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M Ramesh Naik
Department of Agronomy, S.V. Agricultural College, Tirupati, ANGRAU, Andhra Pradesh, India

S Hemalatha
Department of Agronomy, S.V. Agricultural College, Tirupati, ANGRAU, Andhra Pradesh, India

A Pratap Kumar Reddy
Department of Agronomy, ARS, Utkur, Kadapa, ANGRAU, Andhra Pradesh, India

KV Naga Madhuri
Department of Soil Science, IFT, RARS, Tirupati, ANGRAU, Andhra Pradesh, India

V Umamahesh
Department of Crop Physiology, S.V. Agricultural College, Tirupati, ANGRAU, Andhra Pradesh, India

Correspondence
M Ramesh Naik
Department of Agronomy, S.V. Agricultural College, Tirupati, ANGRAU, Andhra Pradesh, India

Need based nitrogen management on growth attributes of *Rabi* maize (*Zea mays* L.) using leaf colour chart under varied plant density

M Ramesh Naik, S Hemalatha, A Pratap Kumar Reddy, KV Naga Madhuri and V Umamahesh

Abstract
Fertilizer nitrogen application to maize crop is generally low or excess in quantity than required amount without assessing the crop demand for nitrogen. To obtain maximum possible yield in maize nitrogen fertilizer should be applied based on the requirement of the crop in different splits. A field experiment was conducted at Wet land farm of Sri Venkateswara Agricultural College, Tirupati during *Rabi* season of 2017-18 to calibrate leaf colour chart (LCC) for nitrogen management under varied plant density to establish threshold leaf colour greenness as a guide for right quantity of fertilizer N to be top dressed in maize. The experiment consisted of twenty seven treatment combinations comprising of three planting densities (66,666, 83,333 and 1,11,111 lakhs ha^{-1}), three nitrogen levels (30, 35 and 40 kg N ha^{-1}) and three LCC threshold values (LCC 4, 4.5 and 5). The results disclosed that the initial & final plant population, plant height, drymatter accrual and finally yield of maize increased significantly with increase in plant density, with application of fertilizer nitrogen at higher dose *i.e.* 40 kg ha^{-1} by using higher LCC threshold value (LCC-5) at different splits. Whereas, leaf area per plant and SPAD chlorophyll readings were lessened significantly with increase in plant densities, however it was increased with application of nitrogen at higher rate *i.e.* 40 kg ha^{-1} by using higher LCC threshold value (LCC-5) at various splits. Finally, experimental results manifested that, fine-tuning of N dose and time of application by number of splits which synchronize the crop demand and is the best management strategy for sustainable crop growth and yield.

Keywords: Plant population, nitrogen, leaf colour chart, SPAD meter, maize and yield

1. Introduction
Maize (*Zea mays* L.) is the primary staple food crop in many developing countries and it grown as high land cereal crop throughout the world. It is the third most important staple food crop both in terms of area and production after rice and wheat in India. In India maize is cultivated in an area of 9.22 million hectares with a production of 28.72 million tonnes and productivity of 3.12 t ha^{-1} (USDA, 2018) [17]. India contributes about 2 per cent of the world's maize produce (NCoMM Special Report, 2017) [10].
Generally in maize, lower plant population and poor nutrient management practices are the major factors affecting the yield (Dawadi and Sah, 2012) [4]. Plant spacing or population is most important factor which plays a significant role on growth, development and yield of maize. Generally, optimum plant population utilize solar radiation and nutrients efficiently. Management of row spacing is a crucial factor for increasing the productivity of maize. Proper plant density is important from the point of intercepting sunlight for photosynthesis besides, efficient use of plant nutrients and soil moisture. The most important role of N in the plant is one of the constituent of protein and nucleic acids and chlorophyll that enables the plant to transfer energy from sunlight through photosynthesis. Therefore, adequate supply of N is necessary to achieve and sustain higher yield potential in crops (Sharifi and Namvar 2016) [13]. Nitrogen plays critical function attributable to increase in grain yield and it affects production more than any other element.
To determine the nitrogen status of standing crops, we need to monitor with different techniques to answer the questions of when, where and how much of fertilizer required. Few techniques like tissue test, inexpensive Leaf colour chart (LCC), SPAD meters and Green seeker will be used to take decision. Real time nitrogen management through innovative gadgets such as chlorophyll meter, which is faster than tissue testing for nitrogen

determination, can render to find when plant requires more nitrogen. Owing to expensive cost of chlorophyll meter, farmers have tendency to restrict its wide spread in developing countries. Now a day's leaf colour chart is ideal to farmers as a visual and subjective indicator of the crop nitrogen status and right time application of nitrogen fertilizer (Ladha *et al.*, 2000) [8]. LCC proves to be an effective tool for detecting requirement of additional quantity of nitrogen to maize for higher yields and increased profit compared with fixed nitrogen levels (Pasuquin *et al.*, 2012) [11]. Keeping the foregoing fact in view, a field experiment was conducted to study the "Real time nitrogen management in *rabi* maize (*Zea mays* L.) using leaf colour chart under varied plant density".

Materials and Methods

A field experiment was carried out at the Wet land farm of Sri Venkateswara Agricultural College, Tirupati (13.6°N, 79.3°E and 182.9 m above mean sea level) during *Rabi* season of 2017-18 to study the response of maize hybrid to different stand density and nitrogen levels using leaf colour chart. The experimental soil was sandy clay loam, alkaline in reaction (pH 7.9) with low organic carbon (0.28%), low available nitrogen (N) (183 kg ha^{-1}), medium available phosphorus (P) (25.3 kg ha^{-1}) and medium available potash (K) (185 kg ha^{-1}). A total amount of rainfall during crop period of 43.3 mm was received in 2 rainy days. Experiment was laid out in Split-Split plot design with three replications. The treatment comprise three planting densities (66,666, 83,333 and 1,11,111 lakhs ha^{-1}) as main plots, three nitrogen levels (30, 35 and 40 kg N ha^{-1}) as sub plots and three LCC threshold values (LCC 4, 4.5 and 5) as sub-sub plots.

Maize crop (Hybrid-DHM-117) was sown on 16th November 2017 using three different spacing *viz.*, 75 cm x 20 cm, 60cm x 20 cm and 45 cm x 20 cm. The recommended dose of 80:80 kg P_2O_5 and K_2O ha^{-1} through SSP and MOP were applied to the soil as basal along with 60 kg N ha^{-1} through urea, in all the treatments. However, remaining 180 kg urea was applied based on leaf colour chart at different splits when it falls below the LCC threshold levels at every 10 days interval from 21 days after sowing to silk emerging stage *i.e.*, 21, 31, 41, 51 and 61DAS. A 'six panel' LCC was used to match leaf colour in ten plants from each net plot. Observations were taken for 10 plants by placing the middle part of the youngest fully expanded and healthy leaf on the top of the colour strips in the chart. When six or more leaves read below a set critical value (4, 4.5 and 5), urea was applied as per the treatment. LCC readings were taken at same time of the day (8:00-11:00 AM). However, during observation LCC was not exposed directly to sunlight and leaf being measured was shielded from the sun. Based on LCC total quantity of N applied in different treatments and total quantity of N saved in each treatment combination was given in Table 1. Planting of maize at lower density required lower amount of nitrogen (157.8 kg N ha^{-1}) in which N saving was 82.2 kg N ha^{-1}. Whereas at higher plant densities total quantity of N applied and N saved was 182.6 and 87.4 kg N ha^{-1}, respectively. Among the different combinations of treatment, total amount of N was applied at higher rate (219.9 kg N ha^{-1}) to $P_3N_3L_3$, however lower amount of N was applied (140 kg N ha^{-1}) at $P_1N_1L_1$ by using LCC over 240 kg N ha^{-1} (recommended). Crop was grown under fully irrigated conditions and two hand weedings were done at 20 and 40 days after sowing to minimize crop- weed competition. There was no serious incidence of any insect-pest and disease during the experimental period.

Data pertaining to growth attributes and yield of five randomly selected plants was recorded from each net plot. The initial plant count after thinning was recorded by counting all the plants from each net plot at 14 days after sowing and final plant count was recorded by counting all the plants from net plot at the time of harvest and computed on hectare basis. Leaf area of crop was measured by using LI-COR model LI-300 portable leaf area meter. The SPAD-502 (Soil and plant analysis development) chlorophyll meter developed from Minolta (Minolta Ltd, Tokyo, Japan) was used for measuring leaf chlorophyll at middle lamina of the third leaf from the top of the plant without destructing leaves.

Table 1: Quantity of fertilizer N applied and saved in various treatments based on LCC

Total quantity of fertilizer N applied and saved (kg ha^{-1})							
		L_1	L_2	L_3	Mean for each treatment combination	Mean N applied	Mean N saved
P_1	N_1	140	150	160	150.0	157.8	82.2
	N_2	141.5	153.2	176.5	157.0		
	N_3	153.2	166.6	179.8	166.5		
P_2	N_1	160	170	180	170	172.6	67.4
	N_2	164.8	164.9	188.2	172.6		
	N_3	166.4	179.4	179.9	175.2		
P_3	N_1	160	170	190	173.3	182.6	57.4
	N_2	166	176.5	188.6	177.0		
	N_3	179.7	193.1	219.9	197.5		
Mean for L		159.0	169.3	184.7			

Results and Discussion

The effect of various plant densities, on initial and final plant population of maize was significantly influenced during the investigation, while interactions were not statistically traceable (Table 2). However, initial and final plant population was maximum when maize crop was sown with closer spacing which accommodate higher plant density *i.e.*, P_3 (1, 11, 111 plants ha^{-1}), which was significantly superior to other plant densities *viz.*, P_2 (83,333 plants ha^{-1}) and P_1 (66,666 plants ha^{-1}). Maximum plant population was recorded at higher plant density *i.e.*, P_3 (1, 11, 111 plants ha^{-1} which might be due to higher number of seeds sown in that treatment during sowing time. These findings are in conformity with the results of Achiri *et al.*, (2019) [1]. A good and uniform emergence is the basic requirement for good plant establishment that is required for the successful raising of crop, which ultimately determines the crop yield. Germination is dependent on moisture absorption by the seed and physiological processes. Nitrogen levels and LCC threshold did not exert a significant influence on initial and final plant population of maize. However, maximum numbers of plants were recorded at initial and final crop growth stages with application of higher nitrogen 40 kg N ha^{-1} (N_3) using higher threshold value of LCC-5 (L_3). These findings corroborate with the results of Amandeep (2016) [3].

Table 2: Initial and final plant population of maize as influenced by plant density, nitrogen levels and LCC threshold value

		Initial plant population					Final plant population				
		L_1	L_2	L_3	Mean for P	Mean for N	L_1	L_2	L_3	Mean for P	Mean for N
P_1	N_1	64646	63838	63030	63021	80995	60783	61590	62196	60963	79312
	N_2	61616	61131	62828			60784	61390	61995		
	N_3	62424	64444	63232			59169	59169	59169		
P_2	N_1	76091	76454	76205	76618	81035	73320	75758	75757	73809	78876
	N_2	77499	74538	77499			73319	73668	74016		
	N_3	77151	77499	76628			73493	72971	72623		
P_3	N_1	101010	103153	104530	103858	81468	100857	102234	101316	102115	78770
	N_2	107438	102387	104377			101469	101010	102234		
	N_3	104433	104245	103153			102999	103153	103765		
Mean for L		81368	80854	81276			78967	78729	79263		

	Initial plant population		Final plant population	
	SEm ±	CD (P = 0.05)	SEm ±	CD (P = 0.05)
P	441	1721	174	680
N	445	NS	602	NS
L	446	NS	22	NS
P x N	771	NS	1043	NS
P x L	772	NS	383	NS
N x L	772	NS	383	NS
P x N x L	1338	NS	664	NS

Generally, optimum plant density is a prerequisite for proper utilization of natural resources. The higher density of planting P_3 (1,11,111 plants ha^{-1}) with the closer spacing of 45 cm x 20 cm produced significantly taller plants (219.8 cm) and it was however on par with P_2 (83,333 plants ha^{-1}) with a spacing of 60 cm x 20 cm (Table 3). This might be owing to intense competition for light at higher plant density in the plant community would compel the plants to grow taller, in search of radiant energy. Obviously, the plants with lower densities P_1 (66,666 plants ha^{-1}) do not enter into such competition for light and hence, grew normally. Similar findings are reported earlier by Fromme *et al.*, (2019) [5]. Increasing plant height with each successive increment of nitrogen might be attributed to the fact that nitrogen as integral part of chlorophyll, protiens, the building blocks of plants helps in maintaining higher auxin levels which might have resulted in better plant height. The present findings corroborate with the findings of Ahmad *et al.* (2018) [2] and Zothanmawii *et al.* (2018) [18].

Table 3: Plant height and leaf area per plant of maize as influenced by plant density, nitrogen levels and LCC threshold value

		Plant height (cm)					leaf area plant^{-1} (cm^2)				
		L_1	L_2	L_3	Mean for P	Mean for N	L_1	L_2	L_3	Mean for P	Mean for N
P_1	N_1	198.0	207.5	214.9	212.7	212.7	3778	3893	4059	3992	3788
	N_2	206.6	207.9	221.2			3823	3981	4168		
	N_3	209.5	214.7	234.1			3866	4088	4284		
P_2	N_1	211.8	216.6	217.6	217.6	216.3	3577	3637	3795	3839	3856
	N_2	213.6	218.6	223.3			3796	3904	3963		
	N_3	209.7	219.3	228.0			3928	3969	3984		
P_3	N_1	209.2	216.4	218.8	219.8	221.1	3580	3741	3751	3795	3981
	N_2	212.5	218.2	227.4			3713	3804	3807		
	N_3	218.8	227.3	229.4			3843	3921	4001		
Mean for L		210.0	216.5	223.6			3767	3882	3978		

	Plant height (cm)		leaf area plant^{-1} (cm^2)	
	SEm ±	CD (P = 0.05)	SEm ±	CD (P = 0.05)
P	0.9	3.5	20.7	81.02
N	1.9	5.8	35.8	110.3
L	1.5	4.4	27.9	80.2
P x N	3.2	NS	62.0	NS
P x L	2.6	NS	48.4	NS
N x L	2.6	NS	48.4	NS
P x N x L	4.6	NS	83.9	NS

The need based N fertilizer applications at five splits using higher LCC threshold (L_3) along with 60 kg basal application of N produced significantly maximum plant height (223.6 cm), it might be attributed to the increased level of N fertilization with LCC-5 treatment and it was at par with L_2 (LCC-4.5). The level of N, encourage the carbohydrate synthesis that resulted in the taller plants at higher LCC value (L_3). Similarly, this result was in accordance with Kumar *et al.* (2018) [7].

The results of field experiment revealed that leaf area plant^{-1} decreased with increasing plant density of maize as shown in Table 3. Similar results have been documented by Ahmad *et al.*, (2018) [2]. Pertaining to various nitrogen levels leaf area plant^{-1} increased with the application of nitrogen at higher rate *i.e.*, 40 kg N ha^{-1} at different splits. The number of green leaves per plant increased with higher rate of nitrogen application which lead to higher leaf area. Finally, leaf area size determine light interception in a crop canopy and effects overall photosynthesis and yield of maize. The findings are in line with those of Ullah *et al.* (2015) [16]. Experimental results disclosed that crop in terms of leaf area plant^{-1} respond more when N was applied at LCC threshold 5>4.5>4. Top dressing of N fertilizer as guided with LCC lead to increased leaf area plant^{-1} in LCC-5 (L_3) which might be due to continuous split application of N till flowering to maintain sufficient N content (dark green colour) might have helped the plant to put forth higher leaf area as evidenced through LCC-5 value. The same results were also quoted by Kumar *et al.* (2018) [7].

A close glance of data shows that the leaf greenness (measured by SPAD meter) of maize decreased with higher stand density *viz.*, 1, 11, 111 plants ha^{-1} (Table 4). Generally, higher density of crop decreased leaf greenness and higher green leaf retention was noticed at lower densities which might be due to increased leaf area and thickness due to optimum availability of resources for crop growth. The negative role of higher plant density on SPAD reading was also reported by Huang *et al.* (2017) [16]. However, increasing SPAD reading with each successive increment of nitrogen might be attributed to the fact that nitrogen is an integral part of chlorophyll, which converts light into chemical energy needed for photosynthesis, on adequate supply of nitrogen might result in high photosynthetic activity, vigorous vegetative growth and a dark green colour. The results are in conformity with the findings of Amandeep (2016) [3]. SPAD meter and LCC, both gadgets considered leaf greenness as indicator of leaf N concentration. Similarly, application of N with increase in LCC threshold value (L_3) exhibited significant increment in leaf greenness which might be attributed to the increased level of N fertilization. The results are in consonance with those obtained by Mathukia *et al.* (2014) [9].

Table 4: Chlorophyll (SPAD values) of maize as influenced by plant density, nitrogen levels and LCC threshold value

SPAD chlorophyll readings		
	SEm ±	**CD (P = 0.05)**
P	0.6	2.5
N	0.5	1.6
L	0.5	1.4
P x N	0.9	NS
P x L	0.8	NS
N x L	0.8	NS
P x N x L	1.5	NS

		SPAD chlorophyll readings				
		L_1	**L_2**	**L_3**	**Mean for P**	**Mean for N**
P_1	N_1	38.7	40.0	42.0	40.6	37.5
	N_2	38.8	40.2	42.0		
	N_3	38.9	42.0	42.9		
P_2	N_1	34.1	36.7	39.4	38.4	38.7
	N_2	36.2	37.4	40.0		
	N_3	38.2	41.5	41.7		
P_3	N_1	33.6	34.5	35.1	37.0	39.9
	N_2	36.4	38.7	39.5		
	N_3	37.3	38.8	39.3		
Mean for L		36.9	38.9	40.2		

The first pre-requisite for higher yield is higher production of total dry matter per unit area. Ultimately the significantly higher mean total dry matter accumulation (17583 kg ha^{-1}) was obtained (Table 5) at higher planting density P_3 (1, 11,111 plants ha^{-1}) compared to other stand densities *i.e.*, P_2 (83,333 plants ha^{-1}) and P_1 (66,666 plants ha^{-1}). Maximum number of plants per unit area coupled with more number of leaves per plant, resulted in accumulation of larger quantities of dry matter compared to the lower plant densities. Similar results have been documented by Revathi *et al.* (2017) [12]. Dry matter accrual of maize was increased with need based N management at different splits and found maximum at 40 kg N ha^{-1} (N_3) and it may be due to increased leaf area and photosynthetic rate which ultimately results in higher biomass production. The results obtained corroborate the findings of Amandeep, (2016) [3] and Zothanmawii *et al.* (2018) [18]. Need based application of N fertilizer at 0.5 unit increase in LCC score from LCC-4 to LCC-5 significantly increased dry matter accumulation in irrigated *rabi* maize, however it was maximum at LCC threshold-5 than LCC-4.5 (L_2) and LCC-4 (L_1). It can be due to adequate supply of nitrogen at different splits at all the stages of crop growth as evidenced by the nutrient supply and might be responsible for rapid growth in stem, leaves and cobs. Kumar *et al.* (2018) [7] also reported similar results. Interaction of plant density and nitrogen levels exerted significant influence during the experiment, while other interactions were not statistically traceable. Whereas, maximum dry matter accumulation was recorded at the combination of P_3N_3 (18660 kg ha^{-1}) and it was statistically superior than all the other combinations (Table 6).

Table 5: Total drymatter accrual and cob yield of maize as influenced by plant density, nitrogen levels and LCC threshold value

		Total drymatter accrual (kg ha^{-1})					Cob yield (kg ha^{-1})				
		L_1	L_2	L_3	Mean for P	Mean for N	L_1	L_2	L_3	Mean for P	Mean for N
P_1	N_1	12573	13093	13826	13615	14815	7277	7348	8590	8321	8208
	N_2	12877	13613	13828			8027	8621	9227		
	N_3	13533	14395	14795			8308	8818	8675		
P_2	N_1	14049	14694	15349	15016	15391	7488	8218	8876	8756	8991
	N_2	14577	15261	15418			8375	8649	8999		
	N_3	14694	15305	15794			8435	9855	9909		
P_3	N_1	15777	16955	17014	17583	16008	7937	8859	8876	9179	9058
	N_2	17022	17533	17970			8199	9153	9568		
	N_3	17711	18837	19433			9827	10004	10187		
Mean for L		14757	15520	15936			7277	7348	8590		

	Total drymatter accrual (kg ha^{-1})		Cob yield (kg ha^{-1})	
	SEm ±	CD (P = 0.05)	SEm ±	CD (P = 0.05)
P	91	354	97	380
N	80	246	59	181
L	138	426	76	219
P x N	117	238	101	313
P x L	144	NS	132	NS
N x L	144	NS	132	NS
P x N x L	249	NS	230	NS

Scrutiny of data pertaining to cob yield (Table 5) revealed that the higher stand density 1,11,111 plants ha^{-1} (P_3) which was significantly superior to other densities *viz.*, P_2 (83,333 pants ha^{-1}) and P_1 (66,666 plants ha^{-1}). This might be due to high stand densitie number of plants were more per unit area, which leads to more cob yield than other densities and similar results were also reported by Thakur *et al.*, (2015) [15]. Nitrogen is a critical input for higher cob production and it was obtained with application of higher nitrogen level @ 40 kg N ha^{-1} (N_3) and the next best nitrogen dose was 35 kg N ha^{-1} (N_2) followed by 30 kg N ha^{-1} (N_1). The improvement in cob yield with enhanced nitrogen application, owing to better availability and efficient uptake of nutrients which in turn lead to efficient metabolism and higher biomass accrual which might be responsible for production of higher cob yield. The results are in conformity with the findings of Thakur *et al.*, (2015) [15]. Significantly higher cob yield was obtained with higher threshold LCC-5 (L_3), over LCC-4.5 (L_2) and LCC-4 (L_1). Supply of nitrogen at higher LCC threshold-5 evidenced by the higher nutrient supply from source to sink is responsible for higher cob yield. Lower cob yield was found with application of nitrogen based on threshold LCC-4 may be attributed to stress of nitrogen at all the stages of growth leading to detrimental effect on growth and yield. These findings are in line with those of Swamy *et al* (2016) [14].

Table 6: Total drymatter accrual and cob yield as influenced by interaction of plant density and N levels

Total drymatter accrual					Cob yield			
	N_1	N_2	N_3	Mean	N_1	N_2	N_3	Mean
P_1	13165	13439	14241	13615	7738	8691	8535	8321
P_2	14698	15226	15124	15016	8235	9400	8633	8756
P_3	16583	17508	18660	17583	8650	8881	10006	9179
Mean	14815	15391	16009		8208	8991	9058	8207
SEm ±		138			SEm ±		105	
CD (P = 0.05)		426			CD (P = 0.05)		313	

Interaction of plant density and nitrogen levels exerted significant influence, while other interactions were not statistically traceable. Whereas, cob yield obtained varied from 7277 to 10187 kg ha^{-1} at various combinations, however P_3N_3 combination produced maximum cob yield (10187 kg ha^{-1}) and it was statistically superior to all other combinations (Table 6).

Conclusions

Plant population and nitrogen management had profound effect on growth, development and yield of maize. Application of nitrogen with blanket recommendation at fixed time is not adequate for obtaining higher crop growth and yield in maize. Whereas, need-based N fertilizer application through LCC saved 57.4 kg N ha^{-1} an average at higher density of maize and 82.2 kg N ha^{-1} at lower planting density. LCC being an inexpensive, simple and farmer's friendly tool and it can be used even by small and illiterate farmers. Experimental results revealed that maize crop produced maximum growth attributes and yield when it sown at a spacing of 75 cm x 20 cm, which accommodate 1,11,111 plants ha^{-1} along with application of 40 kg N ha^{-1} whenever LCC threshold readings falls below 5 (LCC-5).

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