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***Research Paper***

# Effect of elevated carbon dioxide on biology and morphometric parameters of yellow stem borer, *Scirpophaga incertulas* infesting rice (*Oryza sativa*)

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## ABSTRACT

An experiment was conducted at Research Farm, National Rice Research Institute, Cuttack, Odisha, India to quantify the effect of elevated carbon dioxide ($CO_2$) concentrations on the biology and morphometric parameters of yellow stem borer (*Scirpophaga incertulas*, Pyralidae, Lepidoptera). Yellow stem borer is one of the major pest of rice in the whole rice growing regions of South East Asia. The effect of three carbon dioxide concentrations i.e. 410 ppm (ambient), 550 ppm and 700 ppm on the duration of the developmental period as well as morphometric parameters of each stage of the lifecycle of the pest was analysed. It was found that, there was an increase in the duration of the developmental period of each stage of life cycle as the concentration of $CO_2$ increases. However, the life span of the adult moth was significantly lower under the elevated $CO_2$ concentrations when compared with ambient $CO_2$ concentration. Morphometric parameters *viz*., mean length, width and weight of each larval instar, pupa and adult were found to be significantly higher in elevated concentrations of $CO_2$ as compared to ambient concentration.

***Key words*:** Cereals, Insect, Climate change, Growth, Development

Climate change, a prominent and major environmental issue, has been occurring due to several anthropogenic activities and natural variability. Because of this phenomenon, the earth's average temperature along with $CO_2$ concentration is increasing gradually. The concentration of $CO_2$ in the earth's atmosphere is currently around 410 ppm which is rising continuously. The concentration of $CO_2$ has been anticipated to upsurge up to 550 ppm by the year 2050 and to double by the end of the $20^{th}$ century (IPCC 2018). This continuous rise in the concentration of $CO_2$ is known to affect each and every living organism on earth. Insects being living organism also affected by this increase directly or indirectly. Indirectly it affect by altering the chemical composition and nutritional value of the host plant on which insect feed, whereas directly by altering the phenology, distribution, dynamics, abundance, development, fecundity, the number of generation and survivability of insect pest (Merrill *et al.,* 2008). The present investigation was carried out in order to evaluate the possible effects of elevated $CO_2$ concentrations on the biology and morphometric parameters of the yellow stem borer which is a major pest of rice in South East Asia.

## MATERIALS AND METHODS

The experiment was conducted in the Open Top Chamber (OT Chamber) located at Research Farm, ICAR-National Rice Research Institute, Cuttack, Odisha, India during the *kharif* season of 2018-19 which is situated at $20^0$ N latitude, $86^0$ E longitude and elevation of 23.5 m above the mean sea level (MSL). The experiemnatl set-up consisted three OT chambers in which three levels of $CO_2$ concentrations i.e. 700 ppm, 550 ppm and ambient concentration (410 ppm) were maintained. There are three replications of this set-up and in total, there are nine OT chambers. In each OT chambers, plants were grown in the pots with the recommended package of practices and are covered with a mylar cage in order to prevent the infestation of inset pest. When the crop is of two month old, one pair of field collected moth of YSB was released in each pot and again covered with mylar cage. Next days (after 20 hrs) observations were recorded regarding the laying of

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**Fig 1:** Rearing of yellow stem borer (*Scirpophaga incertulas*) with in OT chambers

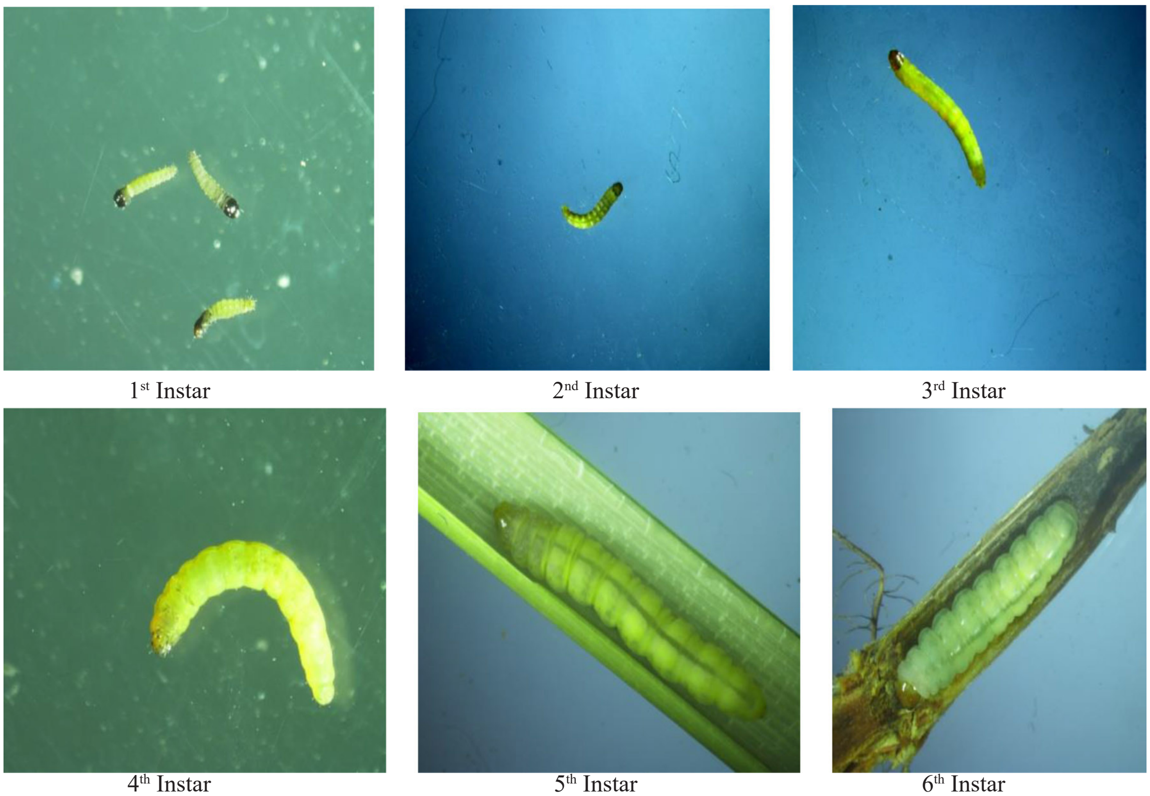


**Fig 2**: Larval instar of yellow stem borer (*Scirpophaga incertulas*)

egg masses. Observations regarding biology, developmental period and morphometric parameters of each and every stage of the life cycle were recorded as per following procedure.

In order to study the biology and morphometric parameters of yellow stem borer, larvae were reared on rice plant grown in pots under OT chambers with varying $CO_2$ concentrations. For this, the egg masses were carefully collected from the plants by removing the mylar cage and observation regarding length and width of egg masses wererecorded, then kept inside the glass vial properly. The glass vials were kept on a tray inside the each OT chamber. On hatching, the egg period or incubation period was recorded. After that, length and width of 1st instar larva wererecorded and the 1st instar larva was reared in a glass beaker lined with filter paper provided with stems of the plant grown in the OT chambers. The stems were split in the every alternate days in order to record the observation regarding developmental period and morphometirc of the each instars. The last instar larvae were reared in the stem having roots (the bottom portion of the rice plant) as it pupates within it (Fig. 1). Prior to pupation, the last larval instar covered the upper portion of stem with white silken thread. Stem plugged with white silken thread were inspected to check the pupal stage of YSB. Stem plugged with white silken thread were then sorted carefully and splited to collect the developing pupa. The collected pupa was kept properly for the emergence of the adult. The pupal period was recorded as the time between the formation of the pupa and emergence of the adult moths. After the emergence of the adult moths, male and female moths were separated, observation regarding morphometric (length and width with wing expansion and without wing expansion) and life span was recorded. The observation regarding morphometric parameters of the various stage as well as instars was recorded by using stereoscopic binocular microscope. The experiment was repeated twice during the *kharif* season of 2018-19 (August to Mid November).

The impact of the elevated concentrations on the biology and morphometric parameters of yellow stem borer was analysed by using one way ANOVA. For study of biology, there were 20

**Table 1:** Comparative developmental period (in Days) of different stages of yellow stem borer (*Scirpophaga incertulas*) reared under elevated $CO_2$ conditions

| Parameters | 410 ppm | 550 ppm | 700 ppm | SEm (±) |
|---|---|---|---|---|
| Egg period (in days) | 5.50 ± 0.51[a] | 5.20 ± 0.41 [b] | 4.55 ± 0.51 [c] | 0.024 |
| 1st Instar (in days) | 2.65 ± 0.67 [c] | 2.95 ± 0.75 [b] | 3.90 ± 0.64 [a] | 0.036 |
| 2nd instar (in days) | 3.50 ± 0.51 [c] | 3.85 ± 0.36 [b] | 4.35 ± 0.48 [a] | 0.023 |
| 3rd Instar (in days) | 4.10 ± 0.55 [c] | 4.45 ± 0.51 [b] | 5.30 ± 0.47 [a] | 0.026 |
| 4th Instar (in days) | 4.90 ± 0.55 [c] | 5.35 ± 0.48 [b] | 5.60 ± 0.50 [a] | 0.026 |
| 5th Instar (in days) | 5.40 ± 0.50 [c] | 5.85 ± 0.67 [b] | 6.20 ± 0.41 [a] | 0.027 |
| 6th Instar (in days) | 5.75 ± 0.71 [c] | 5.95 ± 0.39 [b] | 6.25 ± 0.44 [a] | 0.027 |
| Larval period (in days) | 26.30 ± 1.45 [c] | 28.40 ± 1.09 [b] | 31.60 ± 0.75 [a] | 0.057 |
| Pupa period (in days) | 7.15 ± 0.67 [c] | 7.55 ± 0.51 [b] | 7.70 ± 0.47 [a] | 0.028 |
| Developmental Period (in days) | 38.95 ± 1.66[c] | 41.15 ±1.38b[ab] | 43.85 ±1.53[a] | 0.077 |
| Adult lifespan(in days) | 2.75 ± 0.91 [a] | 2.35 ± 0.87 [b] | 2.30 ± 0.80[bc] | 0.043 |

Values followed by different letters in column are significantly different at $p= 0.05$ by LSD

replications and each replication consisted of 10 larvae, whereas for morphometric studied, there were 10 replications and each replication consisted of 10 numbers of larvae. The experiment was repeated twice in the same season. The results were pooled and presented as mean ± standard deviation. All statistical analysis were performed by using SPSS version 16.0.

## RESULTS AND DISCUSSION

***Biology of YSB under elevated $CO_2$ concentrations***

The developmental period of each and every stage of yellow stem borer under the different concentration of $CO_2$ was investigated and data obtained were presented in Table 1.

***Egg period:*** There is a slight variation in the incubation period under elevated $CO_2$ concentrations (Table 1). The incubation period was significantly less in higher concentration (700 ppm) of $CO_2$ (4.55 ± 0.51days) followed by 550 ppm (5.20 ± 0.41days) and ambient concentration (5.50 ± 0.51 days).

***Larva period:*** There is significant variationin the duration of the developmental period of the larva as well as different instars (Table 1). So far six larval instar were observed among all the treatments. The first instar larva was pale yellow to greenish in colour with blackish to brownish colour head whereas the subsequent instars (2nd , 3rd ,4th and 5th) were light greenish in colour with dark brown head. The last instar larvae were pale green with more of less transparent and are sluggish in nature (Fig. 2). The 1st and 2nd instar larvae feed on tender parts of the stem (internode region) whereas 3rd , 4th , 5th and 6th instar larvae feed on the node as mandibles are strongly developed and sclerotised. From this investigation, it was found that the duration of the larval period was increased with the increase in concentration of $CO_2$. The larva took 26.30 ± 1.45, 28.40 ± 1.09 and 31.60 ± 0.75 days to complete their growth and development in ambient, 550 ppp and 700 ppm $CO_2$ concentration respectively (Table 1).

***Pupal period*:** The pupa were dark brown in colour and the period of their development was significantlyhigher in the elevated $CO_2$concentrations (7.70 ± 0.47, 7.55 ± 0.51 days) as compared to ambient concentrations (7.15 ± 0.67 days) (Table 1).

***Developmental period:*** Delay in the development of 3 to 4 days was observed under elevated concentrations of $CO_2$ as compared to ambient concentration. The insect took 38.95 ± 1.66 days to complete its development from the egg to the emergence of the adult as compared to 41.15 ± 1.38 and 43.85 ± 1.53 days at 550 ppm and 700 ppm, respectively (Table 1).

***Adult:*** Sexual dimorphism is common in yellow stem borer. The female is differentiated from the male by the presence of a black spot on the middle of each forewings and a brownish tuft of hair at anal segments. The males are comparatively smaller than the female. The longevity of adults was found to be significantlylower in elevated concentrations i.e 700 and 550 ppm (2.30 ± 0.80, 2.35 ± 0.87 days) as compared to ambient concentration (2.75 ± 0.91 days) (Table 1).

***Comparative morphometric parameters of YSB under the elevated $CO_2$ concentrations***

The morphometric parameters such as mean length, body width, head width and body weight of each and every stage of the life cycle have been presented in Table 2.

***Egg:*** There was no significant variation in the mean length and width of egg masses laid by the field collected adult female that were reared under the elevated $CO_2$ concentrations (Table-2). The mean length of egg masses varied from 5.264 ± 0.85 to 5.35 ± 0.53 whereas their width varied from 1.712 ± 0.25 to 1.937 ± 0.27 under the different concentrations of $CO_2$.

***Larvae:***The mean length, body width, head width as well as body weight of larvae was found to be vary under the elevated $CO_2$ concentrations. No significant variation in size and weight was observed among the 1st instar larva, whereas significant variation in mean length, body width, head width as well as in body weight was observed in subsequent instar that were reared under the elevated $CO_2$ concentrations (Table 2). It was observed that, mean length, body width as well as head width and body weight increased with the rise in $CO_2$ concentration in all instars except 1st instar. An exception was observed for the body width of the 6th instar larvae, though it was statistically at par, found to be maximum under 550 ppm (2.021 ± 0.16) as compared to 700 ppm (2.013 ± 0.27) concentrations of $CO_2$. The mean length and head width of the 6th instar (final instar) larvae was found to be significantly higher in elevated condition

**Table 2:** Comparative morphometric parameters of different stages of yellow stem borer (*Scirpophaga incertulas*) reared under elevated $CO_2$ concentrations

| Stages of YSB | | | 410 ppm | 550 ppm | 700 ppm | SEm (±) |
|---|---|---|---|---|---|---|
| Egg mass laid by field collected YSB female | | Length (mm) | 5.350 ± 0.53$^{a}$ | 5.264 ± 0.85$^{ab}$ | 5.278 ± 0.81$^{abc}$ | 0.075 |
| | | Width (mm) | 1.712 ± 0.25$^{c}$ | 1.937 ± 0.27$^{a}$ | 1.921 ± 0.41$^{ab}$ | 0.032 |
| Larvae | 1st Instar | Length (mm) | 0.971± 0.04$^{abc}$ | 0.973 ± 0.06$^{ab}$ | 0.982 ± 0.05$^{a}$ | 0.005 |
| | | Body Width (mm) | 0.200 ± 0.01$^{a}$ | 0.190 ± 0.01$^{bc}$ | 0.191 ± 0.01$^{b}$ | 0.002 |
| | | Head Width (mm) | 0.128 ± 0.024$^{abc}$ | 0.129 ± 0.031$^{ab}$ | 0.134 ± 0.02$^{a}$ | 0.003 |
| | 2nd Instar | Length (mm) | 3.258 ± 0.25$^{c}$ | 4.823 ± 0.47$^{b}$ | 7.952 ± 0.55$^{a}$ | 0.045 |
| | | Body Width (mm) | 0.467 ± 0.05$^{c}$ | 0.641± 0.05$^{b}$ | 0.834 ± 0.04$^{a}$ | 0.005 |
| | | Head Width (mm) | 0.296 ± 0.065$^{c}$ | 0.537 ± 0.049$^{b}$ | 0.688 ± 0.051$^{a}$ | 0.006 |
| | | Body Weight (g) | 0.001 ± 0.0007$^{c}$ | 0.004 ± 0.0005$^{b}$ | 0.0092 ± 0.001$^{a}$ | 0.0001 |
| | 3rd Instar | Length (mm) | 5.243 ± 0.286$^{c}$ | 6.959 ± 0.497$^{b}$ | 9.160 ± 0.660$^{a}$ | 0.051 |
| | | Body Width (mm) | 0.667 ± 0.035$^{c}$ | 0.841 ± 0.043$^{b}$ | 1.182 ± 0.073$^{a}$ | 0.005 |
| | | Head Width (mm) | 0.505 ± 0.034$^{c}$ | 0.642 ± 0.041$^{b}$ | 0.780 ± 0.043$^{a}$ | 0.004 |
| | | Body Weight (g) | 0.0042 ± 0.006$^{c}$ | 0.0062 ± 0.001$^{b}$ | 0.0152 ± 0.003$^{a}$ | 0.0002 |
| | 4th Instar | Length (mm) | 7.578 ± 1.01$^{c}$ | 11.309 ± 0.96$^{b}$ | 14.340 ±1.28$^{a}$ | 0.110 |
| | | Body Width (mm) | 0.964 ± 0.04$^{c}$ | 1.298 ± 0.10$^{b}$ | 1.473 ± 0.11$^{a}$ | 0.009 |
| | | Head Width (mm) | 0.784 ± 0.069$^{c}$ | 0.959 ± 0.078$^{b}$ | 1.091± 0.038$^{a}$ | 0.006 |
| | | Body Weight (g) | 0.022 ± 0.041$^{c}$ | 0.037 ± 0.0067$^{b}$ | 0.053 ± 0.011$^{a}$ | 0.0025 |
| | 5th Instar | Length (mm) | 11.163± 1.37$^{c}$ | 15.085 ± 1.24$^{b}$ | 15.686 ± 1.65$^{a}$ | 0.144 |
| | | Body Width (mm) | 1.370 ±0.26$^{c}$ | 1.594 ± 0.23$^{b}$ | 1.748 ± 0.23$^{a}$ | 0.025 |
| | | Head Width (mm) | 0.883±0.093$^{c}$ | 1.018 ± 0.075$^{b}$ | 1.056 ± 0.094$^{a}$ | 0.009 |
| | | Body Weight (g) | 0.031 ±0.0166$^{c}$ | 0.063 ± 0.0133$^{b}$ | 0.078 ± 0.018$^{a}$ | 0.0022 |
| | 6th Instar | Length (mm) | 14.155 ± 1.70$^{c}$ | 17.442 ± 1.85$^{b}$ | 19.757 ± 2.86$^{a}$ | 0.220 |
| | | Body Width (mm) | 1.628 ± 0.19$^{c}$ | 2.021 ± 0.16$^{a}$ | 2.013 ± 0.27$^{ab}$ | 0.022 |
| | | Head Width (mm) | 1.428 ± 0.220$^{c}$ | 1.836 ± 0.152$^{ab}$ | 1.840 ± 0.252$^{a}$ | 0.021 |
| | | Body Weight (g) | 0.058 ± 0.012$^{c}$ | 0.0900 ± 0.014$^{b}$ | 0.0990 ± 0.015$^{a}$ | 0.0014 |
| Pupa | | Length (mm) | 8.126 ± 0.48$^{c}$ | 8.504 ± 0.55$^{b}$ | 8.799 ± 0.47$^{a}$ | 0.051 |
| | | Width (mm) | 1.673 ± 0.26$^{c}$ | 1.866 ± 0.19$^{b}$ | 1.989 ± 0.22$^{a}$ | 0.023 |
| Adult male | | Length (mm) | 10.393 ± 0.45$^{c}$ | 10.748 ± 0.42$^{b}$ | 11.087 ± 0.38$^{a}$ | 0.042 |
| | | Width without wing expansion (mm) | 8.512 ± 0.34$^{c}$ | 8.78 3± 0.27$^{b}$ | 9.009 ± 0.32$^{a}$ | 0.031 |
| | | Width with wing expansion (mm) | 15.380 ± 0.44$^{c}$ | 15.921 ± 0.33$^{b}$ | 16.035 ± 0.33$^{a}$ | 0.038 |
| Adult female | | Length (mm) | 13.491 ± 0.55$^{c}$ | 13.848 ± 0.27$^{b}$ | 14.078 ± 0.32$^{a}$ | 0.041 |
| | | Width without wing expansion (mm) | 11.154 ± 0.34$^{c}$ | 11.754 ± 0.26$^{b}$ | 11.869 ± 0.29$^{a}$ | 0.030 |
| | | Width with wing expansion (mm) | 18.879 ± 0.26$^{c}$ | 19.362 ± 0.26$^{b}$ | 20.169 ± 0.19$^{a}$ | 0.024 |

The unit of Length, Body width and head width is millimetre (mm) whereas that of body weight is gram (g); Values followed by different letters in column are significantly different at $p$= 0.05 by LSD

i.e 19.757 ± 2.86 and1.840 ± 0.252in 700 ppm, 17.442 ± 1.85 and 1.836 ± 0.152 in 550 ppm as compared to ambient condition i.e 14.155 ± 1.70 and 1.428 ± 0.220 in 410 ppm. The body weight of the last instar larva was also found to be more in higher concentration of $CO_2$ (0.099 ± 0.015 in 700 ppm and 0.09 ± 0.014 ) as compared to the ambient concentration (0.058 ± 0.012).

***Pupa:*** The mean length and width of the pupa also varied significantly under theelevated $CO_2$ concentrations (Table 2). The mean length and width of the pupa was found to be higher in 700 ppm (8.799 ± 0.47 and 1.989 ± 0.22) followed by 550 ppm (8.504 ± 0.55 and 1.866 ± 0.19) and 410 ppm (8.126 ± 0.48 and 1.673 ± 0.26).

***Adult:*** The mean length and width of both male as well as female varied significantly under the elevated $CO_2$ concentrations. The males and females that were emerged under elevated conditions were found to be longer and wider than that were emerged under ambient conditions(Table 2). Again the female moths were bigger in size as compared to the male moths. The mean length of male and female moth was found to be 11.087 ± 0.38 mm and 14.078 ± 0.32 mm, 10.748 ± 0.42 mm and 13.848 ± 0.27 mm, 10.393 ± 0.45 mm and 13.491 ± 0.55 mm under 700 ppm, 550 ppm and ambient $CO_2$ conditions, respectively. Similarly, the mean width with wing expansion of male and female moth was found to be 16.035 ± 0.33 mm and 20.169 ± 0.19 mm, 15.921 ± 0.33 mm and 19.362 ± 0.26 mm, 15.380 ± 0.44 mm and 18.879 ± 0.26 mm under 700 ppm, 550 ppm and ambient $CO_2$ concentrations, respectively.

The responses of insect herbivores to elevated $CO_2$ are species specific (Hillstrom *et al.,* 2010). Some species respond positively whereas some species responds negatively to the elevated $CO_2$ levels. In our studied, we observed that increase in concentration of $CO_2$ had no effect on the incubation period of egg masses laid by the field collected adult moths of yellow stem borer. However, there was an increase in duration of the developmental period for each larval instar as well as pupa that is grown under elevated $CO_2$ compared to ambient conditions. Similar findings have been reported previously for different insects like tobacco caterpillar (*Spodoptera litura*) on peanut (Kumari and Verma, 2021), bollworm (*Helicoverpa armigera*) on the artificial diet (Liu *et al.,* 2017) and brown plant hopper (*Nilparvata lugens*) on rice (Bao-kun *et al.,* 2014). However, decrease in duration of each nymphal instar as well as total developmental period with increase in $CO_2$ cconcentration was reported by (Mounica et al., 2020).This increase in the duration of the developmental period may be due to the alternation in nutritional composition of the host plants in terms of the quantity of protein required by an insect herbivore that were grown under the elevated $CO_2$ concentrations. Elevated $CO_2$concentrations led to the increased rate of photosynthesis, growth and production of biomass (Sreenivas *et al.,* 2021) which in turns increases the C:N ration of plants that were grown under the elevated $CO_2$ concentrations (Goverde and Erhardt, 2003). This increase in C:N ratio leads to a reduction in nitrogen concentration on different parts of plants (Kumari and Verma, 2021). Several studies reported that, growth and development as well as performance of the insect herbivore is positively correlated with the nitrogen content of the plant and there is an increase in duration of the developmental period of insect herbivores when they feed on the plant with reduced nitrogen content. Decrease in protein content of the rice plant was also observed when it was grown under elevated $CO_2$ concentration (Ujiie *et al.,* 2019). In order to compensate the protein requirement for development, the larvae have to be consumed more amounts of food materials and required more time to convert these ingested food materials to biomass (Kumari and Verma, 2021). In contrast, the adult that was emerging from the pupa reared under the elevated $CO_2$ concentrations had a shorter lifespan as compared to that emerged from the pupa grown under ambient concentration.

The increase in length, width as well as the weight of each larval instar was observed under the elevated $CO_2$ concentrations as compared to the ambient conditions. This was probability because of compensatory increase in food consumption by most of the phytophagous herbivores under elevated concentrations of $CO_2$ (Lee *et al.,* 2002). The reduction in nutritional quality of different plant parts grown under the elevated $CO_2$ concentrations was well documented (Coviella *et al.,* 2000). Several studies reported that insect herbivores increase their individual consumption rate in order to fulfill their nutritional requirement when reared on plants that were grown under the elevated $CO_2$ concentrations. This increase in individual consumption rate leads to proportionate increase in body size as well as the weight of larvae under elevated $CO_2$ concentration.

## CONCLUSION

The morphometric parameters of larva of yellow stem borer such as body length, width and weight were found to be increased with increase in the concentration of $CO_2$. The rise in the concentration of $CO_2$ led to increase in the duration of the larval and pupal period whereas decreased in the duration of the incubation period as well as life span of the adult moth was observed in the higher concentration of $CO_2$. The total developmental period of yellow stem borer was also found to be increased when they were reared under the higher concentration of $CO_2$. The findings may help in fine tuning of management strategies of the insect-pest.

***Conflict of Interest Statement***: The author(s) declare(s) that there is no conflict of interest.

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