# Effects of carbon dioxide enrichment during different growth periods on flowering, pod set and seed yield in Soybean

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## Abstract

The objective of this study is to elucidate the effects of  $CO_2$  enrichment during different growth periods on flowering, pod set and seed yield in soybean (*Glycine max* (L.) Merr.). Soybean cultivar 'Fukuyutaka' was grown in a growth chamber of the Institute of Bioresources Research Center of Kyushu Electric Power Co., Inc. at Saga, Japan (33?17?-N, 130?18?-E) under natural light. The  $CO_2$  concentrations were maintained at 350µmol  $CO_2$  mol<sup>-1</sup> for ambient  $CO_2$  and at 700µmol  $CO_2$  mol<sup>-1</sup> for  $CO_2$  enrichment.  $CO_2$  concentration was enriched during the whole growth period (WP), vegetative growth period (VP) or reproductive period (RP). Seed yield was increased by 25% by  $CO_2$  enrichment during RP or WP due to the increase of pod number, but was not by  $CO_2$  enrichment during VP. Although  $CO_2$  enrichment had no effect on the number of flowers,  $CO_2$  enrichment during RP increased the pod number on all raceme orders and that during WP increased the pod number in the secondary and tertiary racemes. It is suggested that an increase of seed yield by  $CO_2$  enrichment is mostly brought by the improvement of pod set, mainly on the high-order racemes that opened later during flowering period, and that the response of seed yield is mainly attributed to  $CO_2$  enrichment during RP.

# Media summary

CO<sub>2</sub> enrichment during reproductive period affected mainly the improvement of the pod set on the higherorder racemes that opened later during flowering period.

## **Key Words**

CO<sub>2</sub> enrichment, Growth stage, Flowering, Pod set, Raceme order, Soybean

## Introduction

Since the Industrial Revolution, the concentration of carbon dioxide  $(CO_2)$  in the atmosphere has been increasing. This increase is almost certainly due primarily to the continued burning of fossil fuels. According to the Intergovernmental Panel on Climate Change (IPCC, 2001), the atmospheric  $CO_2$  concentration is predicted to double by the end of this century. The steadily increasing level of  $CO_2$  is expected to enhance plant growth and to increase seed yield in grain crops. Long-term experiments have generally shown that an elevated  $CO_2$  concentration ( $CO_2$  enrichment) brings about high yields through high photosynthetic rates and rapid biomass accumulation (Zelitch, 1982; Kimball, 1983).

In soybean,  $CO_2$  enrichment has been reported to increase the photosynthetic rate (Jones et al., 1984; Rogers et al., 1984) and leaf area (Ziska and Bunce, 1995), and therefore the total dry weight (Cooper and Brun, 1967; Allen et al., 1991). These results suggest that the stimulation of vegetative growth by  $CO_2$  enrichment is important for the increase of seed yield. On the other hand, some researchers reported that seed yield was increased by  $CO_2$  enrichment during the reproductive growth period when vegetative growth was almost completed (Hardman and Brun, 1971; Ackerson et al., 1984). Thus, the response of seed yield to  $CO_2$  enrichment is not always correlated with vegetative parameters. Recently, Ziska et al. (2001) reported that the seed yield response to  $CO_2$  enrichment is associated with plasticity in the ability to set additional pods on branches in a  $CO_2$  enriched environment. However, the morphological developmental process by which this increase in seed yield comes about is not well known. Abortion of flower and young pods is an important limiting factor of seed yield in soybean (Van Schaik and Probst, 1958). Although soybean flowers are produced abundantly, a large number of flowers and young pods abscise rather than develop into mature pods. The flowering period varies from 18 to 50 days in soybean, and the seed yield depends on the pods set on various racemes which flowered at different times. Therefore, an investigation of the number of flowers and the pod set on individual raceme orders may be an effective way of estimating the effect of elevated  $CO_2$  concentrations on the seed yield in soybean.

The objective of this study was to determine the effects of  $CO_2$  enrichment during various growth periods on flowering and pod set to clarify whether and how  $CO_2$  enrichment causes an increase in seed yield in soybean.

## Methods

The experiments were conducted at the Bioresources Research Center of Kyushu Electric Power Co., Inc. in Saga, Japan (33?17?-N, 130?18?-E). Soybean (*Glycine max* (L.) Merr. cv. Fukuyutaka) seeds were inoculated with Bradyrhizobium japonicum (A1018) and planted in plastic pots (20cm in diameter and 21cm in height) filled with fine sand on 19 July, 2000 and 18 July, 2001. After emergence, the plants were moved into the growth chambers under natural light and with CO<sub>2</sub> concentrations maintained at 350 $\mu$ mol CO<sub>2</sub> mol<sup>-1</sup> (Control) and 700 $\mu$ mol CO<sub>2</sub> mol<sup>-1</sup>(CO<sub>2</sub> enrichment). The chambers were constructed with steel frames and covered with a layer of plastic film (about 40% shade in 2000, and about 25% shade in 2001 because of the change of film). The day/night air temperature was controlled at about 28/22?C, except for 2 hours at sunrise (0700 to 0900 Japan Standard Time (JST)) and 2 hours at sunset (1800 to 2000 JST), when it was set at 25?C. Nutrient solution was given automatically twice every day at 0900 and 1400 (JST) five minutes each, but the concentration of the nutrient was maintained at 50% of the standard until flowering. The CO<sub>2</sub> concentration was elevated through whole growth period (WP, from emergence to maturity) in 2000, but in 2001 during the vegetative growth period (VP, from emergence to the onset of flowering), the reproductive growth period (RP, from flowering to maturity), and WP. Before and after the CO<sub>2</sub> enrichment, the plants were exposed to the same as control. Each treatment consisted of six plants and a half of them were used to record flower number. After the first flower opened, the raceme order of the flowers that had opened that day was recorded every two days. Flowers were distinguished at each node and each raceme order following the method described by Kuroda et al. (1992). The flowers that had been counted were marked by giving a cut injury through into the standard vexillum of the flower. All of the plants were harvested after matured, and the vegetative growth parameters, yield components and seed yield were measured after the plants were dried naturally in a room.

#### Results

In 2000,  $CO_2$  enrichment during WP increased the seed yield per plant about 13%, and it was also increased the pod number and seed number by about 12% (Table 1). In 2001,  $CO_2$  enrichment during RP and WP increased seed yield by about 25%, but that during VP had no effect.  $CO_2$  enrichment during WP and RP increased the number of matured pods and seeds. However, seed size was unaffected by that during any of the period (WP, VP or RP). Thus,  $CO_2$  enrichment increased seed yield not by increasing seed size but by increasing the number of matured pods per plant.

The number of flowers on each raceme order was not affected by the  $CO_2$  enrichment during any of the periods (Fig. 1). Although  $CO_2$  enrichment during VP and WP slightly decreased the number of flowers on the terminal, primary and secondary racemes,  $CO_2$  enrichment during any periods had no significant effect on the total number of flowers in 2001 (Table 1).

Table 1. Influence of  $CO_2$  enrichment during the different growth periods on the vegetative growth parameters, the yield components and the seed yield after harvest. The values of the number of flowers and the rate of pod set are means of 3 plants and those of the other parameters are means of 6 plants. \* indicated a significant difference, and ns no significant difference at 5% level according to Student's t-test. Treatments followed by the same letter are not significantly different

Period of CO <sub>2</sub> enrichment	Length of main stem (cm)	Total number of nodes	Dry weight of stem (g)	Number of flowers	Number of matured pods	Number of matured seeds	Rate of pod set (%)	100-seed weight (g)	Seed yield(g)
2000									
С	70.7	69.8	19.6	246.0	168.7	323.7	77.6	31.3	101.9
WP	69.8	82.0	25.3	294.7	192.5	371.6	71.1	32.5	114.9
	ns	*	ns	*	ns	ns	ns	ns	ns
2001									
(C)	44.0 a	65.2 a	26.8 в	285.0 a	170.2 в	307.8 в	64.7 a	44.0 a	135.2 в
VP	43.6 a	66.2 a	27.7 в	231.7 a	161.8 b	290.2 в	70.1 a	45.1 a	130.8 в
RP	48.3 a	70.2 a	28.5 в	268.7 a	210.2 a	384.8 a	76.5 a	43.8 a	168.2 a
WP	45.3 a	69.5 a	35.7 a	253.0 a	210.3 a	393.0 a	77.1 a	43.2 a	169.4 a

at 5% level according to One-way ANOVA and Bonferroni's multiple range test. C, Control; WP, whole growth period; VP, vegetative growth period; RP, reproductive growth period.

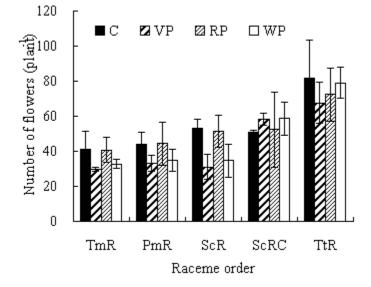


Fig. 1. Number of flowers on each raceme order of the plants exposed to elevated  $CO_2$  concentrations during the different growth periods in 2001. Data are means of 3 plants ? S.D. C, Control; VP,  $CO_2$  enrichment during the vegetative growth period; RP,  $CO_2$  enrichment during the reproductive growth period; WP,  $CO_2$  enrichment during the whole growth period. TmR, terminal raceme (flowering followed PmR after 2 days); PmR, primary raceme(flowering started first); ScR, secondary raceme(flowering followed PmR after 8 days); ScRC, secondary raceme with compound leaves(flowering followed PmR after 6 days); TtR, tertiary raceme (flowering followed PmR after 14 days).

Fig. 2 shows the effects of  $CO_2$  enrichment during different growth periods on the number of pods on the individual raceme order.  $CO_2$  enrichment during RP slightly increased the numbers of matured pods on the terminal, primary and secondary racemes, but that during VP and WP decreased them. However,  $CO_2$  enrichment during any period increased the pod number on the secondary racemes with compound leaves and the tertiary racemes, although that during VP did not increase the pod number of tertiary racemes. Thus,  $CO_2$  enrichment during RP increased the pod number on all racemes and that during WP increased the pod number on the secondary and tertiary racemes.

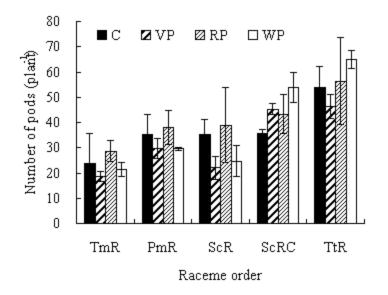


Fig. 2. Number of matured pods on each raceme order in the plants exposed to elevated  $CO_2$  concentrations during the different growth periods in 2001. Data are means of 3 plants ? S.D. Other notes are the same as in Fig. 1.

The effect of  $CO_2$  enrichment on the rate of pod set varied with the raceme order (Fig. 3).  $CO_2$  enrichment during RP increased the rate of pod set on all raceme orders. On the order hand,  $CO_2$  enrichment during WP greatly increased the rate of pod set on the secondary racemes, the secondary racemes with compound leaves and tertiary racemes; that is, on the high-order racemes. Since only the  $CO_2$  enrichment during RP and WP increased the number of pods, we suggest that seed yield can be most effectively increased by  $CO_2$  enrichment during the reproductive growth period.

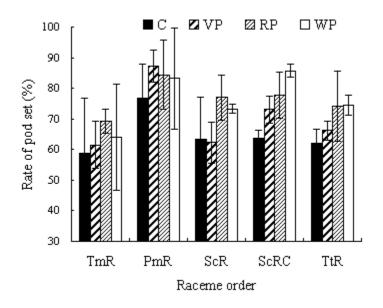


Fig. 3. Rate of pod set on each raceme order in the plants exposed to elevated  $CO_2$  concentrations under the different growth periods in 2001. Data are means of 3 plants ? S.D. Other notes are the same as in Fig. 1.

#### Conclusion

 $CO_2$  enrichment may increase seed yield by improving pod set on the high-order racemes that opened later during flowering period. Moreover, the response of seed yield to  $CO_2$  enrichment is mainly attributed to the response during the reproductive growth stage. However, further research on the accumulation and translocation of assimilate is necessary to clarify the physiological mechanism of the response to  $CO_2$  enrichment in the higher-order racemes.

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