

ARTICLE

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Which active optical sensor vegetation index is best for nitrogen assessment in irrigated cotton?

K. F. Bronson¹  | M. M. Conley¹ | A. N. French¹ | D. J. Hunsaker¹ | K. R. Thorp¹ | E. M. Barnes²

¹USDA-ARS, U.S. Arid Land Agricultural Research Center, 21881 N. Cardon Ln, Maricopa, AZ 85138, USA

²Cotton Incorporated, 6399 Weston Parkway, Cary, NC 27513, USA

Correspondence

Kevin Bronson, USDA-ARS, U.S. Arid Land Agricultural Research Center, 21881 N. Cardon Ln, Maricopa, AZ 85138, USA
Email: kevin.bronson@usda.gov

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Cotton Incorporated

Abstract

Use of active optical sensors (AOS) in nitrogen (N) management of row crops continues to grow. Since the first studies in the mid-1990s, several commercial AOS are now available. Typically, canopy reflectance in red and near infrared (NIR) bands are used to calculate the normalized difference vegetation index (NDVI). More recently, commercially available AOS include a third, red-edge band that allows the calculation of additional vegetation indices (VIs). We present two studies of five site-years of N management studies in Maricopa, AZ, on a Casa Grande sandy loam with cotton (*Gossypium hirsutum* L.). The 2014–2015 study was conducted under an overhead sprinkler irrigation system (OSI), and the 2016–2018 study was in subsurface drip irrigation (SDI). The study objective was to compare the ability of 12 different VIs to detect N deficiency among N treatments from shortly after emergence to mid-bloom. In the OSI study, which showed delayed, small N treatment effects, NDRE and the chlorophyll index using amber (CIRE) detected N deficiency in zero-N and in reduced N-reflectance-based plots 7–23 d before other VIs did. With SDI, the choice of VI was less critical as several VIs could distinguish N deficiency in zero-N and in reflectance plots. The commonly used NDVI red (NDVIR) only detected N deficiency in reflectance plots in one of five site-years. In conclusion, we recommend the use of AOS with NIR and red-edge bands and the calculation of NDRE or CIRE to guide AOS-based in-season N management of irrigated cotton.

Abbreviations: AOS, active optical sensor; CCCI, canopy chlorophyll content index; CIA, chlorophyll index using amber; CIRE, chlorophyll index using red edge; DATT, Datt, 1999; DGPS, differential geopositioning system; LAI, leaf area index; MTCI, Meris terrestrial chlorophyll index; NDARE, normalized difference vegetation index-amber-red edge; NDRE, normalized difference red edge index; NDRRE, normalized difference vegetation index-red-red edge; NDVIA, normalized difference vegetation index amber; NDVIG, normalized difference vegetation index green; NDVIR, normalized difference vegetation index red; NIR, near infrared; OSI, overhead sprinkler irrigation; PRI, physiological reflectance index; SDI, subsurface drip irrigation; VI, vegetation index.

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1 | INTRODUCTION

Interest in canopy reflectance using active optical sensors (AOS) continues to grow for the management of N fertilizer in field crops such as cotton (Arnall, Abit, Taylor, & Raun, 2016; Bronson, Malapati, Scharf, & Nichols, 2011; Chua et al., 2003; Oliveira et al., 2012; Raper, Varco, & Hubbard, 2013; Stamatiadis et al., 2019). The most common AOS technology measure reflectance in visible (450–670 nm, but typically red, i.e., 650–670 nm) and NIR (780–870 nm) wavebands, and vegetation indices like the normalized difference vegetation index (NDVI) are calculated (Tucker, 1979) from

the reflectance values. However, the NDVI can saturate at medium to high leaf area index as the crop canopies close (Aparicio, Villegas, Casadesus, Araus, & Royo, 2000; Erdle, Mistele, & Schmidhalter, 2011; Li et al., 2008). Therefore, there has been interest in testing VIs that remain sensitive to N at leaf area index > 2. Many studies have reported that NDVI calculated using green or amber reflectance (NDVIA) is more sensitive to N than the traditional NDVI with red as the visible waveband (Bronson et al., 2017a; Gitelson, Kaufman, & Merzlyak, 1996; Holland, Schepers, Shanahan, & Host, 2004; Shiratsuchi et al., 2011; Solari, Shanahan, Ferguson, Schepers, & Gitelson, 2008). Other studies have indicated improved sensitivity to N with VIs which include a red-edge waveband of 710–740 nm, which is approximately halfway between red and NIR (Raper & Varco, 2015). Recently, AOS have included a red-edge waveband of 710–740 nm which allows the calculation of the normalized difference red edge (NDRE) index introduced by Gitelson and Merzlyak (1994). Other VIs which utilize the red-edge include DATT (Datt, 1999), Meris terrestrial chlorophyll index (MTCI) (Dash & Curran, 2004), and the chlorophyll index using red edge (CIRE; Gitelson, Vina, Ciganda, Rundquist, & Arkebauer, 2005). The DATT and MTCI have the advantage of being sensitive to N with relatively less effect of water or irrigation level for corn (Shiratsuchi et al., 2011) and durum wheat (Bronson et al., 2017a). The canopy chlorophyll content index (CCCI) is a ratio of NDRE and NDVI and has been reported to have good sensitivity to N in cotton, broccoli, and wheat (Barnes et al., 2000; Cammarano et al., 2011; Long, Eitel, & Huggins, 2009; El-Shikha, Waller, Hunsaker, Clarke, & Barnes, 2007). Although there have been numerous evaluations of a large number of VI's for their ability to assess N status of corn (Clay, Kim, Chang, Clay, & Dalsted, 2006; Shiratsuchi et al., 2011) and wheat (Bronson et al., 2017a), fewer comparisons have been conducted for cotton (Oliveira et al., 2012, Raper & Varco, 2015).

The objectives of this study were to (a) compare the ability of 12 commonly used VIs derived from weekly canopy reflectance from an AOS to detect N deficiency in irrigated cotton N management studies; and (b) examine relations between the 12 VIs and in-season N fertilizer rate, leaf N, biomass, N uptake, cotton lint and seed yields.

2 | MATERIALS AND METHODS

Canopy reflectance was measured weekly using the Crop Circle ACS-470 AOS (Holland Scientific Inc., Lincoln, NE), starting about 2 wk after plant emergence and ending about mid-bloom in two N management irrigated cotton studies in Maricopa, AZ (33.067 N, 111.97 W and 360 m above sea level) on a Casa Grande sandy loam (fine-loamy, mixed, superactive, hyperthermic, Typic Natrargid); an overhead

sprinkler irrigation (OSI) study, 2014–2015 (Bronson et al., 2017b), and a subsurface drip irrigation (SDI) study, 2016–2018 (Bronson et al., 2019). In the OSI study, there were eight N treatments replicated four times, described in detail in Bronson et al. (2017b). In the SDI study there were five N treatments (two of these at a deficit 70% irrigation level) replicated three times as described in Bronson et al. (2019). The experimental design was a randomized complete block in each study. Both studies included soil test-based (0–90 cm pre-plant soil NO_3) N management, canopy reflectance-based N management, and zero-N treatments, which will be the focus of this manuscript. We will not present the 70% irrigation level data in this manuscript. In the OSI study, the reflectance-based N plots initially received 50% of the N rate of the soil test plots. If NDVIA from reflectance plot means fell significantly below the plot mean of NDVIA of the soil test-based N treatments, then the N rate was increased to match the soil test rate (designated “reflectance-based N-1” in Bronson et al., 2017b). In the SDI study, we used NDRE instead of NDVIA to base the same reflectance plot N treatment approach on (Bronson et al., 2019).

The Crop Circle ACS-470 sensor field of view was $30^\circ \times 14^\circ$. Data acquisition rate was 5 Hz, and one pass per plot was made. In 2014 and 2015 in the OSI study, two AOS Crop Circle ACS-470 sensors were mounted in tandem, one oriented forward, one oriented backward to maximize the separation between light sources, over row number three, from the total of six rows in each plot. Sensors were centered over the crop row with the long dimension of the rectangular footprint perpendicular to the row, and with 30 cm separation between light sources of the two sensors, to avoid overlap. In the SDI study of 2016–2018, two additional ACS-470 sensors were added to acquire canopy reflectance from plot rows four and five (of eight total) in each plot. The Crop Circle ACS-470 sensors had interference band-pass filters centered at 800 nm (20 nm width), 590 nm (10 nm width), and 730 nm (10 nm width). The second sensor in each row had filters at 550 nm (10 nm width), 530 nm (10 nm width), and 670 nm (10 nm width). Weekly calibration just prior to field data acquisition was carried out for one AOS at a time connected to the Holland Scientific FieldCAL SC-1. This consisted of zeroing the sensors' output while covered with black foam, followed by spanning the output to 1.0 with sensors 1.3 m above a 1.2×1.8 m titanium white-painted piece of plywood. Additionally, white board readings were taken for 60 s before and after field plot measurements were taken. Reflectance data from the plots for all sensor-wavebands (six in 2014–2015, 12 in 2016–2018) were divided by the unique post-run 60-s-average whiteboard reflectance reading of each waveband to adjust for the variations from 1.0. Sensors were mounted on the front arms of a Hamby high-clearance tractor and adjusted weekly to a height of 1 m above the plant canopy in the first plot that had a soil test-based N rate treatment (and 100% irrigation in

2016–2018). Crop Circle datum were logged to a Holland Scientific GeoSCOUT X datalogger. Differential GPS was logged with a Hemisphere (Hemisphere GPS, Calgary, AB, Canada) Crescent A100 GPS receiver. The offsets of the four sensors from the GPS receiver were entered into and accounted for in the GeoScout X datalogger.

NDVI-Red (NDVIR) (Tucker, 1979) was calculated:

$$(R_{800} - R_{670}) / (R_{800} + R_{670})$$

where R_{800} and R_{670} , are reflectance at 800 and 670 nm, respectively.

NDVI-Amber (NDVIA) (Solari et al., 2008) was calculated:

$$(R_{800} - R_{590}) / (R_{800} + R_{590})$$

where R_{590} , is reflectance at 590 nm.

NDVI-Green (NDVIG) (Gitelson et al., 1996) was calculated:

$$(R_{800} - R_{550}) / (R_{800} + R_{550})$$

where R_{550} , is reflectance at 550 nm.

The chlorophyll index using amber (CIA) (Gitelson et al., 2005) was calculated:

$$(R_{800}) / (R_{590}) - 1$$

The physiological reflectance index (PRI) (Penuelas, Gamon, Freedon, Merino, & Field, 1994) was calculated:

$$(R_{590} - R_{530}) / (R_{590} + R_{530})$$

where R_{530} , is reflectance at 530 nm.

Normalized difference vegetation index-Red-Red edge (NDRRE) (Gitelson, Kaufman, Stark, & Rundquist, 2002) was calculated:

$$(R_{730} - R_{670}) / (R_{730} + R_{670})$$

where R_{730} , is reflectance at 730 nm.

Normalized difference vegetation index-Amber-Red edge (NDARE) was calculated:

$$(R_{730} - R_{590}) / (R_{730} + R_{590})$$

Normalized difference red edge index (NDRE) (Gitelson & Merzlyak, 1994) was calculated:

$$(R_{800} - R_{730}) / (R_{800} + R_{730})$$

Canopy chlorophyll content index (CCCI) (Long et al., 2009; Barnes et al., 2000, Cammarano et al., 2011) was calculated:

$$\text{NDRE/NDVIR}$$

The DATT (Datt, 1999) was calculated:

$$(R_{800} - R_{730}) / (R_{800} - R_{670})$$

The Meris terrestrial chlorophyll index (MTCI) (Dash and Curran, 2004) was calculated:

$$(R_{800} - R_{730}) / (R_{730} - R_{670})$$

The chlorophyll index using red edge (CIRE) (Gitelson et al., 2005) was calculated:

$$(R_{800}/R_{730}) - 1$$

The VI data files were assigned to 6-m \times 1-m (2014-2015) and 6-m \times 2-m (2016-2018) vector polygons centered on two DGPS sampling anchor points per 36-m long OSI plot in 2014–2015 and on four DGPS sampling points per 100-m long SDI plot in 2016–2018 using Geoprocessing/Intersect procedures in ArcMap 10 (ESRI, 2015).

The effects of N treatment (and irrigation level in 2016–2018) on the 12 VIs at first bloom and mid bloom were estimated using the PROC MIXED procedure (SAS Institute, 2013). Replicate and replicate \times N treatment were considered random effects. Nitrogen treatment in both studies and irrigation level in the 2016–2018 SDI study were considered fixed effects. If the F-test for treatment was significant at $P = .05$, then pairwise comparisons of least square treatment means for each of the 12 VIs was estimated at $P = .05$ using PDIF (SAS, 2013). Additionally, we estimated single degree of freedom contrasts within PROC MIXED.

Leaves were sampled starting at first square, from the uppermost fully extended leaf, of 12 plants within 3 m of the DGPS sampling points. Leaf N was analyzed following drying and grinding on a Leco Truspec Leco-Truspec CN analyzer (Leco Corp., St. Joseph, MO). Biomass and total N uptake were determined at first open boll each year from 0.5 m of row from each of two rows of plants sampled at each DGPS point. Leaves, stems, seed, and burrs were analyzed for N content on the Leco CN analyzer. Lint and seed yields were sampled by a cotton picker on the center two rows on a 6-m length, centered on the DGPS points (Bronson et al., 2017b, 2019).

We also performed simple correlation of the 12 VIs (using means of vector polygons) at first bloom and mid bloom, leaf N at first bloom and mid bloom, N fertilizer rate, biomass at first open boll, N uptake at first open boll, lint yield, and seed yield using PROC CORR (SAS, 2013).

3 | RESULTS

Table 1 shows the day of year when the comparisons ‘Zero-N vs. soil test-based N’ and ‘Reflectance-based N management vs. soil test-based N’ were significant at the $P = .05$ level for

TABLE 1 Day of year that N deficiencies appeared for 12 vegetation indices (VIs) in nitrogen management studies of irrigated cotton and lint and seed yield contrasts, Maricopa, AZ, 2014–2018

VIs/yields	2014		2015		2016		2017		2018	
	Zero-N vs. soil test	Reflectance vs. soil test	Zero-N vs. soil test	Reflectance vs. soil test	Zero-N vs. soil test	Reflectance vs. soil test	Zero-N vs. soil test	Reflectance vs. soil test	Zero-N vs. soil test	Reflectance vs. soil test
NDVIA	210	NS ^a	203	217	153	159	150	164	134	176
NDVIR	217	NS	209	NS	153	159	150	NS	134	NS
NDVIG	210	NS	203	210	153	159	150	164	134	176
CIA	217	NS	203	217	153	159	150	164	134	176
PRI	217	NS	217	NS	153	159	150	NS	134	176
NDARE	NS	NS	210	NS	153	159	150	NS	141	176
NDRRE	NS	NS	210	NS	153	159	150	NS	134	NS
NDRE	203	NS	180	188	153	159	150	164	134	176
CCCI	210	NS	NS	NS	159	NS	157	164	148	NS
DATT	210	NS	229	NS	166	166	164	164	148	NS
MTCI	210	NS	NS	NS	166	166	164	164	141	NS
CIRE	203	NS	180	188	153	159	150	164	134	176
Lint yield	*	NS	*	*	*	NS	*	NS	*	NS
Seed yield	*	NS	*	NS	*	NS	*	NS	*	*

^aNS, not significant at $P < .05$.*Significant at $P < .05$.

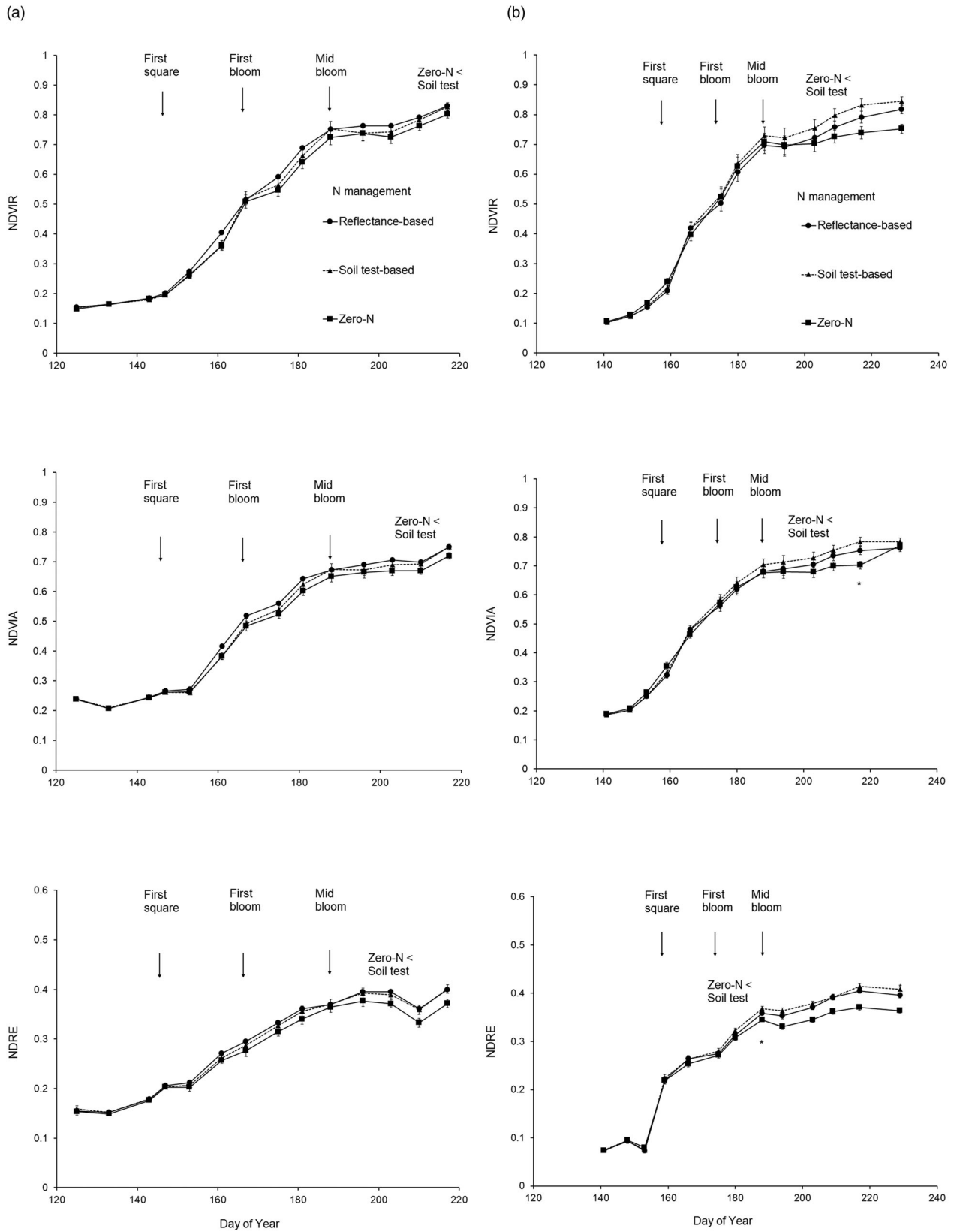


FIGURE 1 Normalized difference vegetation index red (NDVIR), normalized difference vegetation index amber (NDVIA), and normalized difference red edge index (NDRE) as affected by N fertilizer management in overhead sprinkler-irrigated cotton (Maricopa AZ) in (a) 2014 and (b) 2015. Downward arrows indicate N fertilizations. Asterisk (*) indicates Reflectance-based N strategy 1 plots are significantly < soil test-based N at $P < .05$. Standard error bars are shown for each date. The NDVIA figures are adapted from Bronson et al. (2017b)

all 12 VIs. This information is contained for NDVIR, NDVIA, and NDRE in Figure 1 and 2, but Table 1 allows for easily comparing all 12 VIs. In addition, Table 1 shows that lint and seed yields for all five site-years were significantly reduced in zero-N plots vs. soil test plots (Bronson et al., 2017b, 2019). This indicates that N deficiencies in zero-N plots remained until harvest. Nitrogen fertilizer rates were adjusted upwards in the reflectance plots in 2016 and 2017, when NDRE fell significantly < NDRE in soil test plots (Bronson et al., 2019), and yields were similar to the soil test plot yields. In 2014, 2015, and in 2018, reflectance plot N rates were not adjusted, but remained at 50% of soil test plot N rates. However, a lint yield depression in 2015 and a seed yield depression in 2018 were observed relative to the soil test plots (Bronson et al., 2017b, 2019).

The NDVIs showed a smooth, sigmoidal seasonal increase in each of the five site-years (NDVIR, NDVIA shown in Figures 1 and 2). The NDRE, on the other hand had greater variability between dates (Figures 1 and 2). The VIs NDVIR, NDVIA, and NDRE did not exhibit N deficiencies in zero-N plots relative to soil test plots under OSI in 2014 and 2015 until after mid bloom (Figure 1a and B). The NDRE and CIRE detected N deficiency as lower values with zero-N vs. soil test plots 7 and 23 d earlier than other VIs in 2014 and 2015, respectively (Table 1). This was mid bloom, day 203 in 2014, and first bloom, day 180 in 2015. No VI indicated N deficiency in reflectance plots vs. soil plots in 2014. In 2015, NDRE and CIRE showed a brief, 1-wk difference between reflectance-based N and soil test at mid bloom (day 188), the date of the last N fertilization and 22 d before NDVIG showed this difference (Figure 1b, Table 1). The NDVIA and CIA indicated N deficiency in reflectance-plots on day 217 in 2015. This was at peak bloom and was 29 d after the last N fertilization.

Nitrogen treatment differences in the VIs were much more pronounced and appeared early in the season under SDI in 2016–2018 (Figure 2, Table 1). Significantly reduced VIs with zero-N compared to soil test occurred from first squaring to early squaring in all 3 yr. In 2016 and 2017, all VIs except CCCI, DATT, and MTCI detected zero-N plot deficiencies on day 153 and 150 (early squaring), respectively. In 2018, all VIs except NDARE, CCCI, DATT, and MTCI declared zero-N plot deficiencies on day 134 (first square) (Table 1).

In 2016, all VIs showed reflectance plot N deficiency on day 159 (early squaring), except CCCI, and DATT and MTCI, which showed deficiency 7 d later (Table 1). The CCCI was the only VI in 2016 to not show reflectance plot N deficiency. The N fertilizer rate for reflectance plots was adjusted upwards to match the soil test N rate at this time. Figure 2 A indicates that it took 2–3 wk for the reflectance plots to recover from N deficiency with the higher rate of N fertigation in 2016. Reflectance plot N deficiency only occurred for 1 wk at first bloom in 2017 for NDVIA, NDVIG, CIA, CCCI,

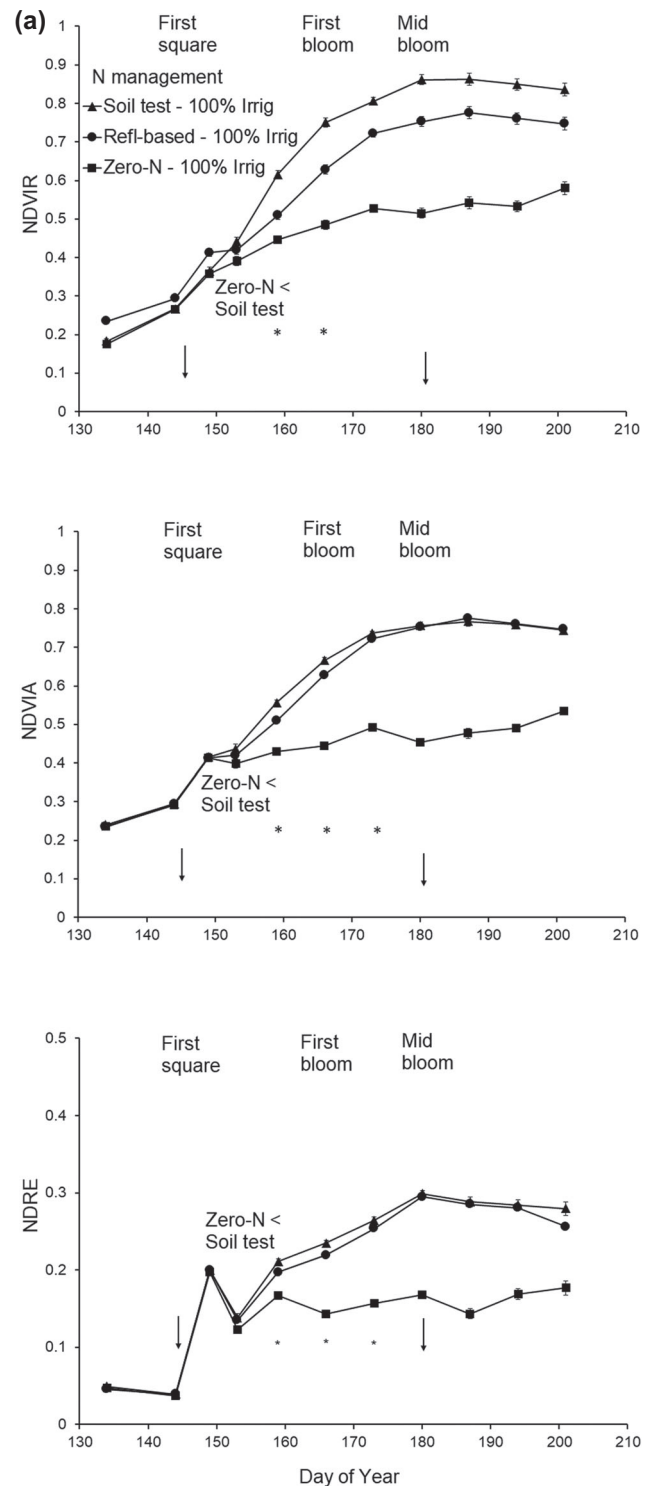


FIGURE 2 Normalized difference vegetation index red (NDVIR), normalized difference vegetation index amber (NDVIA), and normalized difference red edge index (NDRE) as affected by N fertilizer management in subsurface drip-irrigated cotton (Maricopa AZ) in (a) 2016, (b) 2017, and (c) 2018. Downward arrows indicate start and end 6-wk N fertigations. Asterisk (*) indicates Reflectance-based N plots are significantly < soil test-based N at $P < .05$. Standard error bars are shown for each date. The NDRE figures are adapted from Bronson et al. (2019)

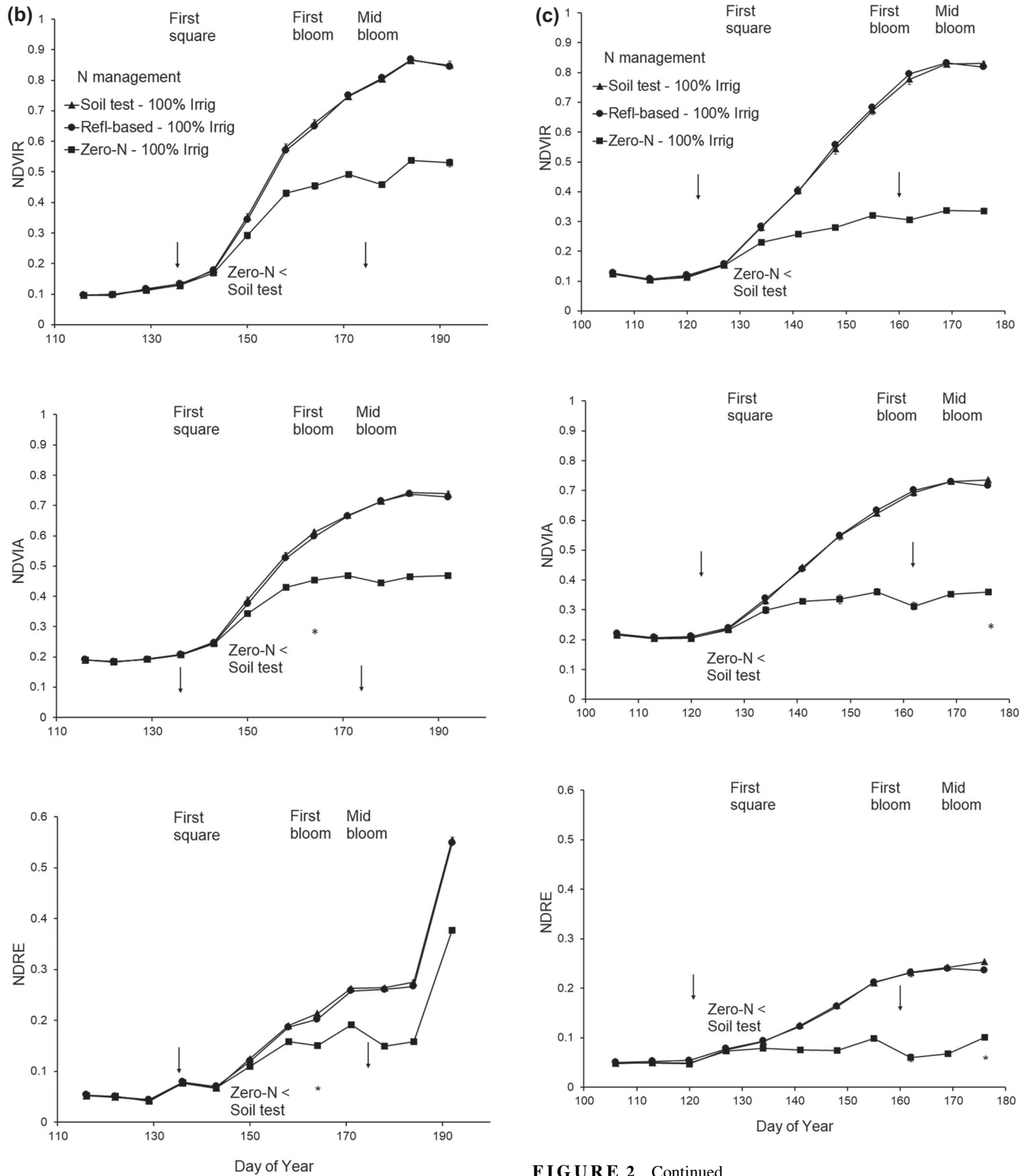


FIGURE 2 Continued

FIGURE 2 Continued

DATT, MTCI, and CIRE, but not for NDVIR, PRI, NADRE, or NDRRE (Figure 2b, Table 1). This was on day 164, when again the N fertilizer rate for reflectance plots was increased to match the soil test N rate. Reflectance plots recovered from N deficiency in just 1 wk in 2017. In 2018, NDVIA, NDVIG, CI, PRI, NDARE, and NDRE showed reflectance plot deficiency at day 176, which was at mid bloom and 17 d after the N fertigation ceased (Table 1).

Correlations between VIs, and individual waveband reflectance and N fertilizer rate, biomass, N uptake, leaf N, and yield were weak under OSI in 2014–2015 (Table 2). There were two weak VI correlations with N rate in 2014, and only weak correlation with four of 12 VIs in 2015. Lint and seed yield-VI correlations in 2014 at first bloom (0.35–0.62), increased to as high as 0.75 at mid bloom (Table 2). In 2015 there was moderate correlations under OSI with VIs and biomass and lint and seed yield of 0.25–0.41 (Table 2). On the other hand, correlations between VIs, waveband reflectance, N rate, and the measured plant variables were high under SDI in 2016–2018, with r ranging from .70 to .98. (Table 2). Exceptions to high correlations included CCCI in all 3 yr, and DATT and MTCI for 2017–2018. Correlations between reflectance at NIR, red edge, and visible wavebands and the plant parameters were strong in SDI from 2016–2018 with similar results among the wavebands (Table 2).

4 | DISCUSSION

The delayed response of VIs to N management treatments with OSI and the weak correlations with N rate and plant measures is consistent with the relatively small lint and seed yield response to N fertilizer reported by Bronson et al. (2017b). The early separation of VIs with N treatments with SDI and the high correlations with plant variables reflects the large lint and seed responses to N rate (i.e., delta yields) in that 3-yr study (Bronson et al., 2019). In addition to high water use efficiency, SDI had very high N recovery efficiency of 60–94% (Bronson et al., 2011; Bronson et al., 2019), which explains the large separation of VI data among N management treatments. Several VIs detected zero-N plot and reflectance plot deficiencies on the same day for 2016–2018. In 2014 and 2015, NDRE and CIRE detected zero-N plot deficiencies 1 wk and 23 d, respectively, before NDVIA did, and reflectance-based treatment deficiencies were evident 29 d earlier in 2015. Thus, a significant finding in the OSI study was that NDRE and CIRE detected N deficiency earlier than NDVIA, or other VIs. This is also reflected in the ANOVAs by date, and pairwise comparisons of N treatment VIs means. The correlation tables showed higher correlations with N rate, biomass, plant N, and yield for most of the 12 VIs in 2016–2018 than in 2014–2015. In general VIs in OSI in 2014 had greater correlation with biomass and less with leaf N, while

in SDI correlations were similar between leaf N and biomass. Separating correlations of VIs with leaf N vs. plant biomass is difficult, because the two plant variables are correlated. This is evident by the correlations of 800 nm NIR reflectance with leaf N, whereas NIR is usually not sensitive to leaf chlorophyll or leaf N (Hatfield, Gitelson, Schepers, & Walthall, 2008).

The greater efficiency of water and N utilization in SDI resulted in greater relative differences among N treatments compared to OSI (Bronson et al., 2019). It is clear that NDRE and CIRE declared zero-N plots deficient earlier than did the NDVIs in OSI, when the differences were small, and that in SDI with a rapid onset of large N treatment differences, the choice of VI is less critical.

The NDVIR only detected reflectance plot N deficiency in one of five site-years (i.e., 2016). We have reported earlier in Texas studies with irrigated cotton and with durum wheat in Arizona that NDVIA often detects N deficiencies slightly earlier than NDVIR (Bronson et al., 2011; Bronson et al., 2017a). This has been previously reported by Gitelson et al. (1996) and Shiratsuchi et al. (2011) and was attributed to the saturation of NDVIR in closed canopies.

It is important to discuss what we term “N deficiency” in this study. In 2014, for example, no differences in any of the VIs were observed across 14 wk for reflectance plots compared to the soil test plots. This, combined with the lack of lint or seed yield differences, clearly indicate that there were no N deficiencies in the reflectance plots. In 2015, the reflectance plot NDRE and CIRE fell significantly below that of the soil test plots on day 188, and a small lint yield depression of 105 kg ha⁻¹ were observed. We did not adjust the N fertilizer rate in the third application of 2015 because we were committed to using NDVIA, which did not show a reflectance plot deficiency on day 188 (but did on day 217) (Bronson et al., 2017b). 2016 and 2017 were cases where in-season N deficiencies in the reflectance plots were declared by several of the VIs, and the N fertilizer rate was adjusted upwards to match the soil test plot rates for the remainder of the 6-wk fertigation periods. The N deficiency disappeared in 3 wk in 2016 and in 1 wk in 2017, and lint and seed yields were similar between the reflectance and soil test plots (Bronson et al., 2019). 2018 was yet a different case, where VIs in the reflectance plots did not decline below VIs in the soil test plots until mid-bloom (day 176), which was after the 6-wk fertigation period. Bronson et al. (2019) suggested that, based on the 2018 result, which was that a seed yield loss in the reflectance plot was observed relative to the soil test plot seed yields, that an 8-wk fertigation period may be preferable to the 6-wk time frame in SDI cotton.

The NDRE is not nearly as well-studied in the literature as the NDVI, but its use in agricultural research is increasing (Ballester, Hornbuckle, Brinkhoff, Smith, & Quayle, 2017; Barnes et al., 2000; Bean et al., 2018; Long et al., 2009; Montealegre, Wortman, Ferguson, Shaver, & Schepers, 2019;

TABLE 2 Simple correlation of 12 vegetation indices (VIs) and reflectance from four wavebands of upland cotton with seed yield, lint yield, total biomass, leaf N, and total N uptake using PROC CORR, Maricopa, AZ, 2014–2018

VIs/wavebands	First bloom						Mid bloom					
	N fertilizer rate	Total biomass	Total N uptake	Leaf N	Lint yield	Seed yield	N fertilizer rate	Total biomass	Total N uptake	Leaf N	Lint yield	Seed yield
2014												
NDVIA	NS	0.49**	0.32**	NS	0.45**	0.42**	NS	0.69**	0.47**	NS	0.72**	0.72**
NDVIR	NS	0.49**	0.32**	NS	0.45**	0.42**	NS	0.58**	0.42**	NS	0.68**	0.68**
NDVIG	NS	0.49**	0.32**	NS	0.46**	0.43**	NS	0.60**	0.26*	NS	0.32**	0.35**
CIA	NS	0.49**	0.33**	NS	0.45**	0.42**	NS	-0.39**	-0.27*	0.27*	-0.59**	-0.60*
PRI	NS	-0.44**	-0.32**	NS	-0.37**	-0.35**	NS	-0.63**	-0.43**	NS	-0.71**	0.70**
NDARE	NS	0.56**	0.36**	NS	0.54**	0.50**	NS	0.60**	0.42**	NS	0.68**	0.68**
NDRRE	NS	0.54**	0.34**	NS	0.51**	0.48**	NS	0.61**	0.43**	NS	0.72**	0.72**
NDRE	NS	0.26*	NS	NS	NS	NS	NS	0.62**	0.43**	NS	0.71**	0.71**
CCCI	NS	-0.60**	-0.39**	NS	-0.60**	-0.55**	NS	0.60**	0.43**	NS	0.70**	0.70**
DATT	NS	-0.60**	-0.38**	NS	-0.62**	-0.56**	NS	-0.63**	-0.41**	0.25*	-0.75**	-0.73**
MTCI	NS	-0.59**	-0.36**	NS	-0.60**	-0.56**	NS	NS	NS	NS	NS	NS
CIRE	NS	0.26*	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
R ₈₀₀	NS	0.50**	0.32**	NS	0.44**	0.43**	NS	0.55**	0.35**	NS	0.51**	0.47**
R ₇₃₀	NS	0.59**	0.37**	NS	0.55**	0.53**	NS	0.65**	0.47**	NS	0.71**	0.71**
R ₆₇₀	NS	-0.46*	-0.29*	NS	-0.44**	-0.39**	NS	0.68**	0.48**	NS	0.74**	0.74**
R ₅₉₀	NS	-0.39**	-0.27*	NS	-0.39**	-0.34**	NS	-0.55**	-0.37**	NS	-0.67**	-0.66**
2015												
NDVIA	NS	0.25*	NS	NS	0.34**	0.31**	NS	0.30**	NS	NS	0.38**	0.36**
NDVIR	NS	NS	NS	NS	0.33**	0.30**	NS	0.26*	NS	NS	0.36*	0.34**
NDVIG	NS	0.25*	NS	NS	0.33**	0.31**	NS	0.33**	NS	NS	0.37**	0.37**
CIA	0.27*	0.28*	NS	NS	0.38**	0.36**	NS	0.31**	NS	NS	0.41**	0.41**
PRI	NS	-0.28*	NS	NS	-0.36**	-0.34**	NS	-0.25*	NS	NS	0.41**	-0.38**
NDARE	NS	NS	NS	NS	0.35**	0.31**	NS	0.28*	NS	NS	0.38**	0.37**
NDRRE	NS	NS	NS	NS	0.35**	0.31**	NS	NS	NS	NS	0.37**	0.34**
NDRE	NS	0.32**	NS	0.36**	0.32**	0.33**	NS	0.38**	0.38**	0.40**	0.30**	0.33**
CCCI	0.27*	NS	NS	0.29**	-0.27*	NS	NS	NS	NS	NS	0.41**	-0.26*
DATT	0.30**	NS	NS	0.35**	-0.25**	NS	0.29*	NS	NS	NS	-0.28*	NS
MTCI	0.28*	NS	NS	0.31**	NS	NS	NS	NS	NS	NS	NS	NS

(Continues)

TABLE 2 (Continued)

ViS/wavebands	First bloom						Mid bloom					
	N fertilizer rate	Total biomass	Total N uptake	Leaf N	Lint yield	Seed yield	N fertilizer rate	Total biomass	Total N uptake	Leaf N	Lint yield	Seed yield
CIRE	NS	0.34**	NS	0.36**	0.33**	0.34**	NS	0.38**	0.39**	0.42**	0.29*	0.33**
R ₈₀₀	NS	NS	NS	NS	0.39**	0.35**	NS	0.25*	NS	NS	0.43**	0.41**
R ₇₃₀	NS	NS	NS	-0.34**	0.38**	0.32**	NS	NS	NS	NS	0.43**	0.40**
R ₆₇₀	NS	-0.29*	NS	NS	-0.31**	-0.29*	NS	-0.28*	NS	NS	-0.33**	-0.31*
R ₅₉₀	NS	-0.45**	-0.39**	-0.55**	NS	NS	NS	-0.24*	-0.42**	-0.44**	NS	NS
2016												
NDVIA	0.96**	0.86**	0.93**	0.94**	0.85**	0.88**	0.95**	0.90**	0.94**	0.94**	0.90**	0.92**
NDVIR	0.95**	0.87**	0.93**	0.91**	0.86**	0.85**	0.93**	0.90**	0.93**	0.92**	0.92**	0.93**
NDVIG	0.96**	0.85**	0.93**	0.94**	0.85**	0.87**	0.98**	0.86**	0.93**	0.94**	0.82**	0.92**
CIA	0.96**	0.86**	0.94**	0.93**	0.84**	0.87**	0.93**	0.92**	0.94**	0.91**	0.89**	0.92**
PRI	-0.82**	-0.70**	-0.78**	-0.93**	-0.70**	-0.72**	-0.88**	-0.91**	-0.91**	-0.87**	-0.89**	-0.92**
NDARE	0.95**	0.88**	0.94**	0.91**	0.86**	0.89**	0.91**	0.91**	0.93**	0.90**	0.90**	0.93**
NDRRE	0.93**	0.89**	0.93**	0.89**	0.87**	0.90**	0.89**	0.90**	0.91**	0.87**	0.90**	0.93**
NDRE	0.98**	0.80**	0.90**	0.95**	0.81**	0.85**	0.98**	0.82**	0.91**	0.96**	0.82**	0.84**
CCCI	0.57**	NS	0.38**	0.62**	0.25*	NS	0.51**	NS	0.37**	0.47**	NS	NS
DATT	0.90**	0.65**	0.77**	0.90**	0.66**	0.67**	0.81**	0.59**	0.71**	0.91**	0.57**	0.58**
MTCI	0.90**	0.65**	0.77**	0.90**	0.65**	0.67**	0.81**	0.59**	0.71**	0.77**	0.57**	0.58**
CIRE	0.97**	0.80**	0.90**	0.95**	0.80**	0.82**	0.98**	0.80**	0.91**	0.97**	0.81**	0.83**
R ₈₀₀	0.90**	0.87**	0.92**	0.86**	0.83**	0.86**	0.76**	0.85**	0.81**	0.51**	0.80**	0.84**
R ₇₃₀	0.83**	0.87**	0.88**	0.78**	0.81**	0.85**	0.69**	0.81**	0.75**	0.44**	0.74**	0.79**
R ₆₇₀	-0.94**	-0.85**	-0.90**	-0.90**	-0.84**	-0.87**	-0.78**	-0.53**	-0.86**	-0.88**	-0.66**	-0.65**
R ₅₉₀	-0.88**	-0.63**	-0.76**	-0.88**	-0.66**	-0.68**	0.27*	0.53**	NS	NS	0.42**	0.46**
2017												
NDVIA	0.89**	0.76**	0.77**	0.87**	0.76**	0.74**	0.92**	0.84**	0.85**	0.96**	0.89**	0.88**
NDVIR	0.86**	0.75**	0.76**	0.86**	0.75**	0.74**	0.90**	0.84**	0.83**	0.95**	0.90**	0.89**
NDVIG	0.90**	0.76**	0.78**	0.88**	0.75**	0.74**	0.93**	0.84**	0.85**	0.97**	0.85**	0.84**
CIA	0.89**	0.77**	0.78**	0.83**	0.76**	0.75**	0.88**	0.84**	0.83**	0.94**	0.91**	0.90**
PRI	-0.85**	-0.77**	-0.77**	-0.86**	-0.75**	-0.73**	-0.86**	-0.84**	-0.82**	-0.92**	-0.91**	-0.90**
NDARE	0.87**	0.75**	0.76**	0.86**	0.76**	0.75**	0.89**	0.80**	0.80**	0.92**	0.85**	0.84**
NDRRE	0.84**	0.73**	0.74**	0.84**	0.76**	0.74**	0.87**	0.82**	0.84**	0.87**	0.94**	0.93**
NDRE	0.91**	0.76**	0.79**	0.86**	0.73**	0.71**	0.96**	0.84**	0.87**	0.97**	0.83**	0.82**
CCCI	NS	NS	NS	-0.43**	NS	NS	0.36**	NS	0.25*	NS	NS	NS

(Continues)

TABLE 2 (Continued)

VIs/wavebands	First bloom					Mid bloom				
	N fertilizer rate	Total biomass	Total N uptake	Leaf N	Seed yield	N fertilizer rate	Total biomass	Total N uptake	Leaf N	Seed yield
DATT	0.53**	0.44**	0.48**	0.41**	0.34**	0.89**	0.70**	0.77**	0.61**	0.57**
MTCI	0.31**	NS	NS	NS	NS	0.77**	0.56**	0.65**	0.61**	0.37**
CIRE	0.91**	0.76**	0.79**	0.87**	0.73**	0.96**	0.84**	0.87**	0.97**	0.82**
R ₈₀₀	0.82**	0.74**	0.74**	0.81**	0.71**	0.79**	0.79**	0.76**	0.87**	0.91**
R ₇₃₀	0.75**	0.70**	0.69**	0.78**	0.68**	0.70**	0.74**	0.69**	0.80**	0.90**
R ₆₇₀	-0.80	-0.68**	-0.69**	-0.80**	-0.70**	-0.89**	-0.82**	-0.82**	-0.94**	-0.87**
R ₅₉₀	-0.82	-0.65**	-0.68**	-0.78**	-0.68**	-0.84**	-0.65**	-0.71**	-0.72**	-0.47**
2018										
NDVIA	0.87**	0.95**	0.91**	0.95**	0.98**	0.88**	0.96**	0.92**	0.97**	0.93**
NDVIR	0.86**	0.95**	0.90**	0.95**	0.98**	0.87**	0.96**	0.91**	0.97**	0.93**
NDVIG	0.87**	0.95**	0.90**	0.95**	0.98**	0.89**	0.96**	0.92**	0.97**	0.93**
CIA	0.84**	0.94**	0.89**	0.93**	0.97**	0.83**	0.97**	0.91**	0.96**	0.92**
PRI	-0.86**	-0.95**	-0.90**	-0.95**	-0.96**	-0.83**	-0.97**	-0.90**	-0.96**	-0.92**
NDARE	0.93**	0.95**	0.89**	0.93**	0.97**	0.95**	0.97**	0.91**	0.96**	0.92**
NDRRE	0.92**	0.95**	0.89**	0.93**	0.97**	0.93**	0.96**	0.90**	0.96**	0.92**
NDRE	0.90**	0.93**	0.90**	0.95**	0.97**	0.90**	0.95**	0.92**	0.97**	0.92**
CCCI	NS	NS	0.26*	NS	0.27*	NS	NS	NS	NS	NS
DATT	0.58**	0.56**	0.55**	0.32**	0.60**	NS	NS	NS	NS	NS
MTCI	NS	NS	NS	NS	NS	0.90**	0.88**	0.88**	0.94**	0.86**
CIRE	0.89**	0.93**	0.90**	0.95**	0.97**	0.90**	0.96**	0.92**	0.97**	0.93**
R ₈₀₀	0.87**	0.94**	0.91**	0.93**	0.97**	0.75**	0.95**	0.87**	0.91**	0.89**
R ₇₃₀	0.85**	0.95**	0.91**	0.92**	0.96**	0.69**	0.92**	0.83**	0.87**	0.87**
R ₆₇₀	-0.80**	-0.91**	-0.84**	-0.90	-0.95**	-0.86**	-0.95**	-0.90**	-0.96**	-0.91**
R ₅₉₀	-0.74**	-0.86	-0.78**	-0.84**	-0.90**	-0.30*	NS	NS	NS	NS

^aNS is not significant at $P = .05$.

^bR₈₀₀, R₇₃₀, R₆₇₀, and R₅₉₀ are reflectance at 800, 730, 670, and 590 nm, respectively.

* **, *** are significant at the $P < .05$, and .01 level, respectively ($n = 64$ in 2014–2015, and $n = 60$ in 2016–2018, except for leaf N, where $n = 32$ in 2014–2015 and $n = 30$ in 2016–2018).

Raper & Varco, 2015; Shiratsuchi et al., 2011; Stamatiadis et al., 2019). Shiratsuchi et al. (2011) reported that in corn, NDRE was more responsive at high N fertilizer rates than was NDVI. The reason why NDRE and CIRE detected N deficiencies ahead of the other VIs in 2014–2015 must be due to the use of the red edge reflectance instead of a visible waveband, as the 800 nm NIR waveband was constant. Hatfield et al. (2008) reported on reflectance vs. chlorophyll content for corn (*Zea mays* L.) leaves. They presented that NIR (780 nm) reflectance was high, but not responsive to chlorophyll level. Red (670 nm) reflectance decreased with increasing chlorophyll, but plateaued quickly. Green reflectance vs. chlorophyll had a similar pattern to red, but leveled off much more gradually. Red edge (710 nm) reflectance, on the other hand was the most responsive waveband to increasing chlorophyll content, and only leveled off at the highest chlorophyll levels. Raper and Varco (2015) found the highest correlation with leaf N in field cotton with reflectance at 710 nm ($r = -.61$) compared to 19 other visible, and NIR wavebands. Read, Tarpley, McKinion, and Reddy (2002) studied reflectance of individual NIR, red edge, red, green, and blue wavebands on individual cotton leaves. They found weak correlation with NIR, red edge and visible reflectance to leaf chlorophyll and leaf N, and that ratioing the bands improved the correlations considerably. This is similar to what we observed in the present study. For example, in 2016, at mid bloom, correlations between 800 nm reflectance and biomass and leaf N were 0.85 and 0.51, respectively. Correlations between 730 nm reflectance and biomass and leaf N were 0.81 and 0.44. Correlation between NDRE, CIRE and biomass was a similar 0.82 and 0.81, respectively. Yet, correlation between NDRE, CIRE and leaf N increased to 0.96 and 0.97, respectively. Tarpley, Reddy, and Sassenrath-Cole (2000) reported that in estimating N content of cotton leaves, ratios of NIR to red edge were superior to ratios of NIR to visible wavebands. They suggested that the including NIR reflectance serves to normalize the data, and provide a baseline for the red edge, resulting in improved accuracy. Other researchers have employed the red edge inflection point to estimate cotton N status, but this requires hyperspectral reflectance data (Raper & Varco, 2015).

It was surprising that CCCI did not perform better in detecting N deficiency, considering that CCCI is the ratio of NDRE to NDVIR and that NDRE performed well in this regard. The CCCI has been reported to correlate well with plant N in wheat and durum wheat (Bronson et al., 2017a; Cammarano et al., 2011) and in cotton (Barnes et al., 2000). Recent reports with cotton indicated that CCCI performed as well as (Ballester et al., 2017) or better than NDRE (Raper & Varco, 2015) in estimating cotton N status in the field. Raper and Varco (2015) state that CCCI standardizes for biomass and would be effective for early-season N assessment. In our

study, however CCCI only correlated with biomass in one of five site-years (2014).

Nitrogen savings of up to 50% of the 0–90 cm soil test-based N recommendation were observed in the five site-years of cotton studies. Yield reductions associated with the reduced N rate reflectance plots were small and infrequent. In 2015, under OSI, a 105 kg lint ha⁻¹ reduction was observed, but not with seed yield (Bronson et al., 2017b). In 2018, under SDI, a 196 kg seed ha⁻¹ reduction with reflectance management was reported, but not with the higher-value lint (Bronson et al., 2019).

Newer commercial AOS products typically offer a NIR, red, and red edge waveband sensing, such as the Holland Scientific RapidSCAN CS-45 and CropCircle ACS-430, and the AgLeader OptRx Crop Sensor. The TOPCON CropSpec provides NIR and red edge wavebands. However, amber wavebands, as in Holland Scientific's CropCircle ACS-210, have largely been discontinued. Amber and green reflectance is required for CIA and PRI calculations. Ballester et al. (2017) recently employed a 5-band RedEdge, MicaSense camera, with visible, NIR, and red edge wavebands on an unmanned aerial vehicle to assess cotton N status in Australia. They reported that CCCI and NDRE were the VIs tested that best distinguished N treatments.

The CIRE, which performed just as well in assessing N status as NDRE, is calculated with NIR and red edge. Therefore, since NDRE and CIRE were the most consistent early N deficiency-detecting VIs in this study, we recommend using either one of them for irrigated in-season cotton N assessment and management.

The algorithm for using NDRE or CIRE to manage cotton could be the “saving N without hurting yield” approach described here, with the initial 50% N rate of a well-fertilized pre-plant soil test-based N rate plot or area (Bronson et al., 2017b; Bronson et al., 2019). Arnall et al. (2016) developed a VI-based N rate calculator for cotton. It entailed dividing NDVI by growing degree days, and the use of a response index relative to N-rich strips. Asking farmers to create N-rich or well-fertilized strips is often questioned as a drawback to the on-farm use of canopy reflectance sensors to manage N fertilizer. To address this, Holland and Schepers (2010) proposed the use of the high 95% VI readings in farmers' fields be used as a virtual well-fertilized reference. Further researcher plot and farmer field research is needed to test the use of NDRE and CIRE with these different VI-based algorithms for cotton N fertilizer management.

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ORCID

K. F. Bronson  <https://orcid.org/0000-0001-9447-5733>

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