (0-20 cm), four parameters were determined: soil organic matter (SOM) content, pH, anaerobically incubated (Nan), and

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NO_3^--N. For the subsoil samples (20-40 and 40-60 cm), only NO_3^--N was determined. Soil NO_3^--N was extracted with potassium chloride (KCl) and determined by colorimetry using a UV-VIS spectrophotometer (Keeney and Nelson, 1982). In order to determine N availability (kg ha^{-1}) in the first 60 cm of soil depth, an average bulk density of 1.2 Mg m^{-3} was assumed (Fabrizzi et al., 2005). Soil organic matter was determined by Walkley and Black method (1934); pH was measured with electrode in a 1:2.5 (soil-water) suspension (Thomas and Hargrove, 1984). Anaerobically incubated N was obtained by soil incubation in anaerobic conditions for 7 days at 40°C, and then produced NH_4^+-N was determined by steam microdistillation (Bremner and Keeney, 1965), as proposed by Gianello and Bremner (1986).

Wheat plant sensing was conducted using a SPAD (Konica Minolta, Japan) and GS-506 (hardware rev. G-K, soft ware ver. 1.6.10) at the Z22 (two tillers), Z24 (four tillers), Z31 (one visible node) and Z39 (visible flag leaf ligule) according to Zadoks et al. (1974). The measurements of SPAD were collected from the uppermost leaf with a fully exposed leaf ligule, midway between the leaf edge and midrib (Peterson et al., 1993). Average SPAD values were obtained by measuring 20 plants from the center of the plot at each N rate level evaluated. The measurements of NDVI were collected in the center of the plots at a constant speed (1.3 m s^{-1}) and distance above the canopy (60–90 cm) while collecting reflectance data. The active sensor was positioned perpendicular to the row in the nadir position (0º angle) at the middle plot of each N rate. Relative indices for each site-year N rate were calculated using the average index value of each sensor divided by the average sensor index value from the highest N rate within each site-year. Relative indices are indicated with a prefix "r".

At maturity, for each plot, harvesting was carried out by cropper, and grain yield was determined by harvesting an area of 6 m^2 corrected to 140 g kg^{-1} grain moisture content.

Statistical analyses were conducted using SAS software (SAS Institute, 1988). Wheat grain yield response to applied N fertilizer was determined for each site-year by using PROC GLM ($P \leq 0.10$). The PROC NLIN procedure was then used to fit regression models for each site-year identified as responsive to applied N. The model statistically significant and possessing the highest coefficient of determination (R^2) and lowest root mean square error (RMSE) was selected. When R^2 values were similar, the quadratic-plateau (QP) regression model was selected as suggested by Barker and Sawyer (2010). The fitted regression model was used to determine EONR for each stage-site-year using the ratio history of fertilizer/grain cost the 5.9:1 (Barbieri et

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al., 2009a). The dEONR was calculated as the EONR minus applied N rates within each site-year (Hawkins et al., 2007). The relationship between sensor index and dEONR was evaluated by fitting a QP regression model using PROC NLIN, and the R^2 and 95% lower confidence limit (LCL) and upper confidence limit (UCL) regression parameters were calculated for each canopy index model (Barker and Sawyer, 2010). The LCL and UCL regression equations, compared to the fitted regression model across relative sensor index values, were used to determine the variability in sensor prescribed N rate for dEONR up to zero N. Since SPAD and NDVI readings were taken from multiple sites and years, PROC MIXED model was used to allow inclusion of site-year as a random variable in the regression model.

RESULTS AND DISCUSSION

Water availability did not limit wheat growth or grain yield because accumulated rainfall during July-December was greater than 380-400 mm at all sites and in all growing seasons, except 2011-G. Madariaga and 2011-Maipú, where registered rainfall reached 355 and 350 mm, respectively (Table 1). For those locations, water stress occurred during grain-filling period.

Observed pH values were among the range for soils typical of the region under long term cropping (Sainz Rozas et al., 2011) (Table 1). The SOM content varied between 49 and 65 g kg^{-1}, while Nan concentration and NO_3^--N availability ranged between 49 and 94 mg N kg^{-1}, and between 47 and 114 kg N ha^{-1}, respectively (Table 1). These variations in SOM, Nan and NO_3^--N could be attributed mainly to the effect of different time under cropping and different soil management practices, because surface soil texture was relatively similar in all sites (data not show). Sainz Rozas et al. (2011) reported SOM contents averaging 88 and 55 g kg^{-1} in the topsoil layer of pristine and continuous cropping soils, respectively. Anaerobically incubated N concentrations were within the values reported by several authors (Echeverría et al., 2000; Cozzoli et al., 2010; Reussi Calvo et al., 2013). Thus, the broad variation documented for the Nan parameter reflected diverse potential for N mineralization and response to fertilizer N applications.

In all experimental sites significant responses in yield by N fertilization were determined ($p<0.05$). Average crop yield was 5116, 5309, 8204, 7299, 6927, 6799, 8837, 6095, 7116 and 4105 kg ha^{-1} at 2010-Balcarce, 2010-Loberia, 2011-Loberia, 2011-Gral. Madariaga, 2011-Maipú, 2011-Miramar, 2011-Necochea, 2012-Miramar, 2012-Necochea and 2012-Lobería, respectively (Table 2). The maximum yield response to N

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(yield difference between treatment 300N or 200N and 0N) was 846, 1582, 1563, 854, 1324, 2629, 2969, 1842, 3559 y 1149 kg ha^{-1} for the same sites, respectively (Table 2). In 2011 and 2012 Necochea, the greatest N response can be partially explained by low $N0_3^-$-N and Nan levels observed at sowing, for one side, and for the high yield potential of the site (Table 1 and 2). Similar responses were determined by other authors in the SEB (Calviño et al., 2002; Barbieri et al., 2012; Reussi Calvo et al., 2013).

(Please, place Table 2 here)

The QP model had the highest nonlinear model fit statistics compared to other models and was statically significant all cases except for Z22 stage, in which, a QP model could not be significantly fit at the $P < 0.05$ level. The adjR^2 and RMSE QP model for different canopy index and growth stages are shown in Table 3. The use of relative values (rather than absolute) improved the fit between the sensor readings and dEONR, as it has been found by others (Schepers et al., 1992; Scharf et al., 2006; Hawkins et al., 2007). Therefore, relative indices were used for N algorithm development instead of direct index values (Table 3). For Z31 and Z39, the rSPAD had the highest goodness of fit statistics when compared to the adjR^2 and RMSE of rNDVI (Table 3). The rSPAD was more capable of measuring N stress resulted from the differences in the N availability. However, rNDVI showed highest goodness of fit statistics at Z24 stage (Table 3). Other authors have reported a significant association between rSPAD and yield during wheat stem elongation (Gandrup et al., 2004), and between rSPAD and dEONR at V_{15} and R_1 corn growth stage (Hawkins et al., 2007). A greater sensitivity of GS in the early stages of crop development has also been reported by others authors (Raun et al., 2001; Girma et al, 2006). This could be due to the fact that a deficiency of N reduces the number of leaves per unit area by a lower tillers production (Novoa and Loomis, 1981).

(Please, place Table 3 here)

Figure 2 shows the QP regression model for the relationship between rSPAD or rNDVI and dEONR. This relationship is similar to that found in corn by Hawkins et al. (2007) and Barker and Sawyer (2010). Table 4 shows canopy index QP regression equation parameters with the largest adjR^2 representing the relationship between the relative canopy index and dEONR for the SPAD and rNDVI. Both canopy indices have a similar value at zero dEONR (0.93-0.96), but different join point (200-220 for SPAD and -20- 100 for NDVI). The rate change of canopy index value per kg N ha^{-1} (model slope) was greater with the SPAD than

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NDVI (Table 4), reflecting a greater variation SPAD than NDVI across dEONR rates. Barker and Sawyer (2010) reported that indices related with canopy biomass have a reduced range of relative values across deficit dEONR than indices related to canopy chlorophyll.

(Please, place Figure 2 here)

In general terms, rSPAD and rNDVI values at plateaus are higher than at zero dEONR (at optimal N) (Table 4). In corn, other authors have reported similar value of rSPAD but slightly superiors to rNDVI (Hawkins et al. 2007; Barker and Sawyer, 2010). However the value at optimal N of rSPAD or rNDVI are close to 0.95, value that other researchers have found as a critical value or sufficiency index indicating plant N stress (Peterson et al., 1993). Moreover, a single quadratic-plateau regression model was fitted for rSPAD at Z31 and Z39 values to growth stage (Table 4). This indicates that N stress development and the relationship between rSPAD values and dEONR is the same for spring wheat in both stages, and therefore, one calibration can be used for determining in-season N stress and application rates during stem elongation the spring wheat.

(Please, place Table 4 here)

Moreover, a unique QP model to estimate with 58% confidence the dDOE was adjusted (RMSE = 0.04) for the Z31 and Z39 growth stages (Table 4 and Figure 3), suggesting there is a period of time during stem elongation vegetative growth, rather than one critical time, that provides a similar indication of plant N stress and determination of N application need from an rSPAD or rNDVI based N rate calibration. This time period is also during significant wheat N uptake, which is an important time for development and expression of N stress, and for making needed fertilizer N applications (Melaj et al., 2003). Moreover, Velasco et al. (2012) reported that significant responses to N added occurred up to Z39 stage. Finally, Barker and Sawyer (2010) suggested that if active canopy sensors will be utilized for assessing the in-season fertilizer N need, a measurement of the prediction accuracy for those models is needed. Figure 4 shows the prescribed N rate variability across deficit dEONR for rSPAD/rNDVI (Table 4). In contrast to these authors, our results indicate for spring wheat that based on the 95% confidence limits for the QP regression models, sensing slight N deficiencies (N algorithm prescribed in-season N application between 0–50 kg N ha^{-1}) produces less variability in prescribed N rate, and therefore not reduce the effectiveness of using active sensor based N stress detection for incremental or fine-tuning N application. Therefore, rSPAD/rNDVI can address spatial N

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variability and has the potential to improve field-scale N management when compared with other N management strategies (Kitchen et al., 2010).

(Please, place Figure 3 and 4 here)

In summary, the results of this paper indicated than active canopy sensors can measure N stress during the mid vegetative growth stages in spring wheat. With this information, the development of a single QP model to predict dEONR was adjusted during stem elongation. This model can provide N rate algorithms capable of directing variable in-season N rate application in SEB and other similar spring wheat production areas. Nitrogen application would be directed when the model index value is less than the value at zero dEONR. This information would be important to secure a high yield, more profitable fertilization levels and also, a high N use efficiency. This last point would allow reducing the environmental impact of N fertilization

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Figure 1. Shaded area shows the Southeastern Buenos Aires Province, Argentina and dots indicate the experimental sites.

Figure 2. Relative canopy index values as related to the differential from the economic optimum N rate (dEONR) for relative SPAD (rSPAD) and relative NDVI (rNDVI). Canopy indices and quadratic-plateau (QP) regression models were chosen from the highest goodness of fit statistics (adjR^2) in Table II. The LCL and UCL represent the 95% lower and upper confidence limits, respectively, of the QP regression models. Z24 (four tillers), Z31 (one visible node) and Z39 (visible flag leaf ligule) according to Zadoks et al. (1974).

Figure 3. Relative canopy index values as related to the differential from the economic optimum N rate (dEONR) for relative SPAD (rSPAD) and relative NDVI (rNDVI). The LCL and UCL represent the 95% lower and upper confidence limits, respectively, of the QP regression models. Z31 (one visible node) and Z39 (visible flag leaf ligule) according to Zadoks et al. (1974).

Figure 4. Sensor prescribed N rate variability across the range of differential economic optimum N rates (dEONR) less than zero for integrated model of rSPAD and rNDVI from Table 4.

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Table 1. Previous crop, years continuous cropping (AA), soil characteristics and rainfall (Pp) between July and December in the different experimental sites. SOM = soil organic matter, Nan= anaerobically incubated N.

Year/site	Preceding crop	AA (years)	SOM ($g\ kg^{-1}$)	pH	Nan ($mg\ kg^{-1}$)	$N0_3^-$-N ($kg\ ha^{-1}$)	Pp (mm)
			----------0-20 cm-------			-0-60cm-	
2010-Balcarce	Soybean	15	50	6.1	54	62	412
2010-Lobería	Sunflower	2	50	6.8	74	84	433
2011-Lobería	Soybean	2	50	6.3	94	47	450
2011-G. Madariaga	Soybean	6	65	5.8	77	74	355
2011-Maipú	Sunflower	6	56	5.9	73	78	350
2011-Miramar	Soybean	10	65	5.6	58	81	450
2011-Necochea	Soybean	15	58	6.4	62	69	508
2012-Miramar	Sunflower	10	61	5.7	62	114	398
2012-Necochea	Soybean	20	49	6.4	49	57	390
2012-Loberia	Soybean	5	53	5.8	74	64	570

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Table 2. Effect of nitrogen (N) fertilization on grain yield at different experimental sites.

Year/site	Grain yield (kg ha^{-1})						
	N rate (kg ha^{-1})						
	0	50	100	150	200	300	Average
2010-Balcarce	4700c	4737c	5419ab	5182b	5546a	5111b	5116
2010-Lobería	4370d	4999c	5028c	5659b	5847ab	5952a	5309
2011-Lobería	7435d	7666cd	8299bc	8530ab	8298bc	8998a	8204
2011-G. Madariaga	6783c	7202b	7412ab	7334ab	7426ab	7637a	7299
2011-Maipú	6205d	6400cd	6741c	7392ab	7529a	7297ab	6927
2011-Miramar	5174e	6002d	6593cd	7711ab	7803a	7513b	6799
2011-Necochea	6887d	7646c	9329b	9659ab	9643ab	9856a	8837
2012-Miramar	4805c	5861b	6228a	6427a	6647a	6604a	6095
2012-Necochea	5364d	5852d	6864c	7646b	8044b	8923a	7116
2012-Loberia	3252c	3846b	4398a	4400a	4401a	4333a	4105
Average	5497	6021	6643	7016	7082	7222	
Standard deviation	1308	1263	1477	1600	1536	1765	

In each row, means followed by the same letter are not significantly different according to least significant difference test (LSD) at 5% probability.

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Table 3. The goodness of fit statistics (adjR^2 and RMSE) for the quadratic- plateau (QP) regression models relating canopy indices and differential from the economic optimum N rate (dEONR) for the chlorophyll meter (SPAD) and Green Seeker Green 506 (NDVI) at different growth stage.

Canopy index	Growth stages†						
	Z22	Z24		Z31		Z39	
		adjR^2	RMSE	adjR^2	RMSE	adjR^2	RMSE
SPAD	ns	0.38	2.50	0.56	2.39	0.54	2.83
rSPAD	ns	0.53	0.04	0.67	0.03	0.68	0.03
NDVI	ns	0.18	0.10	0.31	0.05	0.53	0.05
rNDVI	ns	0.61	0.03	0.57	0.03	0.52	0.04

† z22 (two tillers), z24 (four tillers), z31 (one visible node) and z39 (visible flag leaf ligule) according to Zadoks et al. (1974).

‡ Adjusted R^2. Regression models were statistically significant at P = 0.001 level for each canopy index, except as noted when not significant at $P < 0.05$ (ns).

Relative indices calculated using the mean observed sensor value divided by the mean sensor value from the highest N rate for each site year are indicated with a prefix “r”.

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Table 4. Quadratic-plateau (QP) regression models and parameters for several relative canopy indices with the chlorophyll meter (SPAD) and Green Seeker Green 506 (NDVI). Regression models presented for the relative indices with the highest goodness of fit statistic (adjR^2) from Table 3 and integrated models.

Canopy index	QP regression model†	n	Join point‡	Canopy index at Plateau	Canopy index at Zero dEONR	Adj R^2§	P
			kg N ha^{-1}				
SPAD							
rSPAD-Z31	$y = 0.93 + 0.0005x - 0.00000087x^2$	54	200	1.00	0.93	0.67	<0.001
rSPAD-Z39	$y = 0.94 + 0.0004x - 0.00000046x^2$	48	220	1.00	0.94	0.68	<0.001
rSPAD-Z31-Z39	$y = 0.93 + 0.0004x - 0.0000004x^2$	102	210	1.00	0.93	0.66	<0.001
NDVI							
rNDVI-Z24	$y = 0.96 + 0.0001x - 0.0000027x^2$	60	-20	0.97	0.96	0.61	<0.001
rNDVI-Z31	$y = 0.95 + 0.0003x - 0.00000066x^2$	60	100	0.97	0.95	0.57	<0.001
SPAD and NDVI							
rSPAD and rNDVI-| Z31 and Z39	$y = 0.94 + 0.0004x - 0.0000005x^2$	210	196	1.00	0.94	0.58	<0.001

† For regression model, y is the canopy index value; x is the N rate differential from the EONR (dEONR), kg N ha^{-1}.
‡ Nitrogen rate where the quadratic equation joins the canopy index plateau value.
§ Adjusted R^2.

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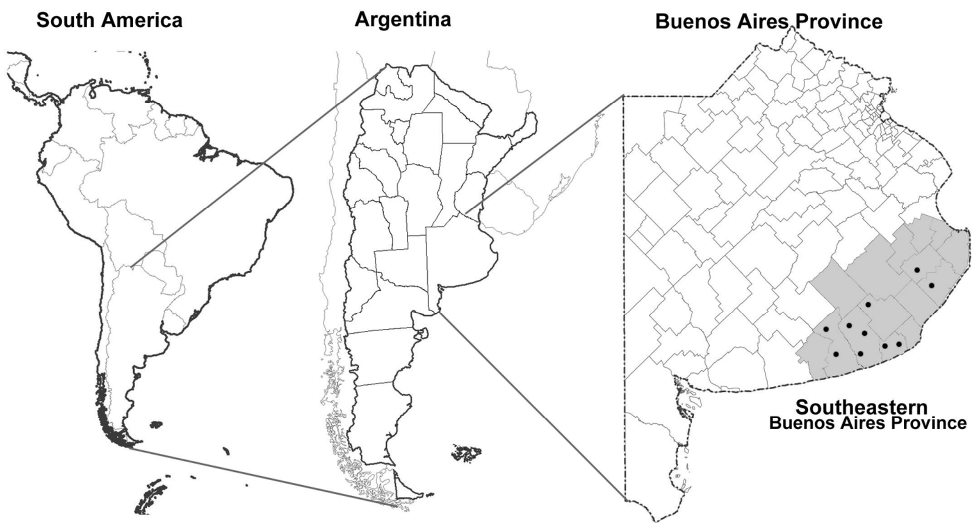

Shaded area shows the Southeastern Buenos Aires Province, Argentina and dots indicate the experimental sites.
187x98mm (299 x 299 DPI)

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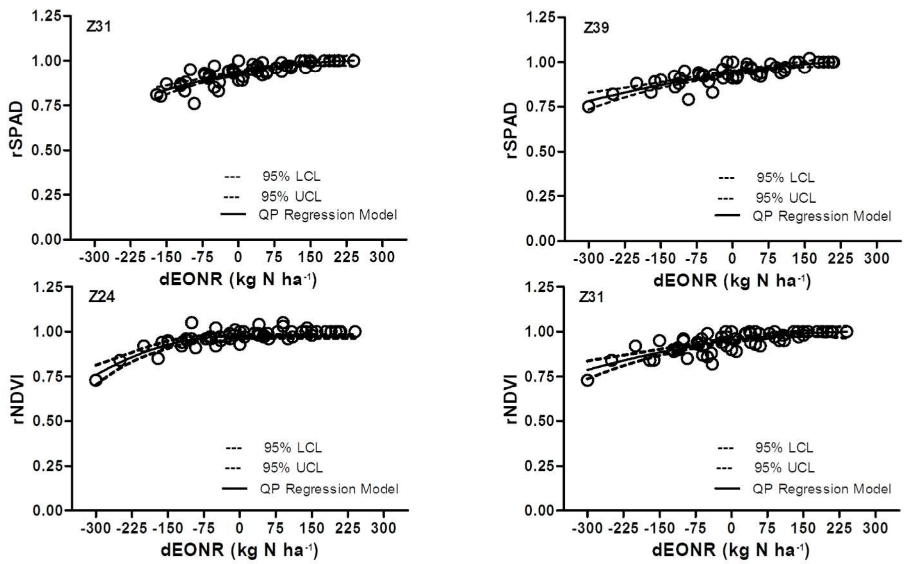

Relative canopy index values as related to the differential from the economic optimum N rate (dEONR) for relative SPAD (rSPAD) and relative NDVI (rNDVI). Canopy indices and quadratic-plateau (QP) regression models were chosen from the highest goodness of fit statistics (adjR2) in Table II. The LCL and UCL represent the 95% lower and upper confidence limits, respectively, of the QP regression models. Z24 (four tillers), Z31 (one visible node) and Z39 (visible flag leaf ligule) according to Zadoks et al. (1974).
183x114mm (300 x 300 DPI)

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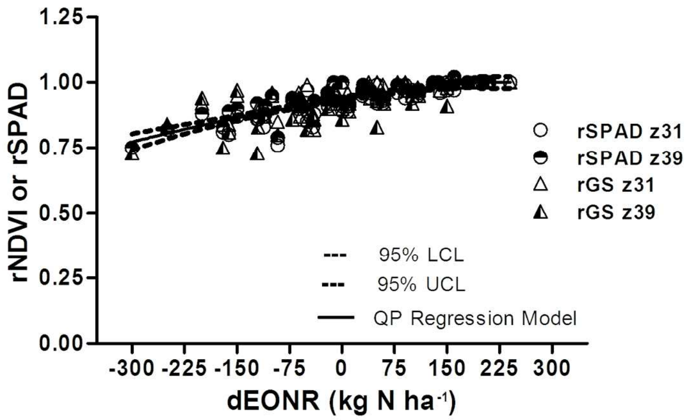

Relative canopy index values as related to the differential from the economic optimum N rate (dEONR) for relative SPAD (rSPAD) and relative NDVI (rNDVI). The LCL and UCL represent the 95% lower and upper confidence limits, respectively, of the QP regression models. Z31 (one visible node) and Z39 (visible flag leaf ligule) according to Zadoks et al. (1974).
165x119mm (300 x 300 DPI)

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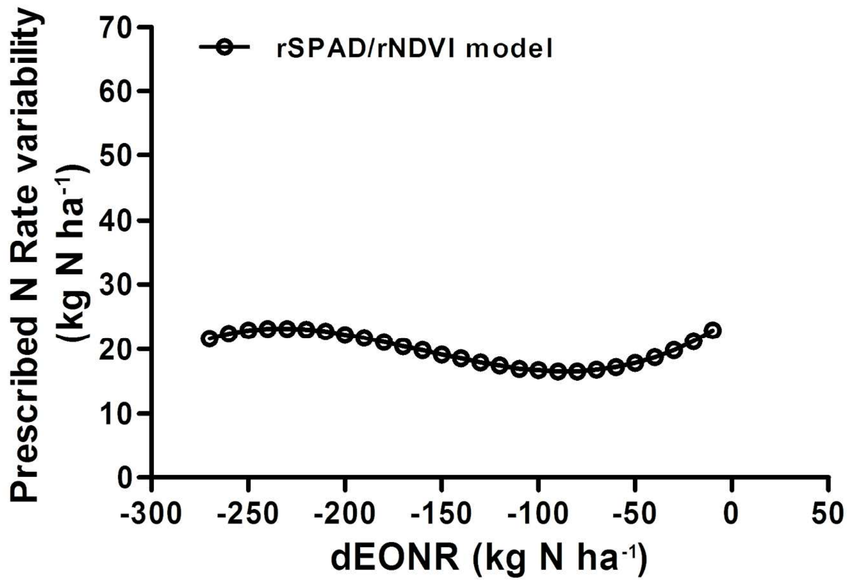

Sensor prescribed N rate variability across the range of differential economic optimum N rates (dEONR) less than zero for integrated model of rSPAD and rNDVI from Table 4.
182x124mm (300 x 300 DPI)