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NITROGEN STIMULATES PHENOLOGICAL TRAITS, GROWTH AND GROWING DEGREE DAYS OF MAIZE

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Field experiments were conducted during 2009 and 2010 to evaluate the effects of nitrogen (N) application timings and rates on phenology of autumn sown maize under semi-arid climatic conditions of Faisalabad, Pakistan. Plant development, growth and yield components were optimized by the N application in three splits; 1/3rd N at V2, 1/3rd N at V16 and 1/3rd N at R1 stages at the rate of 250 kg ha^{-1}. At this rate, the crop achieved more calendar days and thermal time in each growth stage. The treatments T_2 (1/3rd N at V2 stage, 1/3rd N at V16 stage and 1/3rd N at R1 stage) and N_4 (250 kg N ha^{-1}) accumulated the maximum days to silking and maturity (51 and 102 days, respectively), which resulted in the maximum crop growth rate and grain yield (8.38 t ha^{-1}). The highest net benefit and marginal rate of return ($1857 and 22%, respectively) were achieved by N_4 treatment. Therefore, 250 kg N ha^{-1} with three above mentioned splits of N application may produce optimum grain yield of maize under semi-arid environmental and agricultural conditions similar to those of the reported experiments.
Keywords: Crop phenology, economic and marginal analysis, maize growth and yield, nitrogen

INTRODUCTION

Environmental and soil factors such as atmospheric CO_2 concentration, air temperature, precipitation and nutrient availability influence crop development and phenology. Higher temperature, elevated CO_2 concentration and variable precipitation are among serious problems for agricultural production (Dhungana *et al*., 2006; Walker and Schulze, 2008). These changes could strongly affect the physiological processes in plants such as photosynthesis, respiration, partitioning of photosynthesis production and ultimately crop calendar days (Chartzoulakis and Psarras, 2005; Yang and Zhang, 2006). Along with the mean temperature increase, the occurring frequency of extreme temperature may increase, that may shortly affect the crop phenology (Korner *et al*., 2002; Wu *et al*., 2006).

Low nutrient availability is one of the major factors affect crop growth and development (Li *et al*., 2001). Nitrogen has become a very expensive input for maize production in developing countries (Khan *et al*., 2013). In most maize growing areas of Pakistan, inadequate nutrient management at critical crop growth stages adversely affects crop phenology thus limiting maize growth and yield to a lower level. Moser *et al*. (2006) and Grant *et al*. (2002) attributed lower yield of maize to high dose of nitrogen. Additionally, over N fertilization is also a common problem in some agriculture areas for the wheat-maize rotation system (Zhao *et al*., 2006).

Time of N application can improve N use efficiency and protect soil environments. Similarly, deficiency of N is evident in the decrease of light interception by decreasing crop growth, which results in lower grain yield (Aslam *et al*., 2003; Hammad *et al*., 2011a; Maqsood and Shehzad, 2013). Application of optimum N at critical stage can be considered the most important factor in improving crop productivity (Magdoff, 1991). Judicious N management practices do not only optimize grain yield but also reduces the potential N leaching beyond the root zone of the crop (Subedi and Ma, 2005; Yousra *et al*., 2013) and N_2O emission which resulting in safe soil environments.

Shortage of N at early and late growth stages can reduce N uptake. It seems that maize which highly deficient to N would be able to respond the N applied late in the growing season (Cheema *et al*., 2010). There is little data available to indicate how soil N status in early stages of a growing season affect maize response to delay N applications (Darren *et al*., 2000) and how maize response to N when it applied at vegetative stage (Jokela and Randall, 1989). Similarly, Amanullah *et al*. (2009) reported nutrient loss in form of leaching and volatilization in case of application of N fertilizer only at sowing time in one or two splits.

Crop phenology frequently changes as soil nutrients become limiting, but such responses are poorly understood and difficult to quantify without comprehensive research trials. K influences water use efficiency (Quampah *et al*. 2011). In addition to their importance for crop yield, as nutrient availability affects crop phenology (Twine *et al*., 2004). Similarly, crop growing degree days and thermal time are also very important factors that influence crop phenology. Many approaches have been developed for calculating crop growing degree days and thermal time. Many Scientists have tested the accuracy of various forms of the basic growing degree day's equation in predicting various growth and development processes in several crop species.

In the production economics of crop agriculture is a major concern is that a cop responses to inputs at optimum level beyond which the crop does not response to the inputs rather additional inputs might be harmful for the crop (Hennessy, 2009). The effectiveness of any production system is ultimately evaluated on the basis of its economic returns. Economic analysis is the simplest way for determining the highest net benefits of treatments. Marginal analysis shows marginal rates of return of different treatments. Proceeding in steps from the least costly treatment to the most expensive one helps in deciding suitable treatments to the farmers (Byerlee, 1988). The questions arise from the above discussion that: 1) How to manage N application? and 2) How much N fertilizer be applied? The both factors influence crop phenology and ultimately grain yield. The experiments reported here were designed to answer the above questions.

MATERIALS AND METHODS

***Experimental site*:** The reported experiments were conducted at the Agronomic Research Farm of the University of Agriculture Faisalabad, Pakistan (31˚ 25´ N, 73˚ 04´ E) in maize growing seasons autumn during the year 2009 and 2010. Weather data including daily maximum and minimum air temperatures and rain fall were collected from the automated weather station operating at a distance of about one kilometer from the experimental site. Comparatively higher rainfall was observed during the year 2010 than during 2009 (Fig. 1). Similar conditions with respect to maximum and minimum temperatures were recorded at early crop growth stages during both the years i.e. 2009 and 2010.

***Experimental design and treatments*:** Hybrid maize (Pioneer 31-R-88) was sown on August 1, 2009 and August 2, 2010 in split plot arrangements keeping time of N application in the main plot and N rate (100, 150, 200, 250 and 300 kg ha^{-1}) in sub-plot replicated three times. Individual plot size was 15 m^2 with plant population of 7 plants per m^2. Phosphorus (Triple super phosphate) and potassium (Muriate of Potash) each at the rate of 125 kg ha^{-1} were manually broadcasted at the time of sowing. Nitrogen in the form of Urea (46% N) was applied at three splits i.e. T_1 (1/3rd N at seed bed preparation, 1/3rd N at V6 and 1/3rd N at VT stage), T_2 (1/3rd at V2 stage, 1/3rd at V16 and 1/3rd N at R1 stage) and T_3 (1/3rd at seed bed preparation, 1/3rd N at V12 and 1/33rd N at R2 stage) where Vn, stand for V = Vegetative, n = Number of leaves R = Reproductive stage T = Tasseling, R1 = Silking, R2 = Blister.

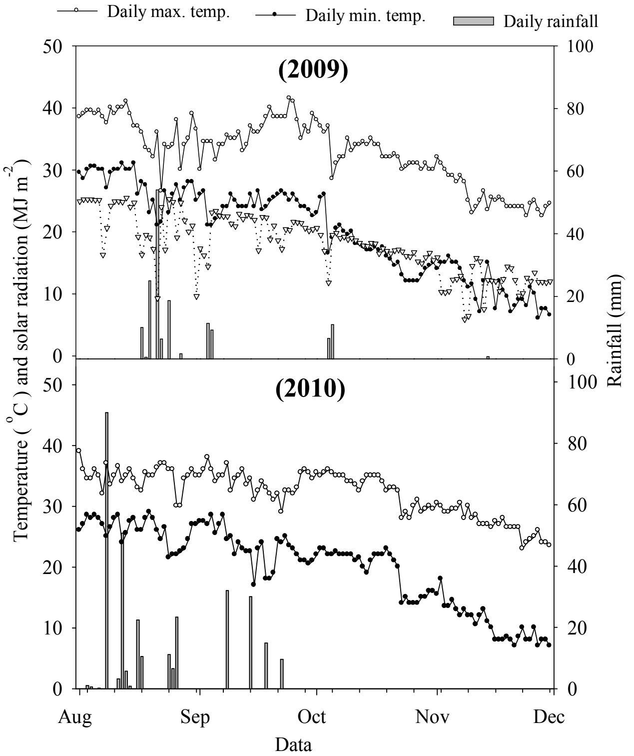

Figure 1. Weather summary of experimental site during growing seasons 2009 and 2010

Irrigation was applied at 30% depletion of total available of soil moisture (TAW) in 0-30 cm soil profile. All other agronomic practices such as earthing up, hoeing, and plant protection measures were kept alike for each treatment. The crop was harvested manually at physiological maturity.

***Soil analysis*:** Initial soil sampling was carried out from each experimental block prior to seed sowing. The soil samples collected from 25 cm depth were analyzed for following soil physical and chemical properties.

The composition of primary soil particles (clay, silt and sand) were determined by their settling rates in an aqueous solution using the hydrometer method. Soil total nitrogen was determined using the micro Kjeldahl distillation and titration method (Bremner and Mulvaney, 1982). Available Phosphorus outlined by Spectrophotometer and exchangeable K was determined using the flame photometer. The properties of the soil are summarized in Table 1.

Table 1. Soil physical and chemical properties of the experimental field (0 - 25 cm)

Soil characteristic	2009	2010
Organic matter (%)	0.91	0.94
Soil pH	7.61	7.57
Nitrogen (g kg^{-1})	0.63	0.66
Phosphorous	6.78	6.86
Potassium	19.48	19.12
Sand (%)	60	59
Silt	16	17
Clay	24	24

Crop phenology: Daily observations of seedling emergence were recorded by in the two inner rows of each plot, starting from two days after sowing. For other phenological observations, 10 plants per plot were randomly tagged and observations were made on daily basis in order to distinguish crop growth stages. Thermal time and growing degree days were calculated as suggested by Gallagher *et al*. (1983) as:

$$Tt = \frac{\Sigma(T\max + T\min)}{2} - Tb$$

Tt is thermal time, *T*max is maximum temperature of a day, *T*min is minimum temperature of a day and *Tb* is base temperature taken as 8 ˚C for maize (O'Callaghan, 1994).

***Plant Sampling*:** One-third of each plot was specified for plant sampling to monitor crop phenology development and growth; the remaining portion of the plots was kept for final harvesting. Five plants were randomly selected in each plot, harvested to the ground level at 14 days intervals. Fresh weight of each plant fraction (stem, leaf, cob) was determined immediately after sampling. For dry weight, the samples were oven dried at 70˚C till constant weight. Crop growth rate (CGR) was calculated as (Hunt, 1978):

$$CGR = \frac{(W2 - W1)}{(t2 - t1)}$$

Where W_1 and W_2 were the total dry weights of harvested sample at times t_1 and t_2, respectively

***Economic and marginal analysis*:** Economic analysis was conducted using input and output prices for the year 2010. Net benefit was calculated as suggested by Byerlee (1988) as:

Net benefit = (Gross income – Variable cost)

Where gross income is the income without expenses and variable cost is that varies in treatments.

Marginal analysis:

Marginal analysis was preceded by using the net benefit and variable cost values. Final recommendations to farmers were given on the basis of marginal rates of return, which were calculated by following the procedure (Byerlee, 1988).

$$\text{Marginal Rate of Return (\%)} = \frac{\text{Change in net benefit}}{\text{Change in cost}} \times 100$$

***Statistical analysis*:** Statistical analysis was conducted by using the SAS (SAS institute 2004). When F-values were significant, the least significant difference (LSD) test was used for comparing treatments means. Response of yield and plant growth to N rates was analyzed by using polynomial contrasts within the analysis of variance structures.

RESULTS AND DISCUSSION

Homogeneity of the two years (2009 and 2010) data was tested by Bartlett's and Levene's test in SAS for phonological variables. Since the Levene's test resulted in non-significant homogeneity in the studied variables for both years, results of the both years are presented together.

The rate and timing of N application did not significantly affect the number of days to emergence in both growing seasons in all treatments (Table 2). This might be due to the newly grown plants that had adequate food reserves in form of cotyledons, which was available for initial plant growth. At initial stages, the plants do not require surplus N because some amount of N was present in the soil (Table 1), which can fulfill early N requirements of the newly emerged plant (Darren *et al*., 2000).

Nitrogen application timing significantly affected number of days to 50% tasseling. Treatment T_2 ($1/3^{rd}$ N at V2 stage, $1/3^{rd}$ N at V16 stage and $1/3^{rd}$ N at R1 stage) took more days to 50% tasseling (51 days) than the other treatments (Table 2). The crop accumulated the maximum number of days to 50% tasseling might be due to application of N fertilizer at critical stages of the crop. Nitrogen rates also showed highly significant effects on number of days to 50% tasseling and the effect of N application was linear. Nitrogen application at the rate of 250 kg ha^{-1} delayed days to 50% tasseling at 51 days, it was statistically similar with treatment N_5 (300 kg N ha^{-1}). Valero *et al*. (2005) concluded that maize crop accumulated 47 days for 50% tasseling when N was applied at the rate of 130 kg ha^{-1} under semiarid environment.

The days to 50% silking parameter was affected by timings as well as rates of N application (Table 2). When $1/3^{rd}$ N at V2 stage, $1/3^{rd}$ N at V16 stage and $1/3^{rd}$ N at R1 stage was applied then the crop achieved more days to 50% silking. Among various N application timings minimum days to 50% silking was observed in treatment T_3. In this treatment during silking stage, the crop might be faced N fertilizer stress. This might be the reason for early days to silking in crop. Various rates of N application significantly influenced days to 50% silking and the effect of N was quadratic. The number of days to 50% silking increased by increasing N dose up to 250 kg ha^{-1}. Beyond this level there was no significant increase. In this two years study it was observed during the both years luxury use of N increased days to silking in the treatments. Amanullah *et al*. (2009) reported that maize crop took 57 days for 50% silking when N fertilizer was applied at the rate of 180 kg ha^{-1} in three splits. These results are similar to those reported by Hammad

Table 2. Effect of nitrogen application timing and nitrogen rates on crop phenology and grain yield

Treatments	Days to emergence (day)	Days to 50 % tasseling (day)	Days to 50 % silking (day)	Days to maturity (day)	Crop growth rate (g m^{-2} d^{-1})	Grain yield (t ha^{-1})
2009	3	50	54	101	18.0	6.90
2010	3	50	55	101	18.1	7.01
Significance	ns	ns	ns	ns	ns	ns
T_1	3	50 b	55 a	100 c	18.6 a	7.00 ab
T_2	4	51 a	56 a	101 b	18.3 a	7.31 a
T_3	3	48 c	54 b	102 a	17.3 b	6.56 b
LSD 5%	0.49	0.65	1.34	0.30	0.68	0.46
Significance	ns	**	**	**	*	*
N_1	4	48 c	53 c	99 d	16.7 d	5.03 e
N_2	3	49 bc	54 bc	100 cd	17.4 c	6.24 d
N_3	4	50 ab	55 ab	101 bc	18.1 b	7.41 c
N_4	4	51 a	56 a	102 ab	19.0 a	8.38 a
N_5	3	50 ab	56 a	103 a	19.2 a	7.72 b
LSD 5%	0.82	1.75	1.50	1.5	0.60	0.35
Significance	ns	**	*	**	**	**
Linear effect	ns	*	**	**	**	**
Quadratic effect	ns	ns	*	ns	ns	**
Cubic effect	ns	ns	ns	ns	ns	*
T × N	ns	ns	ns	ns	ns	ns

Means with in the columns sharing various letters differ significantly at $P \leq 0.05$.
*, ** = Significant at 5 % and 1%, respectively, ns = Non-significant.
T_1 = Application of 1/3rd N at seed bed preparation, 1/3rd N at V6 and 1/3rd N at VT stages,
T_2 = 1/3rd N at V2, 1/3rd N at V16 and 1/3rd N at R1 stages and
T_3 = 1/3rd N at seed bed preparation, 1/3rd N at V12 and 1/3rd N at R2 stages.
N_1= 100, N_2= 150, N_3= 200, N_4= 250 and N_5= 300 kg ha^{-1}.

Table 3. Effect of nitrogen application timing and nitrogen rates on crop phenology

Crop Stages	Treat-ments	Calendar date		Calendar days		Thermal time (˚C days)*		T.T (˚C days)*
		2009	2010	2009	2010	2009	2010	Mean
Sowing	T_1	1-08-09	2-08-10	□	□	□	□	□
	T_2	1-08-09	2-08-10	□	□	□	□	□
	T_3	1-08-09	2-08-10	□	□	□	□	□
Sowing	T_1	3-08-09	4-09-10	3	3	79	71	75
to	T_2	3-08-09	4-09-10	3	3	79	71	75
Emergence	T_3	3-08-09	4-09-10	3	3	79	71	75
Emergence	T_1	19-09-09	20-09-10	47	47	1079	1023	1051
to	T_2	20-09-09	21-09-10	48	48	1103	1043	1071
50% Tasseling	T_3	17-09-09	20-09-10	45	47	1032	1023	1028
50% Tasseling	T_1	24-09-09	25-09-10	5	5	122	99	112
to	T_2	25-09-09	26-09-10	5	5	124	98	112
50% Silking	T_3	22-09-09	24-09-10	5	4	121	80	101
50% Silking	T_1	09-11-09	09-11-10	45	45	773	790	782
to	T_2	10-11-09	10-11-10	46	45	767	783	775
Crop maturity	T_3	10-11-09	10-11-10	49	47	842	822	832
Sowing	T_1	09-11-09	09-11-10	100	100	2053	1982	2018
to	T_2	10-11-09	10-11-10	102	101	2072	1994	2033
Crop maturity	T_3	10-11-09	10-11-10	102	101	2072	1994	2033

T_1 = Application of 1/3rd N at seed bed preparation, 1/3rd N at V6 stage and 1/3rd N at VT stages,
T_2 = 1/3rd N at V2 stage, 1/3rd N at V16 stage and 1/3rd N at R1 stages and
T_3 = 1/3rd N at seed bed preparation, 1/3rd N at V12 stage and 1/3rd N at R2 stages.
*Base temperature = 8 ˚C

(2012). Application of N at later stages of crop delayed crop days to maturity. Similarly, days to maturity of the crop was increased linearly by increasing N dose up to 300 kg ha^{-1}. Akbar *et al*. (2002) found that maize crop took 102 days to maturity when the crop was subjected to 200 kg N ha^{-1}. The Changes in the N application timing and increase in N fertilizer rate and might have enhanced the rate of photosynthesis which resulted in the leaf longevity and delayed tasseling, silking and maturity stage of the crop in maize (Gungula *et al*., 2003).

Crop development is presented in Table 3 indicating calendar days and thermal time (Growing degree days) of both seasons from sowing to emergence. The calendar days from sowing to emergence were similar (3 days) for all treatments which gave mean thermal time of 79 and 71 °C days during 2009 and 2010, respectively. During the year 2010 at sowing time air temperature was low as compared to 2009 due to rainfall (Fig. 1). This shows that autumn maize can germinate well if temperature decreases in August. Application of 1/3 N at seed bed preparation, 1/3 N at V6 stage and 1/3 N at VT stage (T_2) resulted in the maximum thermal time (1103 and 1043 °C day during 2009 and 2010, respectively) from emergence to tasseling stage. For both the years, calendar days were same (48 days) but due to high temperature during 2009, thermal time of the crop increased. Minimum thermal time (1023°C days) was recorded by application 1/3 N at seed bed preparation, 1/3 N at V12 and 1/3 N at R2 stage (T_3) and T_2 in 2010. Calendar days and thermal time for tasseling to silking stage showed little difference for all the treatments because difference between total numbers of days were small (4-5 days). However, for silking to maturity, clear difference was observed in the treatments. Application of N fertilizer at later stages of the crop increased calendar days and thermal time of the crop during both growing season. The results were supported by the findings of Amanullah *et al.* (2009) who reported that application of N fertilizer at later stages increased crop duration in maize (Mohsan, 1999). In general, maximum mean calendar days (102) and thermal time 2072 °C days were taken by the crop in treatment T_2 and T_3 during 2009. Same trend was observed in 2010. The crop matured 2 and 1 days early during 2009 and 2010, respectively when N fertilizer was applied at initial growth stages.

Crop growth rate was significantly affected by time of N application (Table 2). The CGR increased up to 62 days after sowing in all treatments during both growing seasons followed by a gradual decreased up to maturity (Figs. 2a and b). In both growing seasons, the maximum CGR was obtained by application of 1/3rd N at seed bed preparation stage, 1/3rd N at V6 and 1/3rd N at VT stages. The minimum

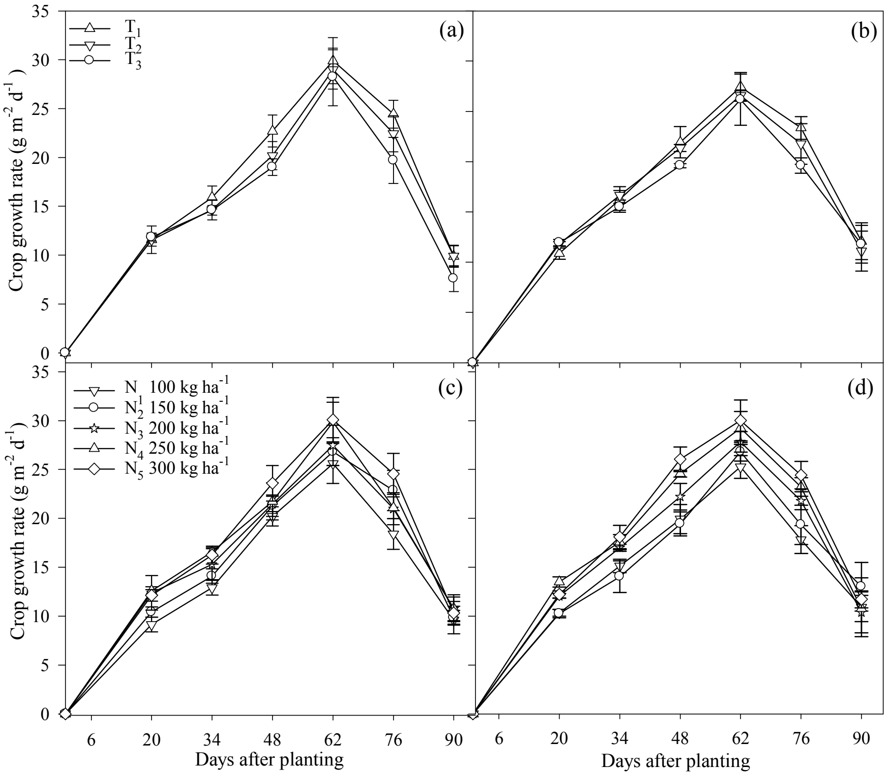

Figure 2. Changes in crop growth rate with time as affected by nitrogen timings and rates during 2009 and 2010; each bar represents standard deviation of three replicates.

Table 4. Economic analysis of nitrogen application timing and rates

Detail	N_1	N_2	N_3	N_4	N_5	Remarks
Total grain yield	5.03	6.24	7.40	8.38	7.72	t ha^{-1}
Adjusted yield	4.53	5.62	6.66	7.54	6.95	To bring at farmer's level (10% decrease)
Gross income	1245	1544	1832	2074	1911	Price of 1 t grain $ 275
Cost of N_1	87	—	—	—	—	Price of one bag urea (23 kg N) 20 $
Cost of N_2	—	130	—	—	—	□ do □
Cost of N_3	—	—	174	—	—	□ do □
Cost of N_4	—	—	—	217	—	□ do □
Cost of N_5	—	—	—	—	261	□ do □
Cost that vary	87	130	174	217	261	$ ha^{-1}
Net benefits	1158	1414	1658	1857	1650	$ ha^{-1}

Table 5. Marginal analysis of different nitrogen application timing and rates

Treatments	Cost that vary ($)	Net benefits ($)	Chang in variable cost ($)	Change in the net benefit ($)	Marginal rate of return (%)
N_1 = 100 kg N ha^{-1}	87	1158	–	–	–
N_2 = 150 kg N ha^{-1}	130	1414	43	256	17
N_3 = 200 kg N ha^{-1}	174	1658	44	243	18
N_4 = 250 kg N ha^{-1}	217	1857	43	200	22
N_5 = 300 kg N ha^{-1}	261	1650	217	D	D

D =Dominated due to less benefits than preceding treatments

CGR was observed by application of 1/3rd N at seed bed preparation stage, 1/3rd N at V12 and 1/3rd N at R2 stages (T_3). The lower CGR in this treatment was likely due to late application of 2nd and 3rd doses of N. Figure 2 illustrates that the rate of N application significantly affected CGR and there was linear effect of N fertilizer (Table 2). This concludes that CGR increased by increasing N dose up to 300 kg N ha^{-1} during both growing seasons. Nitrogen affects crop production by various mechanisms; for example, it accelerates formation of chlorophyll, which is a main part of plant cells resulting into enhanced cell counts and volume of leaves (Shanjani, 2003).

Grain yield was significantly influenced by various times and rate of N application. Application of 1/3rd N at V2 stage, 1/3rd N at V16 stage and 1/3rd N at R1 stage (Treatment T_2) resulted in the maximum (7.35 t ha^{-1}) grain yield. The treatment T_2 took maximum days from emergence to tasseling sowing to maturity and the crop accumulated the maximum thermal time. Therefore, this treatment has more time to accumulate nutrients from the soil and resulted more grain yield as compared to other. The treatment T_2 also accumulated maximum days to 50% tasseling and silking. These characteristics of the crop became helpful for increasing the yield. Freeman *et al*. (2007) stated that N management practices including, methods, time and rate of N application significantly influenced maize grain yield. The grain yield showed positive and significant association (R^2 = 0.63, R^2 = 0.78 during 2009 and 2010, respectively Fig. 3) with CGR.

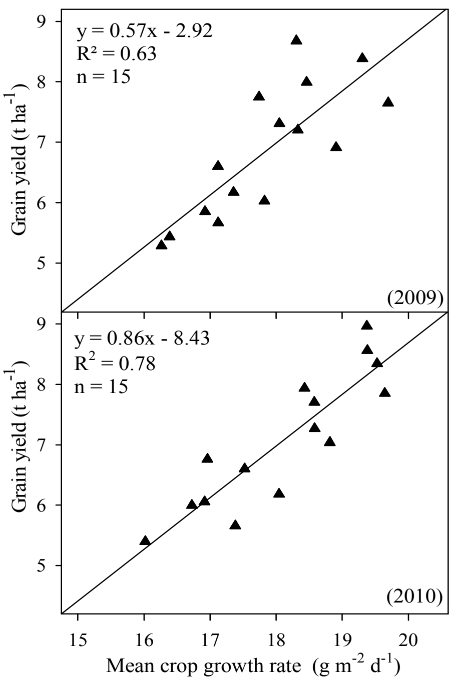

Figure 3. Relationship of grain yield with crop growth rate during 2009 and 2010.

Net benefit increased up to application of N at the rate of 250 kg N ha^{-1} (N_4) (Table 4). Beyond this level, reduction in net benefit was observed. The highest net benefit ($1857) was achieved by application of N_4 input. The maximum marginal rate of return (22%) was obtained by N_4 input (Table 5). Further increase in N input was dominated due to less benefit than proceeding treatment. These results were supported by the finding of Farhad *et al.* (2011a,b) who compared various rates of N from different sources and concluded that application of 250 kg ha^{-1} from synthetic fertilizer gave maximum marginal rate of return as compared to other rates and sources.

Conclusions: The crop growth and phenology significantly responded to time and rate of nitrogen fertilizer application. Application of nitrogen in three splits; 1/3rd at V2, 1/3rd at V16 and 11/3rd at R1 stages expedited plant growth and developments. Increased application of nitrogen fertilizer is not a sound strategy to achieve maximum yield. Optimum maize grain yield and development was achieved by the application of nitrogen up to 250 kg ha^{-1}. The maximum net benefit ($1857) and marginal rate of return (22%) was achieved by application of nitrogen fertilizer input at the rate of 250 kg ha^{-1}. Our findings suggest that farmers of semi-arid region should apply first dose of nitrogen fertilizer at first irrigation (at V2 stage) and the remaining two doses at V16 leaves and R1 stages each at the rate of 250 kg ha^{-1} in hybrid maize grown for silage or grain purpose.

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