

Use of Normalized Difference Vegetation Index to Assess N Status and Predict Grain Yield in Rice

Telha H. Rehman,* Andre Froes Borja Reis, Nadeem Akbar, and Bruce A. Linquist

ABSTRACT

Fine tuning N recommendations requires an understanding of crop N status and yield potential early enough in the growing season when changes to N management can influence yields. Recent studies have demonstrated the ability of Normalized Difference Vegetation Index (NDVI) to assess crop N status and predict yield in wheat (*Triticum aestivum* L.) and maize (*Zea mays* L.); however, there has been relatively little such research on rice (*Oryza sativa* L.). The objectives of this study were to determine how well NDVI measured at the panicle initiation (PI) rice growth stage assesses crop N status and predicts final grain yield. Nitrogen response trials were established over a 4-yr period (10 site-years) at various locations throughout the Sacramento Valley rice growing region of California. Additionally, the relationship between NDVI and crop N status was characterized across 28 on-farm plots representing a range of environmental conditions and management practices. The NDVI at PI was best correlated with total N uptake (N_{UP} , $r^2 = 0.66$), followed by N concentration (N_{CONC} , $r^2 = 0.54$), and aboveground biomass (AGB, $r^2 = 0.51$). The utility of NDVI was greatest at lower values of crop N status, whereas at higher values, NDVI saturated. The NDVI at PI was positively correlated with final grain yield ($r^2 = 0.58$) indicating utility for developing in-season yield predictions. While NDVI is a potentially useful tool to improve N fertilizer management and develop in-season yield predictions in rice, alternative indices that do not saturate would likely provide a basis for a better tool.

Core Ideas

- The ability of NDVI to assess rice N status and predict final grain yield was evaluated across 38 sites and four years.
- NDVI at panicle initiation was most closely related to crop N uptake.
- At high values of crop N status NDVI had limited utility due to saturation.
- NDVI at panicle initiation was positively correlated ($r^2 = 0.58$) with final grain yield.
- NDVI of 0.66 at panicle initiation indicated sufficient crop N uptake to achieve average maximum grain yield.

DESPITE BEING the most studied nutrient worldwide, nitrogen (N) use efficiency in global rice (*Oryza sativa* L.) production is only about 30% (Ladha et al., 2005). In 2017, approximately 16 million Mg of N fertilizer was used for rice production worldwide (IFA, 2017), implying 11.2 million Mg of N was potentially lost to the environment. Nitrogen fertilizer losses from agricultural systems can have many adverse environmental and human health consequences. For example, nitrate leaching due to excessive N fertilization and improper water management can contaminate drinking water and lead to methemoglobinemia in infants (Di and Cameron, 2002; Harter et al., 2012). Significant amounts of greenhouse gases, such as nitrous oxide and methane, can be released from agricultural systems when N availability in the soil exceeds plant N requirements (Smith et al., 2007; Almaraz et al., 2018). Elevated N inputs to aquatic ecosystems from agricultural tailwater can result in hypoxic dead zones due to eutrophication and the proliferation of harmful algal blooms (Conley et al., 2009). Therefore, improved methods need to be designed and adopted that allow farmers to accurately assess crop N needs and make informed management decisions.

In California, the average seasonal N fertilizer requirement for rice is approximately 165 kg N ha⁻¹ (UC-ANR, 2018), which is most efficiently utilized when injected into the soil as aqua-ammonia before planting (Linquist et al., 2009). In recent years, an increasing number of California rice farmers have started applying additional N fertilizer as top-dress around panicle initiation (PI) growth stage. For the short duration varieties commonly grown in California, PI typically occurs around 45 to 50 d after sowing and is considered a critical stage for N management as all pre-plant N fertilizer has been taken up (LaHue et al., 2016), and N applied at growth stages later than PI is less efficiently utilized for grain yield (DeDatta, 1981; Linquist and Sengxua, 2003). The current recommendation at PI is for farmers to first assess crop N status and apply top-dress N only if the crop is deemed N deficient (Linquist et al., 2009). However, assessing crop N status in an accurate and timely manner remains a challenge in these systems, thus most

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Abbreviations: AGB, aboveground biomass; N_{CONC} , nitrogen concentration; NDVI, normalized difference vegetation index; N_{UP} , total nitrogen uptake; PI, panicle initiation.

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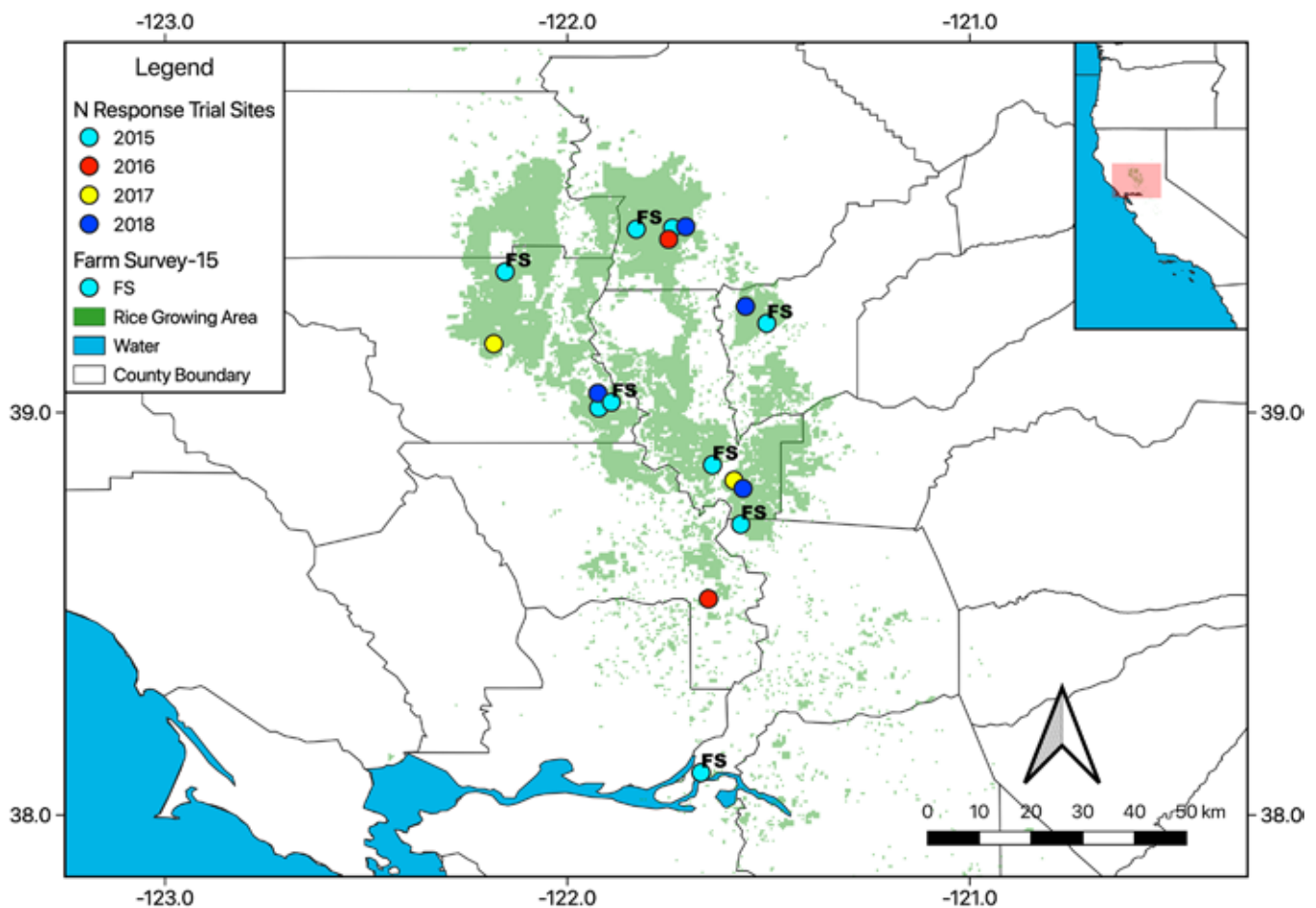


Fig. 1. Map of N response trial sites and Farm Survey-15 locations established during the 2015 to 2018 growing seasons throughout the Sacramento Valley rice growing area of California, USA.

top-dress N applications take place without evaluating crop N status; possibly resulting in inefficiencies due to over application.

Some methods are available to assess midseason plant N status but have not been widely adopted by California rice farmers due to their limitations. Plant tissue analysis provides the most direct measure; however, this technique is also time consuming and lab results are often received past the time when fertilizer decisions need to be made (Daughtry et al., 2000). Alternative technologies are available to expedite in-field N status assessment, such as the Leaf Color Chart (LCC) and the Soil Plant Analysis Development (SPAD) chlorophyll meter (Peng et al., 1996; Balasubramanian et al., 1999). The LCC estimates N content based on leaf greenness, while the SPAD chlorophyll meter measures the difference in transmittance between red and near infrared light passing through the leaf to estimate chlorophyll content (Alam et al., 2005; Uddling et al., 2007). Previous research has demonstrated the ability of these technologies to assess rice N status and promote sustainable N management (e.g., Yang et al., 2003; Islam et al., 2007; Singh et al., 2007). However, both the LCC and SPAD chlorophyll meter are inefficient as they only assess a single leaf at a time, thus requiring considerable time and effort to accurately assess a whole field (Daughtry et al., 2000; Saberioon et al., 2014; Xue et al., 2004).

More recently, remote sensing technology has been developed which utilizes canopy reflectance measurements to assess crop N status in a quick and nondestructive manner. Canopy reflectance data is collected remotely (via satellite, aircraft, or

proximal sensor), and interpreted through a vegetative index. The Normalized Difference Vegetation Index (NDVI) is the most widely adopted (McFarland and van Riper, 2013) and is sensitive to photosynthetic compounds, making it a potentially useful index to measure the productivity of vegetation in a defined area (Tucker, 1979; Tucker et al., 1985).

The ability of NDVI to assess crop N status and develop in-season yield predictions has been studied extensively in wheat (*Triticum aestivum*) and maize (*Zea mays*) production systems. Many have shown NDVI to effectively quantify plant N status across a variety of growth stages and sensor types (Reyniers and Vrindts, 2006; Li et al., 2008; Erdle et al., 2011; Li et al., 2014). Others found NDVI to be useful for developing in-season yield predictions by estimating biomass growth in wheat and maize (Raun et al., 2001; Teal et al., 2006; Inman et al., 2007). Adopting NDVI based N management in wheat and maize production systems has led to improved grain yield, N use efficiency, and net returns (Raun et al., 2002; Mullen et al., 2003; Raun et al., 2005; Tubaña et al., 2008). Comparatively, there have been relatively few such studies in rice. Some have tested the ability of NDVI to assess rice N status (Zhu et al., 2007; Gnyp et al., 2014; Yao et al., 2014; Lu et al., 2017) and few have used NDVI to develop in-season yield predictions (Harrell et al., 2011; Yao et al., 2012; Cao et al., 2016). However, most of these studies have focused their research on single sites, leaving at question the scalability of their findings to other sites representing different soils and management practices. Therefore, the

Table 1. Soil descriptions and selected properties of each N response trial site-year located throughout the Sacramento Valley, California.

Site-year	Soil series	Taxonomic classification	Texture			Organic carbon	Total nitrogen	pH
			Sand	Silt	Clay			
Arbuckle-15	Clear Lake Clay	Fine, smectitic, thermic Xeric Endoaquerts	10	33	57	2.25	0.19	6.2
RES-15	Esquon-Neerdobe	Fine, smectitic, thermic Xeric Epiaquerts	31	25	44	1.49	0.12	5.0
Davis-16	Sycamore Complex	Fine-silty, mixed, super active, nonacid, thermic Mollic Endoaquerts	13	37	50	2.22	0.20	7.0
RES-16	Esquon-Neerdobe	Fine, smectitic, thermic Xeric Epiaquerts	32	24	44	1.75	0.13	5.0
Nicolaus-17	Capay silty clay	Fine, smectitic, thermic Typic Haploxererts	19	36	45	1.44	0.13	5.5
Williams-17	Willows silty clay	Fine, smectitic, thermic Sodic Endoaquerts	21	39	40	1.79	0.16	5.0
Arbuckle-18	Clear Lake Clay	Fine, smectitic, thermic Xeric Endoaquerts	30	21	49	2.12	0.18	6.3
Biggs-18	Eastbiggs	Fine, mixed, active, thermic Abruptic Durixeralfs	50	30	20	1.52	0.12	4.9
Marysville-18	San Joaquin Loam	Fine, mixed, active, thermic Abruptic Durixeralfs	39	39	22	1.19	0.15	4.6
Nicolaus-18	Capay silty clay	Fine, smectitic, thermic Typic Haploxererts	22	36	42	1.75	0.16	4.8

objectives of this study were to determine how well NDVI at PI assesses rice N status and predicts final grain yield across a range of sites and years. Such research will provide the basis for using NDVI as an N management tool in rice.

MATERIALS AND METHODS

Nitrogen Response Trials

Site Description

Eight on-farm and two on-station N response trials were established during the 2015 to 2018 rice growing seasons at various locations (referred to by proximity to nearest town or station and study year) throughout the Sacramento Valley rice growing region of California (Fig. 1; Table 1). On-station sites were established at the California Rice Experiment Station (RES) near Biggs. The Sacramento Valley has a Mediterranean climate characterized by warm and dry conditions during the growing season (May to October). The average air temperature and precipitation during the growing season for the 4 yr of this study were 23.4°C and 7.04 mm, respectively, based on weather data collected from a centrally located California Irrigation Management Information Systems (CIMIS) weather station near Biggs (CIMIS, 2018). In California, most farmers use direct water-seeding to establish the rice crop. In this case, the fields are fertilized following seedbed preparation, flooded, and then soaked seed is broadcast onto the field using an airplane.

Soil samples were collected from the plow layer (approximately 0–15 cm) after tillage, but prior to fertilizer application. Soil taxonomic classification and selected chemical and physical properties for each site-year are provided in Table 1. Most study sites consisted of soils with high clay contents (40–57%), typical of rice soils in California. The only exceptions were soils at Biggs (20% clay) and Marysville (22% clay). Soil pH was measured using saturated paste (United States Salinity Laboratory Staff, 1954) and ranged from 4.6 to 7.0. Soil organic C and N were measured using an elemental analyzer interfaced to a continuous flow isotope ratio mass spectrometer (EA-IRMS) and ranged from 1.19 to 2.25%, and from 0.12 to 0.20%, respectively.

Experimental Design

Nitrogen response trials were arranged in a randomized complete block design with four replicates. In 2015 and 2016, plots measured 5 m by 6 m and in 2017 the plots measured 5 × 7.5 m. A

wide range of crop N status (i.e., biomass and N concentrations) was achieved by broadcasting pre-plant N fertilizer by hand at rates of 0, 75, 125, 175, and 225 kg N ha⁻¹ as urea (0.46 g N g⁻¹). In 2017, additional pre-plant rates of 45 and 275 kg N ha⁻¹ were included. In 2018, pre-plant N fertilizer was injected into the soil subsurface at approximately 7 to 10 cm depth as aqua-ammonia at rates of 0, 101, 135, 168, 202, and 235 kg N ha⁻¹. Plot width was determined by the swath width of the harrowing implement used to apply aqua-ammonia and ranged from 6.5 to 11.5 m. Plot length was 9.1 m at all sites and was taken from the central portion of a 21-m tractor pass to ensure uniform fertilizer application within the plots. Phosphorus (P) and potassium (K) were broadcast across all plots at a rate of 45 kg P₂O₅ ha⁻¹ as triple superphosphate (0.45 g P g⁻¹) and 50 kg K₂O ha⁻¹ as sulfate of potash (0.52 g K g⁻¹; 0.17 g S g⁻¹) to ensure these nutrients did not limit crop growth. Plots did not receive any additional fertilizer after pre-plant applications. Once all fertilizer was applied, fields were flooded and then aerially planted with pre-germinated seeds of medium grain rice variety M-206. Planting dates varied by site-year but were all within the normal timeframe for the Sacramento Valley (early to mid-May). Crop establishment and management followed common grower practice and was either managed by the grower (on-farm sites) or researchers (on-station sites).

Farm Survey

In addition to the N response trials, in 2015 a total of 28 on-farm plots (Farm Survey-15) were established to evaluate the relationship between NDVI and PI N status across a range of rice varieties, fertilizer management, soil types, microclimates, and crop establishment methods. Seven farms were selected (denoted by the nearest town or island) representing the major geographical regions of California where rice is grown (Fig. 1; Table 2). Within each farm, two to seven plots were established. Soil samples (0–15 cm) were collected from each plot and taxonomic classification and selected chemical and physical characteristics are reported in Table 2. All farms were within the Sacramento Valley, except Twitchell Island, which has peat and mineral soils, was dry-seeded (as opposed to water seeded), and has cooler temperatures due to its proximity to the Sacramento-San Joaquin Delta.

NDVI Measurements

A GreenSeeker handheld crop sensor (Trimble Inc., Sunnyvale, CA) was used to measure NDVI. The GreenSeeker is

Table 2. Soil description, selected properties, and rice variety grown at each Farm Survey-15 location.

Farm location (number of plots)	Rice variety	Soil series	Taxonomic classification	Texture			Organic carbon	Total nitrogen	pH
				Sand	Silt	Clay			
Arbuckle (2)	M-206	Clear Lake Clay	Fine, smectitic, thermic Xeric Endoaquerts	12–16	28–30	56–58	2.05–2.32	0.19	6.0–6.5
Biggs (3)	M-205	Lofgren-Blavo	Very-fine, smectitic, thermic Xeric Duraquerts	16–29	15–25	46–63	1.54–2.17	0.12–0.17	4.8–5.6
Marysville (4)	M-401	Kimball loam	Fine, mixed, active, thermic Mollic Palexeralfs	35–47	29–37	24–29	1.01–1.64	0.10–0.14	4.9–5.1
Maxwell (3)	M-206	Willows silty clay	Fine, smectitic, thermic Sodic Endoaquerts	19–31	32–40	37–43	2.52–2.71	0.22–0.23	5.1–5.4
Robbins (4)	Koshi	Clear Lake silt loam	Fine, smectitic, thermic Xeric Endoaquerts	49–83	8–34	9–18	0.40–0.96	0.04–0.08	4.9–5.5
Sacramento (5)	M-104, FRC-22	Clear Lake Clay/ Yuvas Loam	Fine, smectitic, thermic Xeric Endoaquerts/Fine, mixed, active, thermic Abruptic Durixeralfs	21–33	23–38	36–49	1.64–1.97	0.14–0.17	5.0–5.8
Twitchell Island (7)	M-206	Rindge mucky silt loam	Euic, thermic Typic Haplosaprists	15–87	5–39	8–56	2.72–26.14	0.05–1.34	5.1–5.8

an active sensor which measures canopy reflectance (ρ) at specific wavelengths in the red (670 ± 10 nm) and near infrared (780 ± 10 nm) regions of the electromagnetic spectrum and calculates NDVI as $(\rho_{780\text{ nm}} - \rho_{670\text{ nm}}) / (\rho_{780\text{ nm}} + \rho_{670\text{ nm}})$. Measurements were taken at PI, which marks the physiological shift from vegetative to reproductive plant growth (Counce et al., 2000). For the short duration varieties, which were used in most sites in this study, PI occurs approximately 45 to 50 d after sowing and is visually determined by a dark green ring just below the initiating panicle, occurring 5 to 7 d before panicle differentiation (when the panicle becomes visible) (De Datta, 1981). Panicle initiation was visually confirmed in the field prior to measuring NDVI using the method outlined by Dunn et al. (2014). Measurements were taken by holding the GreenSeeker in the nadir position and scanning it over the biomass sampling area at a constant height of 1.0 m above the crop canopy. For each plot, the final NDVI value represented the average of three to four NDVI readings. Canopy closure was achieved by PI in all plots that received N fertilizer, thus the effect of background water or soil on NDVI measurements was considered negligible. For the 0 N plots, some influence of background water was present and was accounted for by taking the average of multiple NDVI readings.

Two GreenSeekers were used to measure NDVI (GreenSeeker 1 in 2015 and GreenSeeker 2 from 2016 to 2018). Consistent differences between the two devices were detected by plotting side by side NDVI measurements ($n = 105$) (Supplemental Fig. S1). Differences were normalized by adjusting NDVI values based on the resulting fitted linear regression equation.

This variability across GreenSeekers is a concern and needs to be addressed when using the device in the field. Often, when using NDVI to inform N fertilizer management, a response index is developed where the NDVI of an N-non-limiting plot and the field test area are measured and the ratio of the two provides the response index (Mullen et al., 2003). In such cases, the variability between GreenSeeker units in terms of direct NDVI measurements would be less a concern.

Biomass Sampling

Immediately following NDVI measurements, all rice plants within a 0.5-m² quadrat were pulled from each plot. After

removing roots, the aboveground biomass was oven dried at 60°C to constant weight, after which the samples were ground in a Wiley mill and then ball-milled. Plant material from each plot was analyzed for total N using EA-IRMS. Two plant samples were collected from each of the 28 Farm Survey-15 plots to calculate an average for each plot. One plant sample was collected per plot for the N response trial sites. From these samples, we quantified the following parameters of crop N status: aboveground biomass (AGB, kg ha⁻¹), N concentration (N_{CONC} , g N kg⁻¹), and total N uptake (N_{UP} , kg N ha⁻¹, = $\text{AGB} \times N_{\text{CONC}}$).

Grain Yield

Grain yield (kg ha⁻¹) was obtained by harvesting mature plants from a 1.0 m² quadrat in each plot (grain yield was not obtained for the Farm Survey-15 plots). Grains were removed from panicles, cleaned using a seed blower, dried to constant moisture at 60°C, and weighed. Final yields are reported at 14% moisture.

Statistical Analysis

Construction of plots, development of regression models, and the analysis thereof was performed using the statistical program R (version 3.5.2; R Core Team, 2019). The package ‘ggplot2’ (Wickham, 2009) was used to visualize the data and construct plots. For the purpose of analysis, data from the Farm Survey-15 plots were combined into a single site-year. The relationship between NDVI and each N status parameter was described using a quadratic linear regression model. The horizontal asymptote for each model was determined as the y-value at the vertex, which was calculated from the resulting model coefficients. Quadratic models were selected over complex higher order models as both model types explained a similar amount of variability in the data and the quadratic models allowed for direct comparisons of results with previous studies.

The relationship between N_{UP} and grain yield was described by a segmented linear regression model from the package ‘segmented’ (Muggeo, 2017). The segmented model identifies breakpoints in the data (i.e., significant changes in the slope parameter) and describes the data before and after the breakpoint using separate linear segments. The relationship between NDVI and grain yield was described by a simple linear regression model.

Table 3. Descriptive statistics (sample number, minimum, maximum, and mean) of rice N status parameters and Normalized Difference Vegetation Index (NDVI) measured at panicle initiation growth stage.

Site-year	N	Aboveground biomass		N concentration		Total N uptake		NDVI	
		Min–Max	Mean	Min–Max	Mean	Min–Max	Mean	Min–Max	Mean
		kg ha ⁻¹		g N kg ⁻¹		kg N ha ⁻¹			
Arbuckle-15	20	3400–8540	6334	13.6–30.5	21.4	48.9–255.8	141.4	0.49–0.78	0.71
Farm Survey-15	28	1260–7260	5090	10.9–33.6	21.9	13.8–196.4	114.9	0.18–0.82	0.65
RES-15	20	3520–6540	5084	11.9–37.3	23.8	41.7–230.5	126.5	0.53–0.80	0.73
Davis-16	20	1332–3714	2609	14.6–31.7	21.5	20.3–114.7	58.9	0.56–0.72	0.67
RES-16	20	1466–4960	3428	18.5–38.8	28.6	30.9–192.6	103.2	0.36–0.75	0.64
Nicolaus-17	28	3970–7426	5559	15.5–36.1	25.7	61.7–240.2	147.7	0.49–0.80	0.68
Williams-17	28	2740–7270	5471	12.3–30.6	22.1	33.8–194.3	124.6	0.36–0.82	0.71
Arbuckle-18	24	730–8006	3397	12.1–30.2	21.4	9.7–160.6	76.5	0.15–0.75	0.61
Biggs-18	23	1962–6812	5019	10.4–32.9	21.5	20.4–193.4	113.6	0.36–0.79	0.69
Marysville-18	24	2384–5472	4604	16.1–37.0	29.9	38.3–202.4	142.0	0.45–0.75	0.66
Nicolaus-18	24	3242–7282	6069	13.1–30.7	23.3	46.0–223.5	146.0	0.58–0.77	0.72
All	289	730–8540	4840	10.4–38.8	23.7	9.7–255.8	118.9	0.15–0.82	0.68

Graphical and numerical summaries were examined to ensure the assumptions of linear regression were satisfied for all regression models. Model goodness of fit was assessed by comparing adjusted coefficient of determination (r^2) and root mean squared error (RMSE), calculated as:

$$RMSE = \sqrt{\frac{1}{n} \sum_{i=1}^n (y_i - \hat{y}_i)^2}$$

RESULTS

Nitrogen Response Trials

Considerable variability in crop AGB, N_{CONC} and N_{UP} was present at PI both within and across N trial sites. As expected, the within-site variability was due to the different N rates with crop AGB, N_{CONC} and N_{UP} all increasing with increasing N rate (Table 3). Across site-years, mean AGB ranged from 2609 kg ha⁻¹ (Davis-16) to 6334 kg ha⁻¹ (Arbuckle-15); N_{CONC} ranged from 21.4 g N kg⁻¹ (Arbuckle-15, Arbuckle-18) to 29.9 g N kg⁻¹ (Marysville-18); and N_{UP} ranged from 58.9 kg N ha⁻¹ (Davis-16) to 147.7 kg N ha⁻¹ (Nicolaus-17).

At any given N trial site, NDVI increased with increasing N rate to a point then leveled off (i.e., saturated). Therefore, the lowest NDVI values tend to represent the 0 N rate, while the

Table 4. Descriptive statistics (sample number, minimum, maximum, and mean) of final grain yield at the N response trial site-years (yields were not obtained for Farm Survey-15).

Site-year	N	Grain yield†	
		Min–Max	Mean
		kg ha ⁻¹	
Arbuckle-15	20	6,469–14,529	12,072
RES-15	20	5,235–14,140	11,753
Davis-16	20	6,664–13,969	10,599
RES-16	20	6,653–14,675	11,246
Nicolaus-17	28	10,345–13,375	12,005
Williams-17	28	6,096–12,829	10,159
Arbuckle-18	24	2,948–13,648	9,980
Biggs-18	23	6,767–13,069	11,468
Marysville-18	24	8,046–12,246	11,000
Nicolaus-18	24	8,961–14,391	12,793
All	231	2,948–14,675	11,291

† Adjusted to 14% moisture.

highest NDVI value represented the higher N rates. Minimum NDVI varied considerably among the site-years, ranging from 0.15 (Arbuckle-18) to 0.58 (Nicolaus-18) whereas maximum NDVI only ranged from 0.72 (Davis-16) to 0.82 (Williams-17) (Table 3).

As expected, the lowest grain yields in the N trials were in the 0 N treatments, with grain yield increasing at most sites to a maximum and then leveling off or decreasing at higher N rates. Minimum site-year grain yield ranged from 2948 kg ha⁻¹ (Arbuckle-18) to 10,345 kg ha⁻¹ (Nicolaus-17) (Table 4). Despite different maximum AGB, N_{CONC} , and N_{UP} at PI across site-years, maximum yields were relatively similar and ranged from 12,246 kg ha⁻¹ (Marysville-18) to 14,675 kg ha⁻¹ (RES-16). Overall, there was no segregation in crop AGB, N_{CONC} , N_{UP} , or grain yield between site-years based on different sources of pre-plant N fertilizer (i.e., urea and aqua-ammonia).

Farm Survey

Crop N status data taken at PI from the Farm Survey-15 plots varied considerably, as may be expected, given the large number of farms within Farm Survey-15 and the variability among them. The range of AGB (1260–7260 kg ha⁻¹), N_{CONC} (10.9–33.6 g N kg⁻¹), and N_{UP} (13.8–196.4 kg N ha⁻¹) was considerably larger across Farm Survey-15 plots relative to N trial site-years (Table 3). Variability in crop N status was also reflected by the wide range of NDVI (0.18–0.82). Grain yield was not obtained for Farm Survey-15.

Panicle Initiation N Status and NDVI

An increase in PI N status led to a corresponding increase in NDVI, until a threshold was achieved, after which NDVI values leveled off (Fig. 2). The NDVI saturated within a narrow range (0.76 to 0.78), when AGB, N_{CONC} , and N_{UP} exceeded 7597 kg ha⁻¹, 29.9 g N kg⁻¹, and 185 kg N ha⁻¹, respectively. Overall, the nature of the relationship between each N status parameter and NDVI was similar across the N response trials and Farm Survey-15. Of the three N status parameters, N_{UP} explained the largest amount of variation in NDVI ($r^2 = 0.66$), followed by N_{CONC} ($r^2 = 0.54$) and AGB ($r^2 = 0.51$).

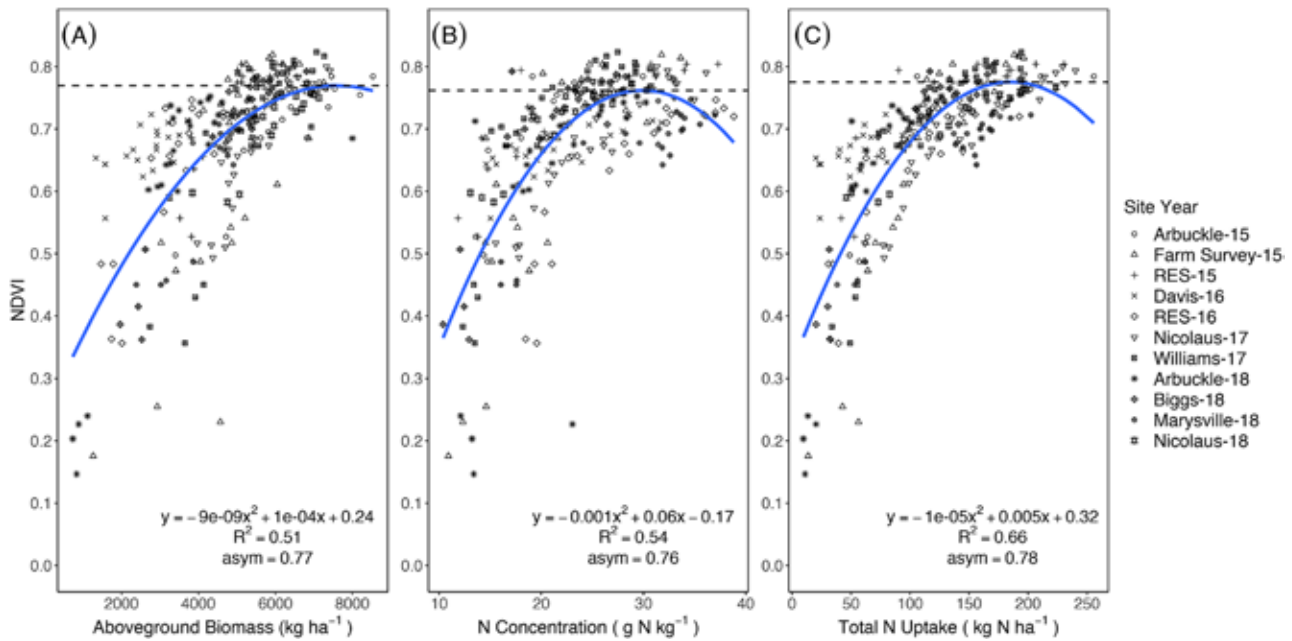


Fig. 2. Relationship between rice (A) aboveground biomass, (B) N concentration, and (C) total N uptake at panicle initiation rice growth stage and Normalized Difference Vegetation Index (NDVI) as described by quadratic linear regression models. The horizontal asymptote (asym) represents the NDVI value at which the relationship saturates. Data were collected during the 2015 to 2018 growing season from ten N response trial sites and 28 on-farm plots (Farm Survey-15) throughout the Sacramento Valley rice growing region of California.

Panicle Initiation Total N Uptake, NDVI, and Final Grain Yield

Based on the segmented model, PI N_{UP} explained a large portion of the variation in final grain yield ($r^2 = 0.63$; RMSE = 1321 kg ha⁻¹) (Fig. 3a). The segmented model estimated a breakpoint at 93.9 kg N ha⁻¹ (95% confidence interval: 85.1 to 102.9 kg N ha⁻¹) (data not shown), indicating that an increase in crop PI N_{UP} beyond this value did not result in a significant increase in average final grain yield. The slope before the breakpoint was 81 kg kg⁻¹ N, and after the breakpoint was not statistically different than a zero slope. The breakpoint of 93.9 kg N ha⁻¹ corresponded to an average maximum grain yield of 12,314 kg ha⁻¹. Based on the simple linear regression model, NDVI at PI was positively correlated with final grain yield ($r^2 = 0.58$; RMSE = 1415 kg ha⁻¹) (Fig. 3b).

DISCUSSION NDVI Saturation

Quadratic linear regression models were developed to describe the relationship between NDVI and crop N status. In each case, as crop N status increased, so did NDVI, until a horizontal asymptote was reached and additional increases in crop N status led to minimal change in NDVI (Fig. 2). This saturation of two-band indices such as NDVI is a well-known phenomenon (Asrar et al., 1984; Hatfield et al., 1985; Thenkabail et al., 2000; Cao et al., 2013; Gu et al., 2013). NDVI saturation is a result of the crop reaching 100% canopy cover, but AGB and leaf area index continuing to increase (Gitelson, 2003). Once the canopy reaches 100% cover, near infrared reflectance continues to rise, but red reflectance only exhibits a modest decrease, resulting in only slight changes in the ratio (i.e., the denominator will have a much greater impact on the ratio than the numerator) (Thenkabail et al., 2000). In our study, NDVI saturated within a narrow range (0.76 to 0.78), when AGB,

N_{CONC} , and N_{UP} exceeded 7597 kg ha⁻¹, 29.9 g N kg⁻¹, and 185 kg N ha⁻¹, respectively (Fig. 2). Our result is similar to the findings of Yao et al. (2014) who reported the relationship between NDVI and AGB and N_{UP} to saturate at about 0.80 and 0.78, respectively. Gnyp et al. (2014) reported the relationship between AGB and NDVI to saturate at approximately 0.90, which is higher than our study and may be because they simulated GreenSeeker NDVI from passive hyperspectral data, while we have used actual GreenSeeker measurements.

Recent studies suggest indices which incorporate a red-edge band (690 to 730 nm) may improve rice N status assessment by overcoming the saturation problem (Wang et al., 2012; Cao et al., 2013; Dunn et al., 2016). Cao et al. (2013) found several red-edge based indices to explain a large portion of rice N_{UP} variability when described by linear regression models. Wang et al. (2012) developed a red-edge based three band index which estimated N_{CONC} with high accuracy while reducing saturation. Dunn et al. (2016) confirmed the strong correlation of red-edge bands with rice N_{UP} based on their analysis of fine-resolution hyperspectral data. Given the saturation of NDVI, and strong linear relationships observed between red-edge based indices and rice N status, further research is warranted to investigate the potential improvement of red-edge based indices over NDVI to assess rice N status.

Assessing Panicle Initiation N Status with NDVI

Of the three N status parameters, N_{UP} explained the largest amount of variation in NDVI ($r^2 = 0.66$), followed by N_{CONC} ($r^2 = 0.54$) and AGB ($r^2 = 0.51$) (Fig. 2). The relationship between NDVI and crop N status was similar across the N trial site-years and Farm Survey-15, indicating NDVI assessed crop N status consistently across the wide range of environmental conditions and management practices included in this study. Importantly, within the observations in this study, AGB at

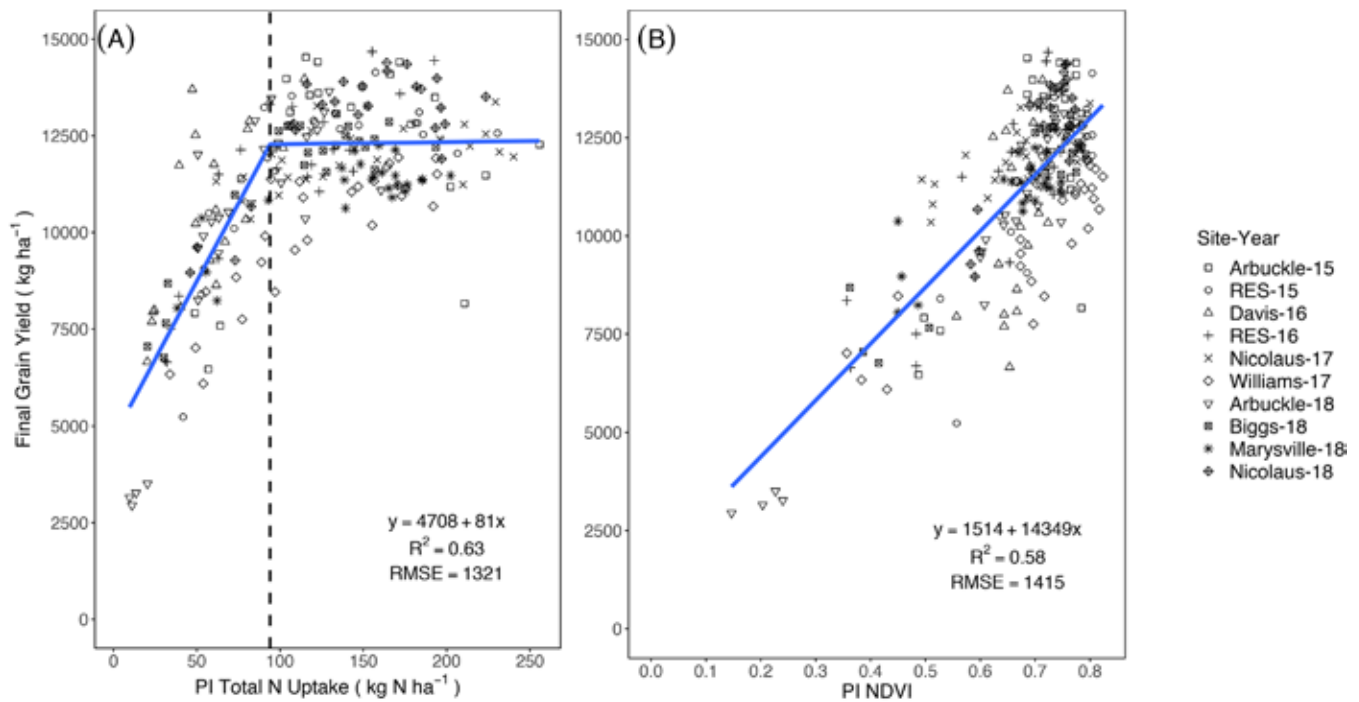


Fig. 3. (A) Relationship between rice total N uptake at panicle initiation (PI) growth stage and final grain yield as described by a segmented model. The vertical dashed line at 93.9 kg N ha⁻¹ represents the average minimum amount of crop N uptake required by PI to achieve average maximum grain yield. (B) Relationship between Normalized Difference Vegetation Index (NDVI) at PI and final grain yield as described by a simple linear regression model. Data were collected during 2015 to 2018 from ten N response trial sites throughout the Sacramento Valley rice growing area of California.

NDVI saturation was closer to the maximum observed AGB, whereas NDVI saturated earlier for N_{CONC} and N_{UP} (Fig. 2). This suggests at PI, NDVI saturation may pose less of a limitation when assessing AGB as it would with N_{CONC} or N_{UP} . That said, the relationship between NDVI and AGB is still poorer than for N_{CONC} or N_{UP} .

The relationship between NDVI and crop N status observed in this study is similar in strength to what others have found in wheat and maize (Reyniers and Vrindts, 2006; Li et al., 2008; Erdle et al., 2011; Li et al., 2014). To our knowledge, only one other study has examined the relationship between NDVI and AGB, N_{CONC} , and N_{UP} in rice. In that study, Yao et al. (2014) reported the strongest correlation between NDVI and AGB ($r^2 = 0.76$), followed by N_{UP} ($r^2 = 0.70$), and N_{CONC} ($r^2 = 0.38$). This is in contrast to our study where NDVI predicted N_{UP} and N_{CONC} better than AGB. We are not sure why this difference between studies, but it may be because Yao et al. (2014) conducted all their research at a single location, thus resulting in less variation of AGB during the course of their study. Others have looked at the relationship between NDVI and N_{CONC} and have reported both strong ($r^2 = 0.81$) and weak ($r^2 = 0.08$) correlations (Zhu et al., 2007; Lu et al., 2017), which may be due to differences in rice varieties or the growth stage when data was collected. In other studies, Gnyep et al. (2014) examined the relationship between NDVI and AGB and reported the same correlation ($r^2 = 0.51$) as our study; while Li et al. (2018) examined the relationship between leaf N_{UP} and NDVI and found a similar correlation ($r^2 = 0.70$) to our study with plant N_{UP} .

The strength of our study relative to most of the other studies mentioned above is that it considered multiple N status parameters

over a large range of sites and years. The strong correlation observed between NDVI and rice N status in this study suggests that the GreenSeeker could be a scalable tool to assess N status. However, as previously discussed, NDVI saturation limits its utility to lower values of crop N status, suggesting alternative indices that do not saturate could potentially improve N status assessment.

Predicting Final Grain Yield at PI with N_{UP} and NDVI

The utility of NDVI to develop regional scale rice yield predictions has received considerable attention (e.g., Huang et al., 2013; Son et al., 2014; Pagani et al., 2019), while fewer studies have focused on the farm scale. The ability to estimate rice yield early in the season is of interest to farmers and private companies for a number of reasons, including refining N fertilizer recommendations, planning harvest, forecasting milling and storage needs, and defining marketing strategies.

We observed a positive correlation ($r^2 = 0.63$) between PI N_{UP} and final grain yield (Fig. 3a). Yields increased strongly with increasing N_{UP} until they reached a plateau at a breakpoint of 93.9 kg N ha⁻¹ (Fig. 3a). This breakpoint represents the average minimum amount of crop PI N_{UP} required to achieve average maximum grain yield. Across sites the actual N_{UP} value varied as indicated by the 95% confidence interval ranging from 85.1 to 102.9 kg N ha⁻¹ (data not shown) and final grain yield at the breakpoint also varied considerably (Fig. 3a). Part of this variability may be explained by differences in soil indigenous N supply after PI. For example, achieving the average maximum grain yield at the breakpoint (12,314 kg ha⁻¹) requires a total seasonal N_{UP} of approximately 215 kg N ha⁻¹ (assuming N concentrations in rice grain and straw of 1.10 and 0.65%,

respectively; Dobermann and Fairhurst, 2000), indicating that an additional 121 kg N ha⁻¹ is required after PI. Given that pre-plant N fertilizer is completely taken up by PI (LaHue et al., 2016), and additional N fertilizer was not applied, this requirement must have been satisfied by soil indigenous N. Previous studies have shown that indigenous N supply from rice soils can vary significantly across sites and over time and is closely linked with soil properties such as organic carbon (Cassman et al., 1998; Espe et al., 2015). In theory, the breakpoint of 93.9 kg N ha⁻¹ N_{UP} and corresponding NDVI could potentially serve as a target for farmers when assessing midseason crop N requirements. However, accounting for site-specific differences in soil N supply may be needed to refine this target and further research could explore this. In this study, N_{UP} of 93.9 kg N ha⁻¹ corresponds to a NDVI value for 0.66 (derived from Fig. 2c), and importantly, this NDVI value is below the saturation value.

Given the relationship between PI N_{UP} and final grain yield (Fig. 3a) and PI N_{UP} and NDVI (Fig. 2c), the positive correlation between NDVI at PI and final grain yield ($r^2 = 0.58$) was expected (Fig. 3b). This is similar to Cao et al. (2016), who found a comparable correlation ($r^2 = 0.63$) in their experiments at a single location. Others (e.g., Harrell et al., 2011; Yao et al., 2012) have reported a poorer relationship between NDVI and grain yield with r^2 values ranging from 0.36 to 0.44.

Importantly, for short duration varieties grown in California, PI usually occurs about 45 to 50 d after seeding; thus, only one-third of the entire growing season. Grain yield can be altered in a number of ways after PI due to many abiotic and biotic factors. For example, in California and elsewhere, cold nighttime temperatures at meiosis (between PI and heading) causes floret sterility and reduced grain yields (Board et al., 1980; Espe et al., 2016). High temperatures at flowering can result in yield losses in many rice growing areas, including California (Espe et al., 2016; Fahad et al., 2018). Additionally, differences in soil N supply late in the season can affect yields as discussed above. Biotic factors such as insects and diseases can all negatively affect yields after PI (Sesma and Osbourne, 2004; Brooks et al., 2009; Hasanuzzaman et al., 2018). The greater the variability in these stresses across sites or years, the poorer the relationship will be between NDVI at PI and final grain yield. Given this, one should not expect the relationship between final grain yield and any plant measurement taken at PI to be very high. However, if those relationships were developed under optimal conditions where post PI stresses did not limit grain yield, then such measurements may provide a good estimate of yield potential. Although, the incidence of these stresses was not measured directly in this study, the fact that maximum grain yields were similar across all site-years suggests that post PI stresses did not have a significant impact on yields, thus providing optimal conditions to predict final grain yield at PI using NDVI.

CONCLUSION

The significant correlation between GreenSeeker NDVI and crop N status suggests that it may be developed into a useful tool to guide midseason N management decisions. However, NDVI saturated at high values of crop N status, suggesting further research in alternative indices (e.g., red-edge based NDVI) is warranted and could potentially improve estimates of midseason N status. Interestingly, in this study we identified an N_{UP}

value at PI (93.9 kg N ha⁻¹) at which average maximum grain yield was achieved. This value could serve as a midseason target in similar systems and may identify when further N applications are needed. The NDVI corresponding to this N_{UP} value is 0.66 which, importantly, is below the saturation point. Finally, as technology advances, future research focusing on large scale production systems will likely shift away from handheld proximal sensors, like the GreenSeeker used in this study, in favor of sensors that can be mounted to drones or satellites.

SUPPLEMENTAL MATERIAL

Supplemental material is available with the online version of this article.

DATA AVAILABILITY

GreenSeeker NDVI values, crop N uptake data, and rice yield data from sites established across the Sacramento Valley rice growing area of California during the 2015 to 2018 growing seasons. This material is available for downloading at dryad (<https://doi:10.5061/dryad.k0r39vv>).

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