

Comparison of optical sensors in assessing the nitrogen (N) status in corn

Comparación de sensores ópticos en la evaluación del estado de nitrógeno (N) en cultivo de maíz

Rodrigo Nogueira Martins^{1*}, Francisco de Assis de Carvalho Pinto¹,
Jorge Tadeu Fim Rosas¹, Fernando Ferreira Lima dos Santos¹, Lucas de Arruda Viana¹

ABSTRACT

Nitrogen (N) plays a key role for optimal plant growth and, in particular, for profitable crop production. Optimal N management using optical sensors can provide a rapid assessment of a crop N status. Therefore, this study aimed at comparing the efficiency of two chlorophyll meters in assessing the N status in corn plants when using an economical optimum dose of nitrogen under variable rate application. To do so, a field experiment using a randomized block design with 5 treatments and 5 replicates was carried out in Januária - Brazil (15° 28' 55" S, 44° 22' 41" W; 474 m) in 2018. Treatments consisted of comparing the pre-fixed N fertilization system with three strategies of variable rate N application using different thresholds of the nitrogen sufficiency index (NSI) in the V4, V8, V10, and V14 vegetative stages. As a result, both chlorophyll meters were capable of detecting the need for N application and differentiating between the plants that received or did not receive N application. In addition, readings from both sensors were highly correlated (> 0.92) from the V6 to the V18 stage. Lastly, both chlorophyll meters were able to identify the crop development variability and could be a suitable tool to monitor the nutritional status and for guiding N fertilization in corn crops.

Keywords: chlorophyll meter, nitrogen sufficiency index, site-specific fertilization, *Zea mays* L., precision sensing.

RESUMEN

El nitrógeno (N) desempeña un papel clave para el crecimiento óptimo de las plantas y, en particular, para la producción rentable de cultivos. La gestión óptima de N mediante sensores ópticos puede proporcionar una evaluación rápida del estado de N de un cultivo. Por lo tanto, este estudio tuvo como objetivo comparar la eficiencia de dos medidores de clorofila en la evaluación del estado de N en las plantas de maíz cuando se utiliza una dosis óptima de nitrógeno económica bajo una aplicación de tasa variable. Para hacerlo, se realizó un experimento de campo utilizando un diseño de bloques al azar con 5 tratamientos y 5 réplicas en Januária - Brasil (15 ° 28 ' 55' ' S, 44 ° 22' 41 ' ' O; 474 m) en 2018. Los tratamientos consistieron en comparar el sistema de fertilización de N preestablecido con tres estrategias de aplicación de N de velocidad variable utilizando diferentes umbrales del índice de suficiencia de nitrógeno (NSI) en las etapas vegetativas V4, V8, V10 y V14. Como resultado, ambos medidores de clorofila fueron capaces de detectar la necesidad de la aplicación de N y diferenciar entre las plantas que recibieron o no la aplicación de N. Además, las lecturas de ambos sensores estaban altamente correlacionadas (> 0,92) desde la etapa V6 a la etapa V18. Por último, ambos medidores de clorofila pudieron identificar la variabilidad del desarrollo del cultivo y podrían ser una herramienta adecuada para monitorear el estado nutricional y para guiar la fertilización con N en los cultivos de maíz.

Palabras claves: medidor de clorofila, índice de suficiencia de nitrógeno, fertilización específica del sitio, *Zea mays* L., detección de precisión.

Introduction

Brazil is one of the world's largest producer and exporter of corn (*Zea mays* L.). In the 2018/19 harvest, the corn production reached approximately 100 million tons of grain with an average yield of 5.71 tons per hectare (CONAB, 2019). Among the nutrients demanded in its production, Nitrogen (N)

plays an important physiological role for optimal plant growth and, in particular, when aiming at maximizing the economic performance of crops (Schlichting *et al.*, 2015). However, there is still a challenge in improving the synchronism between the application time and the plant demand since this nutrient's requirement varies spatially and temporally.

¹ Department of Agricultural Engineering, Federal University of Viçosa, Viçosa, Brazil.

* Corresponding author: rodrigonmartins@hotmail.com

One way to optimize N application in corn plants is by using optical sensors, which are non-destructive leaf sampling methods and that enable rapid detection and correction of the plant nutritional status. Differently, lab-based methods require destructive plant sampling and chemical analysis, which are expensive, time-consuming, and labour-intensive (Fitzgerald *et al.*, 2010). The suitability of optical sensors for guiding N application has been widely documented for several crops such as corn, wheat, and rice (Ali *et al.*, 2018; Ali *et al.*, 2015; Bragagnolo *et al.*, 2013; Kitchen *et al.*, 2010; Varinderpal-Singh *et al.*, 2010). However, regarding corn, there is still a lack of information about the potential of different chlorophyll meter models in assessing N status when refining an economical optimum dose.

There are many optical sensors models available at different costs of acquisition; however, studies comparing their efficiency and capability of detecting N status in plants are still incipient. Schlichting *et al.* (2015) evaluated the effectiveness of two chlorophyll meters (Minolta SPAD 502 and Falker ClorofiLOG 1030) in assessing the nutritional status of wheat plants and concluded that there was no difference between them concerning the indirect determination of leaf chlorophyll content. Other comparison studies in crops like corn, potato, and Tifton 85 grass have showed that readings acquired by optical sensors, such as SPAD-502, N-Tester, and the Falker ClorofiLOG 1030 were capable of detecting the N status in the crop and its nutritional variation in the field (Barbieri junior *et al.*, 2012; Gianquinto *et al.*, 2004).

Thus, despite the availability of several studies regarding the use of optical sensors in crops such as corn, no study has yet compared the efficiency of different hand-held chlorophyll meters under the perspective of improving the nitrogen use efficiency (NUE), through the application of an economical optimum dose. Therefore, this study aimed at comparing the technical efficiency of two chlorophyll meters (Minolta SPAD-502 and Falker ClorofiLOG 1030) in assessing the N status in corn plants when using an economical optimum dose of nitrogen under variable rate application.

Material and Methods

Optical sensors specifications

Two optical sensors were evaluated: SPAD-502 (Soil and Plant Analysis Development) of

Minolta Corporation, Osaka, Japan (1989); and the Falker ClorofiLOG 1030 (Falker Agricultural Automation, Brazil). These sensors are active and function based on the quantification of the chlorophyll index (i.e., sensor readings), which is determined by the light flux, transmitted through the leaf. The SPAD-502 measures leaf transmittance centered at two wavelengths (λ), 650 nm (red) and 940 nm (infrared), and results in the SPAD index. Leaf transmittance measured at 650 nm range lies between the two primary wavelengths associated with chlorophyll activity (645 and 663 nm). The 940 nm wavelength serves as an internal reference to compensate for differences in leaf thickness, water status, or other factors (Waskom *et al.*, 1996). In contrast, the ClorofiLOG 1030 measures leaf transmittance at three wavelengths (λ), 635 and 660 nm (red) and 880 nm (infrared), and indicates the Falker index, which is proportional to chlorophyll absorbance.

Field experiment

The study was carried out in Januária-Minas Gerais state, Brazil (15° 28' 55" S; 44° 22' 41" W; 474 m) from February to June 2018. The local climate is defined as tropical humid with a mean temperature of 27 °C and mean rainfall of 900 mm.

Prior to sowing, the soil classified as Quartzarenic Neosol had the fertility of each block assessed and corrected with exception to N (Alvarez, 1999). After that, the corn hybrid DKB310 PRO 2 was manually sown in 02/09/2018 with 5 plants m^{-1} (62500 seeds ha^{-1}). Each experimental plot was composed of five lines with plants spaced 0.8 m between themselves and 6.0 m long (6.0 x 3.2 m, 19.2 m^2). The useful area consisted of the three central lines, excepting 0.5 m from each line's end.

The experiment was arranged in a randomized block design with 5 treatments and 5 replicates. The treatments were: T0: Control treatment; T1: Reference treatment, with 112 kg ha^{-1} of N split into two equal doses, which were applied in the V4 (four expanded leaves stage) and the V8 stage; T2: Application of 44.8 kg ha^{-1} of N in V4 + 22.4 kg ha^{-1} of N under variable rate considering a fixed value of the nitrogen sufficiency index (NSI < 95%) in the stages V8, V10 and V14, respectively; T3: 44.8 kg ha^{-1} of N in V4 + 22.4 kg ha^{-1} of N under variable rate considering a variable NSI (< 98, 95 and 93%) in the same stages; and T4: 33.6

kg ha⁻¹ of N in V4 + 33.6, 28.0 and 16.8 kg ha⁻¹ of N applied under variable rate using a variable NSI (< 98, 95 e 93%) in V8, V10 and V14, respectively.

The reference dose (112.00 kg ha⁻¹ of N) corresponded to an economical optimum dose of N, which was defined according to the methodology proposed by Silva *et al.* (2016) using data from another field experiment under similar conditions (Andrade *et al.*, 2014). The NSI of each plot was obtained using the equation: $NSI = SR_{VRP} / SR_{RP}$, where: SR_{VRP} corresponded to the mean of thirty sensor readings in the variable rate application plots; and SR_{RP} , which were the same amount of sensor readings in the reference plot (Francis; Piekielek, 1999).

Thus, from the V2 until the V18 vegetative stage, readings of both chlorophyll meters were taken disregarding the margins and the midrib of the last fully expanded leaf on 30 plants chosen randomly in the useful area of each plot. Additionally, at the V8, V10 and V14 stages, discs with 0.7 cm in diameter were collected from the

same leaves to quantify the total leaf nitrogen content using the Kjeldahl method.

After the field experiment, both sensors readings from V2 to the V18 stage were submitted to a simple linear correlation to compare the accuracy between their readings. Additionally, the relative chlorophyll index and the leaf N content of the V8, V10, and V14 stages were analyzed through the F test (Anova) and Tukey's test to validate both sensors as an indicator of the N status. All statistical analyses were performed using the Rstudio software (R Core Team, 2019).

Results and Discussion

The linear correlation analysis showed that the readings from both chlorophyll meters, with exception to the V2 stage (two expanded leaves), were significant ($p < 0.01$), and positively correlated with an increasing tendency throughout the development of the plant (Figure 1).

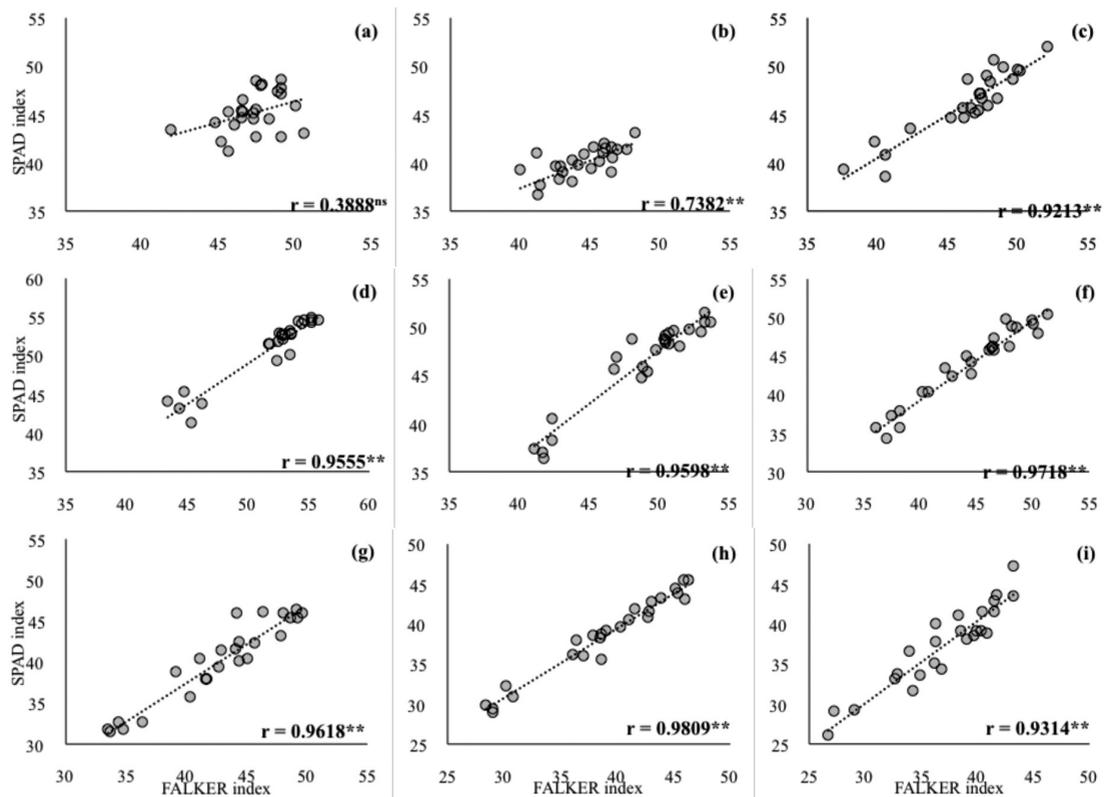


Figure 1. Pearson's correlation between readings of the SPAD-502 and Falker CFL 1030 chlorophyll meters in corn plants from the V2 (a) until the V18 stage (i) in Januária, Brazil, 2018.

** and ^{ns} are respectively significant to 1% ($p < 0.01$) probability and non-significant.

Lower correlations in early stages of the crop can be explained by the high variation of the chlorophyll index among plants since the field sampling was performed using different plants for each sensor. Moreover, the greatest variation in readings of these sensors in initial stages is possibly associated to the critical period of establishment of the crop in the field, in which the physiological condition of the plants is directly affected by the seed nutrient reserve (Muhammad *et al.*, 2015).

In addition, it is possible that the sandy characteristic of the soil (Quartzarenic Neosol) in which the experiment was implanted, associated to the processes that interfere in the N dynamics in the soil (leaching, volatilization, immobilization-mobilization, nitrification, denitrification and mineralization) may have caused changes in N availability during this period as reported in another study (Rambo *et al.*, 2004). Furthermore, lower soil cover at early stages of the plant probably resulted in lower N uptake by roots, which led to a variation in leaf chlorophyll concentration and, consequently, in the sensor's reading.

The correlations tended to be more uniform from the V6 stage, as it can be observed in Figure 1c, where from this stage to the V18 the value was higher than 0.92. Although the relationship of leaf N content with these sensors readings was not evaluated, the correlation between the sensors readings reinforces affirmations from other studies, in which optical sensors of foliar contact presented greater efficiency from the V8, since from this stage the plant, already established in the field, defines the ear's components, such as, the number of rows, length, and number of grains per row (Argenta *et al.*, 2004; Ritchie *et al.*, 1993; Varvel *et al.*, 1997).

The SPAD and FALKER indexes obtained by both chlorophyll meters and leaf N content

differed as a function of the different nitrogen management in coverage and in function of the evaluated phenological stages, as shown in Table 1. In the V8 stage, both the SPAD-502 and ClorofiLOG 1030 were capable of detecting the chlorophyll index difference between treatments that received or not the initial doses of 56.00, 44.80 and 33.60 kg ha⁻¹ of N in V4.

Regarding leaf N content, the result was different, since only the reference treatment (T1) was superior, whereas the variable rate treatments (T2, T3, and T4), although they presented the same average to T1, they did not differ to the control treatment (T0). This behavior is possibly attributed to the higher presence of ammoniac nitrogen at the early stages of the crop (i.e., stages of lower nutritional demand), and which was accounted in the foliar N analysis.

On the other hand, since ammoniac nitrogen is not accounted for the chlorophyll molecule and is not taken into consideration by these sensors' readings, they were less sensitive in differentiating the SPAD or Falker index among the treatments that received fertilization. Regardless, the results showed that the difference observed in the sensor reading was also obtained in the foliar N analysis for treatments that received or did not receive N fertilization.

In the V10 stage, the mean SPAD index of T1 was higher than treatments T2, T3, and T4 and in relation to T0. This difference was expected, since in the V8 stage, the T1 received the second dose of 56.00 kg ha⁻¹ of N, while treatments T2, T3, and T4 received, based on the NSI value, the doses of 22.40 kg ha⁻¹ of N (T2 and T3) and 33.60 kg ha⁻¹ of N (T4), respectively. Regarding the Falker index, there was an overall reduction in the mean values of each treatment compared to the SPAD

Table 1. Average values of the SPAD and FALKER index and leaf N content in the V8, V10, and V14 stages from the corn hybrid DKB310 PRO2 in Januária, Brazil, 2018.

Treatment	SPAD index			FALKER index			Leaf N content (g kg ⁻¹)		
	V ₈	V ₁₀	V ₁₄	V ₈	V ₁₀	V ₁₄	V ₈	V ₁₀	V ₁₄
T1	54.52 ^a	53.12 ^a	48.94 ^a	53.88 ^a	50.72 ^a	46.54 ^a	33.65 ^a	27.91 ^a	24.28 ^a
T2	53.02 ^a	49.32 ^b	42.40 ^b	52.44 ^a	47.18 ^b	39.50 ^b	28.89 ^{ab}	26.85 ^{ab}	21.65 ^a
T3	53.76 ^a	49.68 ^b	43.94 ^b	52.20 ^a	47.78 ^b	41.28 ^b	29.73 ^{ab}	25.37 ^{ab}	21.24 ^a
T4	53.36 ^a	49.82 ^b	44.04 ^b	52.86 ^a	48.18 ^b	42.84 ^{ab}	31.13 ^{ab}	27.19 ^{ab}	23.81 ^a
T0	44.86 ^b	41.88 ^c	34.56 ^c	43.40 ^b	37.92 ^c	32.00 ^c	26.68 ^b	22.77 ^b	17.49 ^b

Means followed by the same letter do not differ statistically at 5% probability, by Tukey's test.

index. For leaf N, the behavior did not differ from that obtained in the V8 stage, which reinforces the existence of differences in the nutritional status between the treatments that received or did not receive N fertilization.

For the V14 stage, the SPAD index presented a behavior similar to that obtained in V10. On the other hand, the highest Falker index average was observed again in T1 treatment. However, it was not different from the T4 treatment, which was also not superior to T2 and T3. Differently, all treatments were higher than T0, and both chlorophyll meters were sensitive to detect this difference in the readings. The leaf N content again showed that all the treatments that received N were superior to the control treatment (T0).

In the decision-making evaluation, regarding the performance of nitrogen fertilization in the V8, V10, and V14 stages, the number of correct decisions by the sensors was defined by the number of repetitions of each treatment in the field (five repetitions as previously described). The Figure 2 shows the number of correct decisions between the SPAD-502 and ClorofiLOG 1030 following the proposed NSI thresholds at each monitored stage.

In the V8 stage, the percentage of correct decision-making between both chlorophyll meters was 100% (15 correct) independent of the NSI used of 0.95 (T2) or 0.98 (T3 and T4). In V10 and V14 stages, a reduction in the number of correct decisions was observed, since in V10 the value was 86.67% (13 correct), while in V14 it was 93.33% (14 correct). This variation in the percentage of correct decisions among chlorophyll meters was possibly caused by the variation in chlorophyll content among the different plants sampled in each plot.

Regardless of the percentage of correct decisions observed, it is not possible to confirm whether the SPAD-502 or the ClorofiLOG underestimated or overestimated the need for N application, as in the correlation analysis the observed values from the V6 stage were higher than 0.9, in other words, the readings of both sensors are highly correlated. In addition, the comparison of the means showed that they were sensitive in differentiating the nutritional status of plants that received or did not receive N fertilization. In addition, it is possible that the application of lower doses in coverage from the V8 resulted in a lower accumulation of chlorophyll molecules in the plant tissues and, consequently, a low variation in the reading of these sensors between the different treatments.

In general, both the SPAD-502 and ClorofiLOG 1030 were efficient in detecting the need for N application and in differentiating between plants that received or did not receive N fertilization. This confirms the results observed in several studies using the SPAD-502. However, the ClorofiLOG 1030 is an advantageous alternative to the SPAD-502 since besides presenting similar technical performance, it presents an acquisition cost lower than the SPAD-502 model.

Conclusions

The chlorophyll meters SPAD-502 and ClorofiLOG 1030 were both capable of detecting the need for N application and in differentiating between plants that received or did not receive nitrogen fertilization when an economical optimum dose was used. In addition, readings from both sensors were highly correlated (> 0.92) from the V6 to the V18 stage. Lastly, both

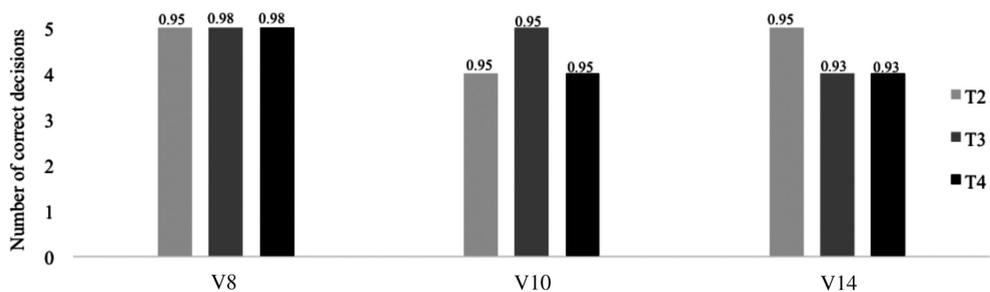


Figure 2. Number of correct decisions for recommendation of nitrogen fertilization by the SPAD-502 and ClorofiLOG 1030 using the NSI thresholds.

chlorophyll meters were able to identify the crop development variability and could be a suitable tool to monitor the nutritional status and for guiding N fertilization in corn crops.

Acknowledgments

This study was financed by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brazil (CAPES) - Finance code 001.

Literature Cited

- Ali, A. M.; Abou-Amer, I.; Ibrahim, S.M.
2018. Using GreenSeeker active optical sensor for optimizing maize nitrogen fertilization in calcareous soils of Egypt. *Archives of Agronomy and Soil Science*, 64 (8): 1083-1093.
- Ali, A.M.; Thind, H.S.
2015. A framework for refining nitrogen management in dry direct-seeded rice using GreenSeeker™ optical sensor. *Computers and Electronics in Agriculture*, 110: 114-120.
- Alvarez V.V.H.; Ribeiro, A.C.
1999. Calagem. In: *Recomendações para o uso de corretivos e fertilizantes em Minas Gerais - 5a aproximação*. Viçosa, MG, Brazil. pp. 43-60.
- Andrade, F.R.; Petter, F.A.; Nóbrega, J.C.A.; Pacheco, L.P.; Zuffo, A.M.
2014. Desempenho agrônômico do milho a doses e épocas de aplicação de nitrogênio no Cerrado piauiense. *Revista de Ciências Agrárias Amazonian Journal of Agricultural and Environmental Sciences*, 57(4): 358-366.
- Argenta, G.; Silva, P.D.; Bortolini, C.G.; Forsthofer, E.L.; Strieder, M.L.
2001. Relação da leitura do clorofilômetro com os teores de clorofila extraível e de nitrogênio na folha de milho. *Revista Brasileira de Fisiologia Vegetal*, 13(2): 158-167.
- Argenta, G.; Silva, P.R.F.D.; Sangoi, L.
2004. Leaf relative chlorophyll content as an indicator parameter to predict nitrogen fertilization in maize. *Ciência Rural*, 34(5): 1379-1387.
- Junior, É.B.; Rossiello, R.O.P.; Silva, R.V.M.M.; Ribeiro, R.C.; Morenz, M.J.F.
2012. Um novo clorofilômetro para estimar os teores de clorofila em folhas do capim Tifton 85. *Ciência Rural*, 42(12): 2242-2245.
- Bragagnolo, J.; Amado, T.J.C.; Nicoloso, R.S.; Jasper, J. Kunz, J.; Teixeira, T.G.
2013. Optical crop sensor for variable-rate nitrogen fertilization in corn: I. Plant nutrient and dry matter production. *Revista Brasileira de Ciência do Solo*, 37(5): 1288-1298.
- CONAB.
2019. Acompanhamento da safra brasileira de grãos: décimo levantamento. Companhia Nacional de Abastecimento. Julho 2019. v. 7, safra 2018/19, n. 10. Available: <https://www.conab.gov.br/info-agro/safra/graos>. Consulted: 13/Mar/2020.
- Falker Automação Agrícola Ltda.
2008. Manual do medidor eletrônico de teor clorofila (ClorofiLOG / CFL 1030). Falker Porto Alegre, Brazil. 33 p.
- Fitzgerald, G.; Rodriguez, D.; O'Leary, G.
2010. Measuring and predicting canopy nitrogen nutrition in wheat using a spectral index - The canopy chlorophyll content index (CCCI). *Field Crops Research*, 116 (3): 318-324.
- Francis, D.D.; Piekielek, W.P.
1999. Assessing crop nitrogen needs with chlorophyll meters. Site-Specific Management Guidelines, Potash & Phosphate Institute. SSMG-12. 1-4.
- Gianquinto, G.; Goffart, J.P.; Olivier, M.; Guarda, G.; Colauzzi, M.; Dalla Costa, L.; Mackerron, D.K.L.
2004. The use of hand-held chlorophyll meters as a tool to assess the nitrogen status and to guide nitrogen fertilization of potato crop. *Potato Research*, 47 (1-2):35-80.
- Kitchen, N.R.; Sudduth, K.A.; Drummond, S.T.; Scharf, P.C.; Palm, H.L.; Roberts, D.F.; Vories, E.D.
2010. Ground-based canopy reflectance sensing for variable-rate nitrogen corn fertilization. *Agronomy Journal*, 102 (1):71-84.
- Minolta Camera Corporation Ltd.
1989. Manual for Chlorophyll Meter SPAD 502. Minolta Radiometric Instruments Division. Osaka, Japon. 22 p.
- Muhammad, I.; Kolla, M.; Volker, R.; Günter, N.
2015. Impact of nutrient seed priming on germination, seedling development, nutritional status and grain yield of maize. *Journal of plant nutrition*, 38 (12): 1803-1821.
- R Core Team.
2019. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.
- Rambo, L.; Silva, P.R.F.D.; Argenta, G.; Bayer, C.
2004. Testes de nitrato no solo como indicadores complementares no manejo da adubação nitrogenada em milho. *Ciência rural*, 34 (4): 1279-1287.
- Ritchie, S.W.; Hanway, J.J.; Benson, G.O.
1993. How a corn plant develops. (Special Report, 48) University of Science and Technology. Ames, Iowa State, US. 21 p.
- Schlichting, A.F.; Bonfim-Silva, E.M. ; Silva, M.D.C.; Pietro-Souza, W.; da Silva, T.J.; Farias, L.D.N.
2015. Efficiency of portable chlorophyll meters in assessing the nutritional status of wheat plants. *Revista Brasileira de Engenharia Agrícola e Ambiental*, 19 (12): 1148-1151.
- Silva, S.; Costa, C.T.S.; do Nascimento, R.; Teodoro, I.; Cantarelli, A.L.D.; da Silva Soares, M.A.
2016. Épocas de plantio e dose econômica de nitrogênio para o milho cultivado nos Tabuleiros Costeiros Alagoanos. *Revista Agro@mbiente On-line*, 10 (2): 114-122.
- Tremblay, N.; Wang, Z.; Ma, B.L.; Belec, C.; Vigneault, P.
2009. A comparison of crop data measured by two commercial sensors for variable-rate nitrogen application. *Precision Agriculture*, 10 (2): 145.
- Waskom, R.M.; Westfall, D.G.; Spellman, D.E.; Soltanpour, P.N.
1996. Monitoring nitrogen status of corn with a portable chlorophyll meter. *Communications in Soil Science and Plant Analysis*, 27 (3-4): 545-560.

Thind, H.S.; Gupta, R.K.

2010. Need based nitrogen management using the chlorophyll meter and leaf colour chart in rice and wheat in South Asia: a review. *Nutrient Cycling in Agroecosystems*, 88 (3): 361-380.

Varvel, G.E.; Schepers, J.S.; Francis, D.D.

1997. Ability for in-season correction of nitrogen deficiency in corn using chlorophyll meters. *Soil Science Society of America Journal*, 61 (4): 1233-1239.

