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Effect of elevated CO_2 and temperature on thermal constants and lower threshold temperatures of maize aphid, *Rhopalosiphum maidis* (Fitch.) (Aphididae:Hemiptera) on maize, *Zea mays* (Linn.)

D.MOUNICA[1], M. SRINIVASA RAO[2], P.V. KRISHNAYYA[3], A.K. PATIBANDA[1] and V. SRINIVASA RAO[1]

[1]Department of Entomology, Agricultural College, Bapatla, ANGRAU, Guntur, Andhra Pradesh*
[2]Central Research Institute for Dryland Agriculture, Santhosh Nagar, Hyderabad, Telangana
[3]Agricultural College, Naira, ANGRAU, Srikakulam, Andhra Pradesh
Corresponding author: mounicaagri47@gmail.com

ABSTRACT

The objective of this study was to examine the development of corn leaf aphid,*Rhopalosiphum maidis* Fitch (Aphididae: Hemiptera) on maize *Zea mays* Linnaeus at elevated and ambient concentrations of CO_2 (550 and 380ppm ± 25 ppm, respectively) at six temperatures (20, 25, 27, 30, 33 and 35°C) and to estimate thermal constants and lower temperature thresholds for the forecasting models based on heat accumulation units which could be developed for use in forecasting. The duration of different growth stages of *R.maidis* were reduced with an increase of temperature from 20°C to 35°C under both ambient and elevated CO_2 conditions. The lower development threshold for first nymphal instar,second nymphal instar, third nymphal instar, fourth nymphal instar, adult duration and total development period required 10.1, 5.04, 13.42, 26.96, 10.9, 23.22 and 20.20°C under *e*CO_2 whereas it was 13.32, 9.41, 19.13, 30.48, 16.38, 22.88 and 20.89°C under *a*CO_2 conditions,respectively. The mean lower temperature threshold for nymph was slightly higher (16.38°C) at *a*CO_2 compared to that of *e*CO_2 (10.90°C) whereas for adult the mean lower temperature threshold was slightly higher (23.22°C) at *e*CO_2 compared to that of *a*CO_2 (22.88°C). The thermal requirement of *R. maidis* from first nymphal instar to adult (total development period) was found to be 100.00 degree days (DD) under *e*CO_2 conditions as against 111.11degree days under *a*CO_2 conditions. These estimated temperature thresholds and thermal constant can predict the pest scenarios and population dynamics of *R. maidis*.

Key words : *Rhopalosiphum maidis,* lower temperature threshold, thermal constant, degreedays

Maize aphid *Rhopalosiphum maidis* (Fitch.) (Aphididae: Hemiptera), a sap-sucking homopteran insect is the largest group of phloem feeding insect. It is a polyphagous species occurring worldwide on sorghum, barley and wheat besides maize (Smith and Boyko, 2007). It is now distributed worldwide in the tropics and warmer temperate regions (Blackman and Eastop, 2000). Species of aphids are diverse and ubiquitous but their importance among crops differs (Srinivasa Rao *et al*., 2016). Aphids cause mechanical harm and malnutrition to plants by the removal of phloem sap.

Agriculture is one of the most vulnerable sectors to the anticipated climate change with an adverse effect on crop yields. The increased levels of atmospheric CO_2 concentrations can have a direct effect on the growth rate of crop plants. Temperature has a direct influence on insect activity, rate of development and also plants. The predicted changes in temperature and CO_2 concentration affect the population dynamics and the status of insect pests of various crops. Plants with C_4 photosynthesis will respond little to rising atmospheric CO_2 because a mechanism to increase the concentration of CO_2 in leaves causes CO_2 saturation of photosynthesis at ambient conditions. Numerous studies have shown that the annual average temperature of the earth will increase 1°C by 2025 and the probable rise in temperature by the end of the century is expected to reach 3°C (Solomon *et al*., 2007). The parameters associated with life table are crucial for understanding population growth potential and for establishing effective management tactics to pest control in general because they provide information on development, reproduction and mortality of a pest population (Maia *et al*., 2000).

So far, there is no published report from India on the effect of *e*CO_2 and variable temperatures on *R. maidis*.

Hence, in the present study, life table parameters of *R. maidis* were examined at two levels of CO_2 and six different temperatures to estimate the temperature thresholds and thermal constants which would be useful in status of the pest populations.

MATERIALS AND METHODS

Maintenance of Rhopalosiphum maidis culture

The corn leaf aphids, *R. maidis* were collected from the field and maintained at an optimum temperature of 27 ± 1°C and 75 ± 5% RH in the insectary of Entomology section, CRIDA, Hyderabad. The nymphs and adults were reared individually in petridishes (110 X 10 mm) to obtain bulk population for experiments. Light intensity of 30, 000 Lx was provided by 26 W florescent bulb inside the chambers during the 14 hours light period with a relative humidity of 60 % (day) and 70 % (night). Light illumination is provided through fluorescent lamps horizontally mounted in pairs above each shelf. Air circulation inside the chamber was maintained from a specifically designed air diffuser. The period of light, CO_2 concentrations and temperature levels were automatically monitored and controlled using Intellus Ultra Controller. The maize plants (DHM-117) and insects were maintained in open top chambers (OTC) and CO_2 growth chambers under respective set conditions at elevated and ambient concentrations of CO_2 (550 and 380ppm ± 25 ppm, respectively) at six temperatures of 20, 25, 27, 30, 33 and 35±1°C. Fully grown foliage (30 days after sowing) obtained from respective treatments was used for feeding trials and leaf quality analysis. The crop was maintained at insecticide free condition throughout the experiment to understand the impact of eCO_2 and temperature on insect pests. Experiments on life-tables of *R. maidis* were conducted by adopting cut leaf method at elevated and ambient concentrations of CO_2 (550 and 380ppm ±25 ppm, respectively) at six temperatures of 20, 25, 27, 30, 33 and 35±1°C and a photoperiod of 14L:10D. The cutcorn leaf (6x6 cm) from the top of corn seedlings of 1-2 months old was detached and placed in a petridish with a moist cotton swab on one side of the leaf margin. The cotton swabs were moistened daily to keep the leaf fresh and the leaves were changed on alternate days (Srinivasa Rao *et al.*, 2013). In order to construct life-tables, newly hatched 50 first instar nymphs were collected carefully from the stock culture with the help of wet camel hair brush and transferred individually into each petridish containing maize leaves obtained from respective set conditions with 50 replications per each treatment. The data on durations of first instar, second instar, third instar, fourth instar, adult and total development period at each temperature under eCO_2 and aCO_2 conditions were recorded.

Calculation of lower temperature thresholds and thermal constants

Quantification of the relationship between insect development and temperature is useful to predict the seasonal occurrence and population dynamics of the insects. The degree day model (thermal summation model) was used to estimate the linear relationship between temperature and the rate of development of insect pests (Campbell *et al.*, 1974). The reciprocal of developmental period for each stage was calculated to obtain the development rate (1/day) at each temperature. Development threshold and thermal constant were determined by regressing development rate on temperature. Thermal constant is estimated by the formula Thermal constant (K) = (T – T0) x D;

where, where, T - Temperature at which insect species is reared,

T0 - Development threshold temperature,

D - Duration of development,

K was estimated as reciprocal of regression coefficient (b) between development rate and temperature. K = 1/ b.

To was determined as ratio of regression intercept 'a' and 'b' T0 = -a /b. The life table parameters were estimated for eCO_2 and aCO_2 separately at abovementioned six temperatures and were plotted against the tested temperatures to compare the lower threshold temperatures and thermal constant under both eCO_2 and aCO_2 conditions.

RESULTS AND DISCUSSION

The durations of different growth stages *R. maidis* were significantly affected by eCO_2 and temperature conditions. The results pertaining to the variation in durations of first instar, second instar, third instar, fourth instar, adult and total development periods of *R. maidis* on maize at elevated and ambient concentrations of CO_2 (550 and 380ppm ± 25 ppm, respectively) at six different temperatures of 20, 25, 27, 30, 33 and 35±1°C are presented inTable 1.

The first nymphal instar duration (3.00 to 1.00 days), second nymphal instar (1.92 to1.00 days), third nymphal instar (1.08 to 1.00 days), fourth nymphal instar (1.00 day), adult duration (23.1 to 1.20 days) and total development

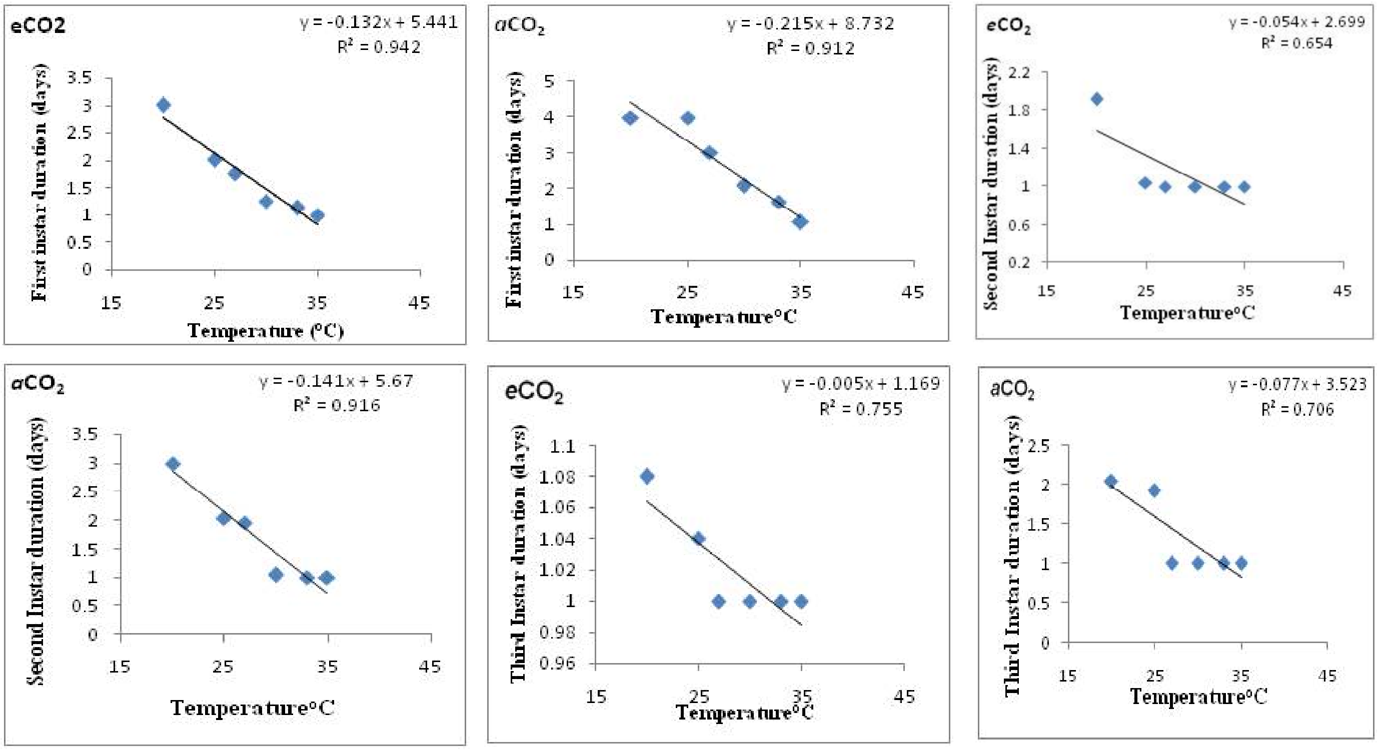

Fig. 1a: Effect of elevated CO_2 and temperatures on durations of first instar, second instar and third instar (days) of *R. maidis* on maize

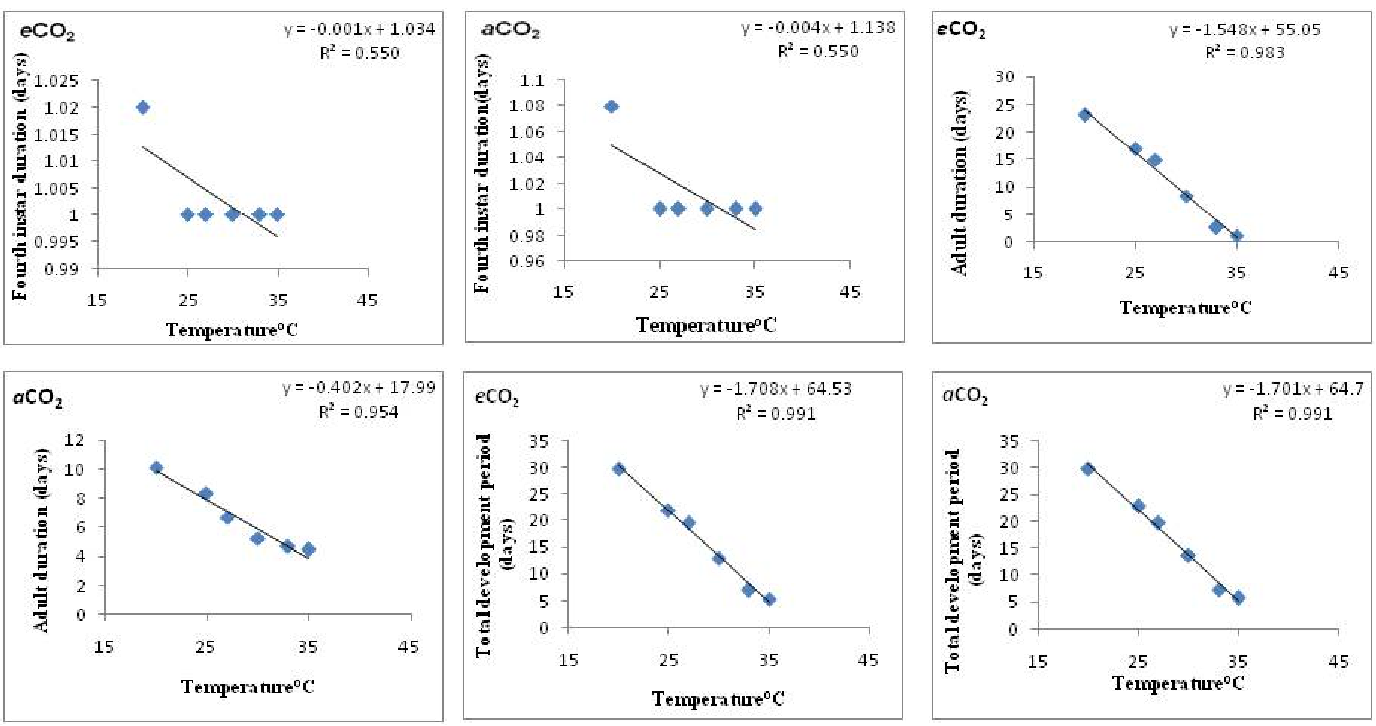

Fig 1b: Effect of elevated CO_2 and temperatures on fourth instar, adult duration and total development period (days) of *R. maidis* on maize

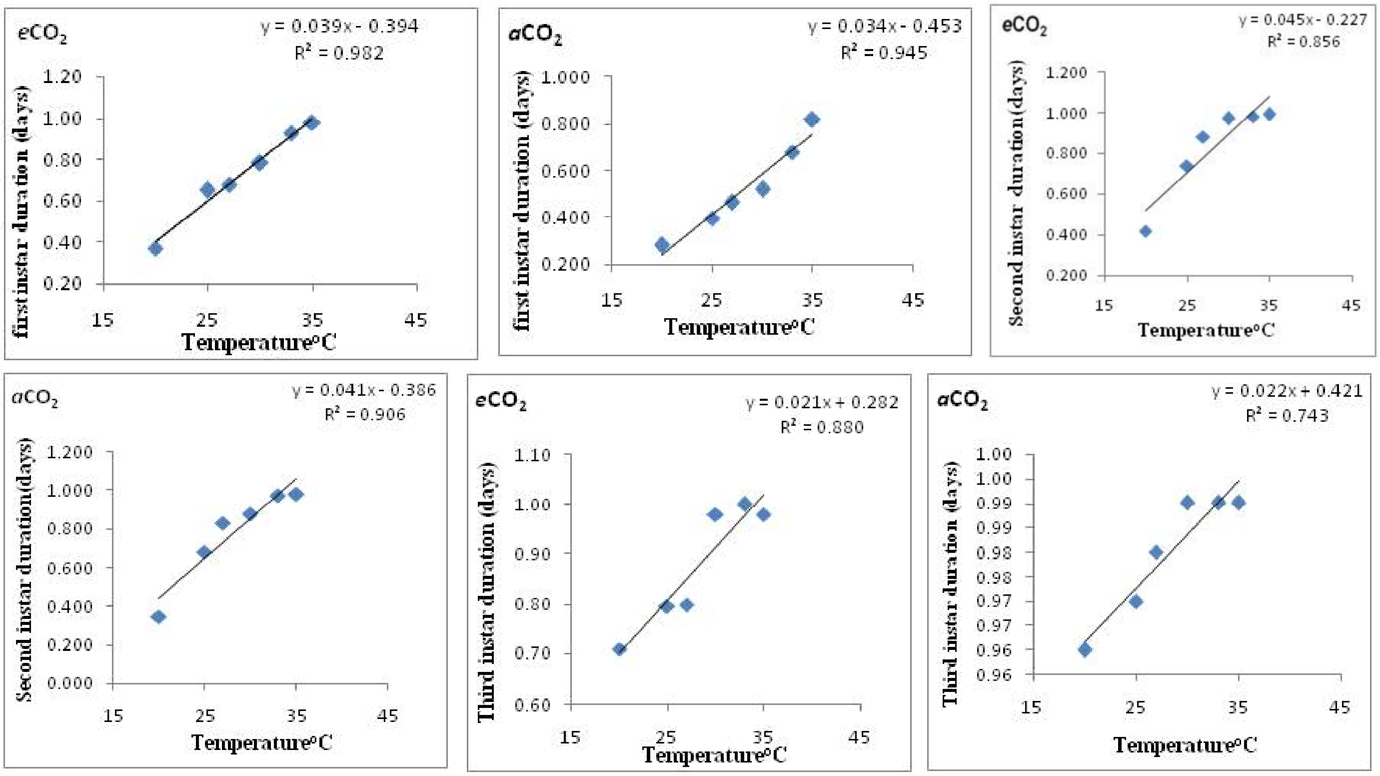

Fig.2a: Regression between temperatures and development rate to determine lower temperature threshold and thermal constant for durations of first instar, second instar and third instar duration (days) of *R. maidis* on maize under eCO_2 and aCO_2 conditions

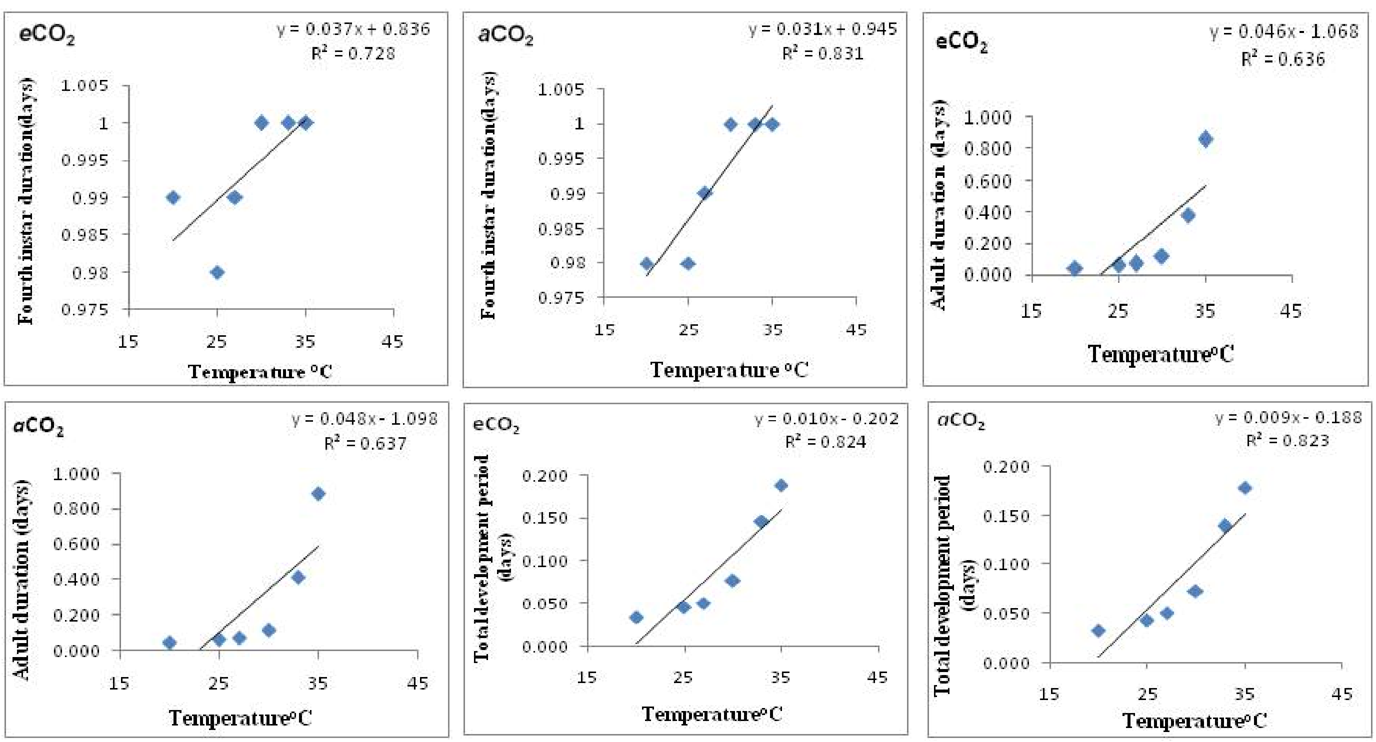

Fig. 2b: Regression between temperatures and development rate to determine lower temperature threshold and thermal constant for durations of fourth instar, adult duration and total development period (days) of *R. maidis* on maize

Table 1: Effect of elevated CO_2 and temperature on duration of different growth stages of *R. maidis* on maize

Temperatures (°C)	First instar duration (days)		Second instar duration (days)		Third instar duration (days)		Fourth instar duration (days)		Adult duration (days)		Total development period (days)	
	aCO_2 (380ppm)	*e*CO_2 (550ppm)	*a*CO_2 (380ppm)	*e*CO_2 (550ppm)	*a*CO_2 (380ppm)	*e*CO_2 (550ppm)	*a*CO_2 (380ppm)	*e*CO_2 (550ppm)	*a*CO_2 (380ppm)	*e*CO_2 (550ppm)	*a*CO_2 (380ppm)	*e*CO_2 (550ppm)
20	4.00±0.00	3.00±0.00	3.00±0.00	1.92±0.27	2.04±0.20	1.08±0.27	1.08±0.27	1.00±0.00	19.80±0.40	23.10±0.30	29.94±0.54	29.84±0.47
25	4.00±0.00	2.00±0.00	2.04±0.20	1.04±0.20	1.92±0.27	1.04±0.20	1.00±0.00	1.00±0.00	14.50±0.50	16.86±0.35	22.82±0.77	22.00±0.53
27	3.00±0.00	1.76±0.43	1.96±0.27	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	13.00±0.47	14.90±0.35	19.68±0.68	19.64±0.52
30	2.08±0.27	1.24±0.40	1.04±0.20	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	8.40±0.50	8.50±0.50	13.66±0.65	13.0±0.74
33	1.64±0.48	1.12±0.33	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	2.60±0.46	2.70±0.60	7.26±0.72	6.90±0.58
35	1.04±0.20	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	1.20±0.45	1.30±0.42	5.66±0.62	5.36±0.52
F test	80.44**		121.24**		145.51**		2.09**		242.73**		5.98**	
S.Em±	0.075		0.046		0.039		0.023		0.092		0.123	
LSD(p=0.05)	0.148		0.092		0.078		0.045		0.182		0.243	
LSD (p=0.01)	0.197		0.122		0.103		0.060		0.241		0.321	
CV(%)	12.30		11.9		11.88		7.94		4.27		3.81	
Factor 1 (CO_2)												
aCO_2 (380 ppm)	2.620		1.67		1.32		1.013		9.91		16.50	
eCO_2 (550 ppm)	1.680		1.18		1.02		1.000		11.21		16.12	
F test	965.88**		1059.87**		378.99**		2.09NS		959.34**		60.86**	
S.Em±	0.030		0.015		0.016		0.009		0.042		0.048	
LSD(p=0.05)	0.062		0.031		0.033		NS		0.084		0.097	
LSD (p=0.01)	0.085		0.042		0.044		NS		0.112		0.129	
CV(%)	12.18		9.20		11.63		7.94		4.85		3.62	
Factor 2 (Temperature(°C))												
20	3.50		2.46		1.56		1.04		21.45		29.89	
25	3.00		1.60		1.48		1.00		15.67		22.41	
27	2.36		1.48		1.00		1.00		13.94		19.66	
30	1.64		1.02		1.00		1.00		8.45		13.34	
33	1.38		1.00		1.00		1.00		2.63		7.08	
35	1.02		1.00		1.00		1.00		1.25		5.51	
F. test	675.21**		563.19**		187.06**		2.09**		30482.27**		22952.31**	
S.Em±	0.053		0.034		0.028		0.016		0.064		0.088	
LSD(p=0.05)	0.104		0.067		0.055		0.032		0.125		0.173	
LSD (p=0.01)	0.137		0.088		0.073		0.042‘		0.165		0.228	

All values are mean ± standard deviation;

** Significant @ 1% level of significance NS = Not-significant

Table 2: Effect of elevated CO_2 and temperature on lower temperature thresholds and thermal constants of each developmental stage of *R. maidis* on maize

Development stage	Regression equation		R^2 value		Lower temperature Threshold (T_0=a/b) (°C)		Thermal constant (K=1/b) (Degree Days-DD)	
	Ambient CO_2 (380 ppm)	Elevated CO_2 (550 ppm)	Ambient CO_2 (380 ppm)	Elevated CO_2 (550 ppm)	Ambient CO_2 (380 ppm)	Elevated CO_2 (550 ppm)	Ambient CO_2 (380 ppm)	Elevated CO_2 (550 ppm)
First instar	y = 0.034x - 0.453	y = 0.039x - 0.394	0.945	0.982	13.32	10.10	29.41	25.64
Second instar	y = 0.041x - 0.386	y = 0.045x - 0.227	0.906	0.856	9.41	5.04	24.39	22.22
Third instar	y = 0.022x + 0.421	y = 0.021x + 0.282	0.743	0.880	19.13	13.42	45.45	41.66
Fourth instar	y = 0.031x + 0.945	y = 0.037x + 0.836	0.831	0.728	30.48	26.96	32.25	27.02
Nymphal duration	y = 0.008x + 0.131	y = 0.009x – 0.098	0.962	0.966	16.38	10.9	125.00	111.11
Adult	y = 0.046x – 1.068	y = 0.048x – 1.098	0.637	0.636	22.88	23.22	20.83	21.74
Total development period	y = 0.009x – 0.188	y = 0.010x – 0.202	0.823	0.824	20.89	20.20	111.11	100.00

period (29.84 to 5.36 days) were recorded under eCO_2 conditions. Similar trend of reduction of the first nymphal instar duration (4.00 to1.04 days), second nymphal instar (3.00 to 1.00 days), third nymphal instar (2.04 to 1.00days), fourth nymphal instar (1.08 to 1.00 days), adult duration (19.8 to 1.30 days) and total development period (29.94 to 5.66 days) were recorded under aCO_2 conditions (Fig 1a &1b). The total development period was decreased with increasing temperature from 20 to 35°C under both eCO_2 and aCO_2 conditions. The present results are in conformity with the findings of Kuo *et al*. (2006), who reported that the duration of total immature stages of *R. maidis* from birth to adult decreased as temperature increased from 6°C (51.7 days) to 30°C (5.2 days). The total immature stages of *R. maidis* from birth to adult fed on barley and maize foliage, the estimated low developmental threshold (4.44°C and 6.1°C) and the shortest developmental time at 26°C and 30°C (Elliott *et al*., 1988), respectively. Total immature developmental duration of *Nilaparvata lugens* decreased from 74.4 days at 15°C to 22.4 days at 30°C (Vattikuti *et al*., 2019).This means that *R.maidis* have a wider temperature range for immature development on corn leaves than on barley leaves.The present results indicated that the lower degree days requirement for completion of nymphal stage at eCO_2 and might be due to shortening of development time.

Lower threshold temperatures and thermal constants of R. maidis

Linear regression model was used to calculate the lower temperature threshold and thermal constant. The development rate of each stage of insect was regressed on temperature and linear regression equations are depicted in the Table 2. The linear regression equations of development rate with temperature under eCO_2 conditions indicated significant multiple coefficient of determination for first nymphal instar (y = 0.039x - 0.394, R^2= 0.982), second nymphal instar (y= 0.045x - 0.227, R^2= 0.856), third nymphal instar (y= 0.021x + 0.282, R^2 = 0.880), fourth nymphal instar (y = 0.037x + 0.836, R^2= 0.728), total nymphal duration (y = 0.009x – 0.098, R^2= 0.966), adult duration (y = 0.048x – 1.098, R^2= 0.636) and total development period (y = 0.010x – 0.202, R^2= 0.824). Similarly the linear regression equations for different stages of *R. maidis* under aCO_2 indicated significant multiple coefficient of determination for first nymphal instar (y = 0.034x-0.453, R^2= 0.945), second nymphal instar (y= 0.041x - 0.386, R^2= 0.906), third nymphal instar (y= 0.022x + 0.421, R^2 = 0.743), fourth nymphal instar (y = 0.031x + 0.945, R^2= 0.831), total nymphal duration (y

= 0.008x + 0.131, R^2= 0.962), adult duration (y = 0.046x – 1.068, R^2= 0.637) and total development period (y = 0.009x – 0.188, R^2= 0.823) (Fig 2a & 2b). Development rate and temperature were separately plotted to determine the lower development threshold of *R. maidis* at two CO_2 conditions. The lower development threshold for durations of first, second, third, fourth nymphal instar, total nymphal duration, adult duration and total development period required 10.1, 5.04, 13.42, 26.96, 10.9, 23.22 and 20.20°C under *e*CO_2 whereas it was 13.32, 9.41, 19.13, 30.48, 16.38, 22.88 and 20.89°Cunder *a*CO_2 conditions, respectively. The mean lower development threshold for nymph was slightly higher (16.38°C) at *a*CO_2 compared to that of *e*CO_2 (10.90°C) whereas for adult the mean lower development threshold was slightly higher (23.22°C) at *e*CO_2 compared to that of *a*CO_2 (22.88°C). The rate of insect development is affected by the temperature to which insects are exposed. The thermal requirement of *R. maidis* from first nymphal instar to adult(total development period) was found to be 100.00 degree days (DD) under *e*CO_2 conditions as against 111.11 degree days under *a*CO_2 conditions. Durations of first, second, third andfourth nymphal instar, total nymphal duration, adult duration required 25.64, 22.22, 41.66,27.02, 111.11 and 21.74 DD under *e*CO_2 conditions as against 29.41, 24.39, 45.45, 32.25,125.00 and 20.83 DD under *a*CO_2 conditions, respectively. Thus, the thermal requirements indicated that the lower DD requirement for completion of nymphal stage at *e*CO_2 compared to that of *a*CO_2. In contrast, adult required higher DD at *e*CO_2 with the corresponding temperatures of 20-35°C (Table 2).

According to lower temperature thresholds and thermal constants, the results were inconformity with the findings of Kuo *et al*. (2006) who reported that the lower thermal constants for *R. maidis* from 33.15 DD for first instars to 25.96 DD for fourth instars. Similar results were observed by Srinivasa Rao *et al*. (2017) who reported that the thermal requirement of nymph varied from 74-102 DD on *e*CO_2 with temperature in the range of 20-30°C as against 90-130 DD on *a*CO_2 and the results indicated that the lower degree days requirement for completion of nymphal stage at *e*CO_2. The lower thermal constants of *A.craccivora* at *e*CO_2 conditions might be due to reduction of DT as reported earlier (Berberet*et al*., 2009). Similar kind of variation of 513-290 DD for soybean aphid was reported by McCornack *et al*. (2004) in the 20-35°C temperature range. These lower thermal requirements might be due to feeding on low nutritious maize foliage obtained from *e*CO_2conditions. It was suggested that the thermal constant is influenced not only by the temperature but also by the biochemical constituents present in the host plant viz., nitrogen, proteins, aminoacids and tannins (Marchioro and Foerster, 2011). The estimated temperature thresholds and thermal constants are useful in the prediction of population peaks (Taveras *etal*., 2004) and the present data will be relevant in future climate change scenarios to understand the distribution and abundance of insect pest.

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