

## **NO<sub>2</sub> Absorption and Physiological Response of Lettuce in a Semi-closed Plant Production System**

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### **Abstract**

The environmental characteristics, absorption rate of NO<sub>2</sub>, physiological response, and quality of lettuces were examined in a semi-closed plant production system consisted of human and plant modules. Leaf lettuces were transplanted to 10-cm diameter pots at 4- to 5-leaf stage, and grown in a gas-fumigated plant module. Air containing NO<sub>2</sub> was circulated 10 min-ON/ 20 min-OFF between the human and plant modules by a diaphragm. At 0.13, 0.30, and 0.45 μmol · mol<sup>-1</sup> NO<sub>2</sub>, NO<sub>2</sub> decrements in the chamber during the 10 min were 0.040, 0.109, and 0.149 μmol · mol<sup>-1</sup>, respectively. The NO<sub>2</sub> absorption rate of the plants was estimated to be 0.078 μmol · mol<sup>-1</sup> · m<sup>-2</sup> · min<sup>-1</sup> at 0.45 μmol · mol<sup>-1</sup> NO<sub>2</sub>, and no visible injuries occurred. The photosynthetic rate of the lettuce exposed to 0.45 μmol · mol<sup>-1</sup> NO<sub>2</sub> was less than that of the control plant at 4th and 5th days, whereas the respiration rate was the same. The lettuce grown at 0.45 μmol · mol<sup>-1</sup> NO<sub>2</sub> had similar leaf width, leaf length, leaf area, dry and fresh weights, and quality to that grown without NO<sub>2</sub>.

**Key words:** plant production, air purification, CO<sub>2</sub> utilization, office, urban-type plant factory

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### **Introduction**

Recently industrialization and concentration of the population to cities made worse living environment in the city, and the concerns about air pollution and indoor air quality (IAQ) increased. Indoor air cleanliness became very important for human because they spend over 80% of a day indoors. Advanced architecture techniques made the building highly closed and adiabatic for saving the energy (Hansen, 1992), increasing the contamination of indoor air. Cleaning the ambient air with forced-ventilation facilities is possible, but there are some limits to solve the indoor air-pollutants problems.

Generally CO<sub>2</sub> concentrations in offices were 400-1300 μmol·mol<sup>-1</sup> at cooling season and 600-1500 μmol·mol<sup>-1</sup> at heating season (Son et al., 2000). They were the lowest at 8:30 a.m. before people arrived at work, and the highest between 10:00 and 15:00. The characteristics of CO<sub>2</sub> change coincided with the photosynthesis cycle of plants during the daytime (Park et al., 1994; Son et al.,

2000). During the period, CO<sub>2</sub> concentrations in the offices, however, exceed the standard for indoor environments (below 1000 μmol·mol<sup>-1</sup>). High CO<sub>2</sub> concentration often causes sick building syndrome, which might make the occupants feel sick and uncomfortable, and gradually lose productivity (Chow et al., 1992).

It was suggested that the surplus CO<sub>2</sub> could be applied for the plant production and the O<sub>2</sub> emitted from plants could be used for the people by exchanging air between plant and human modules (Daunicht, 1996; Gitelson and Okladnikov, 1996; Son et al., 1999). For successful introduction of a semi-closed plant production system as urban-type plant factory (UPF), many kinds of air-pollutants in the building such as NO<sub>2</sub>, SO<sub>4</sub>, CO, etc. and their effects on plant growth have to be considered.

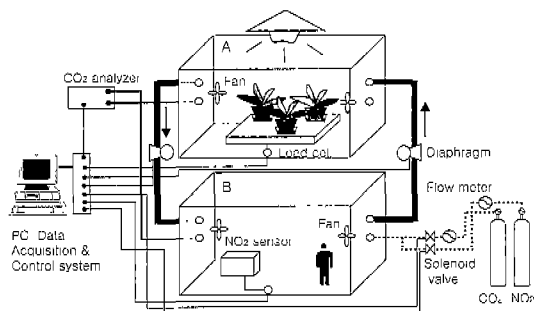
The objectives of this study were to measure the absorption rate of NO<sub>2</sub>, one of the air-pollutants, by lettuce and examine the physiological response and quality of the lettuce when the contaminated air in the human module was circulated between plant and human modules.

## Materials and Methods

**Plant materials and culture methods.** Leaf lettuce (*Lactuca sativa* L. cv. Chungchima) seeds were germinated on polyurethane cubes and hydroponically grown in a greenhouse. Twenty days after germination, the seedlings at 4- to 5-leaf stage were transplanted to 10-cm diameter pots filled with granular rockwool. Nine days after transplantation, the potted lettuces were moved to the plant module chamber and grown under metal halide lamps (250 W). Yamasaki nutrient solution at full strength was supplied. After acclimatization of plants under the artificial lights for 1 day, the experiment was started. In this stage, the lettuce radically grows fast and is sensitive to air-pollutants around plants.

In NO<sub>2</sub> experiments, air temperature and relative humidity (RH) in the chamber were maintained constant at 23±2°C and 60±10% RH during the daytime, and 18±2°C and 70±10% RH during the nighttime. The leaf area of the plants was measured by using a leaf area meter (LI-COR, 3100, USA). The photosynthetic photon flux density was 200 μmol·m<sup>-2</sup>·s<sup>-1</sup> at the top of the plants. The light period was 12 hours per day. In the relative-humidity experiment, the plant module was treated with 50 to 95 % RH, and maintained at 25±1°C during the daytime and 18±1°C during the nighttime.

**Gas-fumigated growth chamber.** The small-scale human and plant modules were constructed with 5-cm thick transparent acrylic plates and had inside a dimension of 60 cm × 50 cm × 50 cm (length × width × height)



**Fig. 1.** Schematic diagram of gas exchange, exposure, and measurement systems constructed for the experiment.

(Fig. 1). The leakage rate was less than the 1% of the chamber volume per hour. CO<sub>2</sub> was added to reach 600 – 1200 μmol·mol<sup>-1</sup> like an office in the city to duplicate the CO<sub>2</sub> concentration of a human module. The air temperature in the chamber was controlled by the room temperature. Two fans constantly agitated the air in the chamber. Environmental sensors were interfaced with a CR-23 digital and analog input/output controller (Campbell Scientific Co., CR-23, USA). This signal conversion relay system communicated with a dedicated control computer over a RS-232 communication link. The lid of chamber was opened in the nighttime.

**NO<sub>2</sub> fumigation treatment.** The set values of NO<sub>2</sub> concentrations in the human module were 0.15, 0.30, and 0.45 μmol·mol<sup>-1</sup> considering the real measured values and indoor limit values. These values were controlled using an electric-luminescent NO<sub>2</sub> analyzer (HS 1000, Mapo industry Co., Korea). The contaminated condition of NO<sub>2</sub> in the human module was controlled constant in the daytime. The air in the two modules was circulated by diaphragm 10 min-ON/ 20 min-OFF during the daytime with maintaining 0.15, 0.30 and 0.45 μmol·mol<sup>-1</sup> NO<sub>2</sub> in each human module. The sampled gas containing NO<sub>2</sub> was analyzed with chemiluminescences analyzer (Dasibi, NO<sub>2</sub>-2108, USA). In order to confirm the effect of inside moisture on the reduction of NO<sub>2</sub> in the chamber, the change of NO<sub>2</sub> concentration with time was measured at various NO<sub>2</sub> levels.

**Measurement.** L (lightness), a (green-yellow), and b (red-blue) values were measured to compare the plant quality-related colors using a colorimeter (Minolta, Japan) between 0.45 μmol·mol<sup>-1</sup> NO<sub>2</sub> treatment and the control. Tension in the leaf was measured by using a texture analyzer (Stable micro system TA-XT2, USA). The size of leaf sample was 2 cm × 4 cm.

The photosynthetic rate of plants was measured with photosynthesis analyzer (LI-COR, 6400, USA) attached with standard CO<sub>2</sub> gas in order to see physiological disorders during the experiment period. After the experiment, differences in leaf width, leaf length, leaf area, and fresh weight of the plant between NO<sub>2</sub> treatment and

the control were compared. The images of leaves were taken to determine the increase in leaf area over the 6-day treatment period, and the leaf area was calculated with image analyzer software, IMAGE PRO.

### Results and Discussion

**CO<sub>2</sub> and NO<sub>2</sub> control.** The CO<sub>2</sub> characteristics simulating CO<sub>2</sub> condition of an office in the city were executed in the gas-fumigated growth chamber (Fig. 2). We assumed attendance time to be 8:00, lunchtime from 12:00 to 13:00 and leaving time to be 18:00 in the experiment. NO<sub>2</sub> concentrations in the chamber were well controlled at 0.13, 0.30, and 0.45 μmol·mol<sup>-1</sup> in the human module (Fig. 3). The operation of the diaphragm,

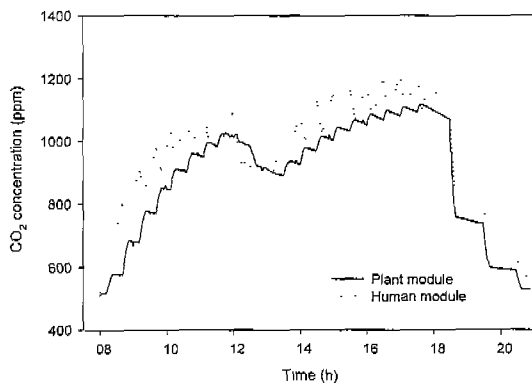


Fig. 2. Change in CO<sub>2</sub> concentration in plant and human modules under air exchange condition of 10-min ON and 20-min OFF. Lunch time was assumed to be 12:00-13:00.

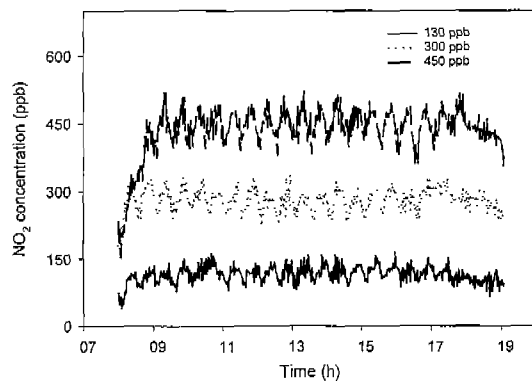


Fig. 3. Change in NO<sub>2</sub> concentration in human module during the daytime.

Table 1. NO<sub>2</sub> decrement in plant module under air circulation of 10-min ON and 20-min OFF between plant and human modules.

| Initial NO <sub>2</sub> concentration (μmol·mol <sup>-1</sup> ) | At starting point (μmol·mol <sup>-1</sup> ) | After 10 min (μmol·mol <sup>-1</sup> ) | NO <sub>2</sub> decrement (μmol·mol <sup>-1</sup> ) |
|---|---|--|---|
| 0.15  | 0.041±0.0069 <sup>z</sup>                   | 0.001±0.0025 <sup>y</sup>              | 0.040   |
| 0.30  | 0.201±0.0165                                | 0.092±0.0087                           | 0.109   |
| 0.45  | 0.393±0.0084                                | 0.244±0.0145                           | 0.149   |

<sup>z</sup>NO<sub>2</sub> concentration at 5 min after stopping air exchange.

<sup>y</sup>NO<sub>2</sub> concentration at 15 min after stopping air exchange.

<sup>x</sup>Mean±SE, n=4.

10 min-ON/ 20 min-OFF, caused the fluctuation of NO<sub>2</sub> concentration in the human module.

**NO<sub>2</sub> absorption.** Decreased NO<sub>2</sub> concentrations were 0.040, 0.109, and 0.149 μmol·mol<sup>-1</sup>, respectively, when the air contaminated by 0.13, 0.30, and 0.45 μmol·mol<sup>-1</sup> NO<sub>2</sub> in the human module was exchanged with the plant module (Table 1). The NO<sub>2</sub> decrement by absorption of the lettuce revealed a good proportion to NO<sub>2</sub> concentration treated (R<sup>2</sup>=0.93) (Reinert, 1984). It is very similar to the result of treating 0 – 1 μmol·mol<sup>-1</sup> NO<sub>2</sub> to sunflower and maize (Okano et al., 1985).

In order to find the actual absorption rate of NO<sub>2</sub> by lettuce, the amount of NO<sub>2</sub> dissolved in moist air was measured. Initial NO<sub>2</sub> concentration was 0.48 μmol·mol<sup>-1</sup>

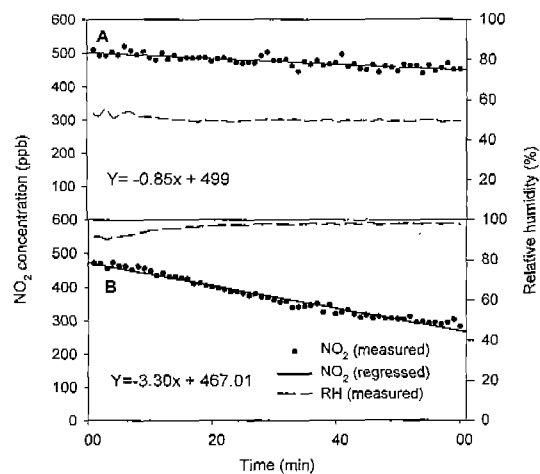


Fig. 4. Decrease of NO<sub>2</sub> concentration in the chamber at relative humidity treatments of 50 (A) and 95% (B). Initial NO<sub>2</sub> concentration in the chamber was 480 ppb.

in the chamber. Decreased slopes by moisture in the chamber without lettuce were  $0.85 \times 10^{-3}$  and  $3.30 \times 10^{-3} \mu\text{mol}\cdot\text{mol}^{-1}\cdot\text{min}^{-1}$  at relative humidities of 50 and 95%, respectively (Fig. 4). As relative humidity in the chamber increased,  $\text{NO}_2$  gas was dissolved more in moist air.

Since more  $\text{NO}_2$  is in a gaseous state at lower relative humidity,  $\text{NO}_2$  decrement measured at as low as 50% of relative humidity could be supposed to be the amount adhered to the acrylic plates. The quantities of  $\text{NO}_2$  absorbed by the lettuce, decreased by the moisture, and adhered to the acrylic plates were 72, 22, and 6%, respectively, when the total amount disappeared was regarded as 100%. From the data above, the  $\text{NO}_2$  absorption rate of lettuce could be estimated to be  $0.078 \mu\text{mol}\cdot\text{mol}^{-1}\cdot\text{m}^{-2}\cdot\text{min}^{-1}$  at  $0.45 \mu\text{mol}\cdot\text{mol}^{-1} \text{NO}_2$ , and lower at below  $0.45 \mu\text{mol}\cdot\text{mol}^{-1} \text{NO}_2$ .

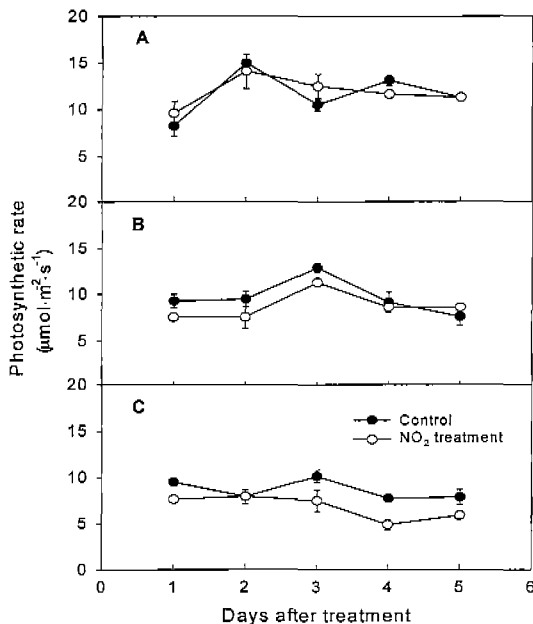
**Photosynthetic rate.** Photosynthetic rate of lettuce was measured for 5 days at 15:00. The photosynthetic rate at  $0.45 \mu\text{mol}\cdot\text{mol}^{-1} \text{NO}_2$  was lower than at the con-

trol at 4th and 5th days, but no significant difference was found at  $0.30 \mu\text{mol}\cdot\text{mol}^{-1} \text{NO}_2$  or less (Fig. 5). Similarly to our results, the growth of sunflower increased with time at  $0.2 \mu\text{mol}\cdot\text{mol}^{-1} \text{NO}_2$ , but decreased at  $0.5 \mu\text{mol}\cdot\text{mol}^{-1} \text{NO}_2$  (Okano et al., 1985). We supposed that the lettuce treated with  $0.45 \mu\text{mol}\cdot\text{mol}^{-1} \text{NO}_2$  would be a little injured as time passed. The decreasing tendency of photosynthetic rate with both  $\text{NO}_2$  treatment and the control in this experiment might be due to the increased water vapor pressure in the plant module.

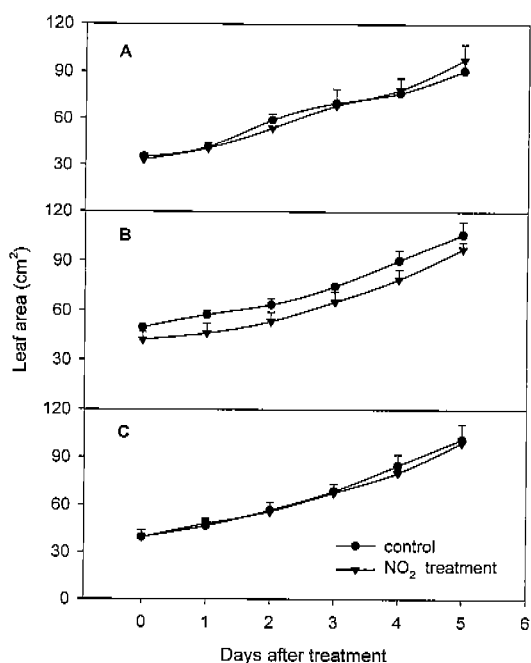
The plants contaminated by air pollutants had a decreased transpiration rate with generation of ABA, which is known to be involved in stomata closure. If transpiration rate decrease, the leaf temperature will increase. And white fleck, which is one of the typical symptoms of visible injury caused by  $\text{O}_3$  and  $\text{NO}_2$ , will appear by exchanging assimilation materials such as lipid and protein from old leaf (Shimizu et al., 1984). Injury on the plant in response to the treatment of pollutant gases such as  $\text{SO}_2$ ,  $\text{NO}_2$  and  $\text{O}_3$  depends on the exposure time and the concentration of the pollutant (Srivasiava et al., 1974). When plants are grown at high  $\text{NO}_2$  concentration, the plants gradually close the stomata to prevent gas exchange. If they do that, the transpiration and photosynthetic rates will become lower (Omasa and Onoe, 1985).

Stomatal conductance of leaves at 4th and 5th days under  $0.45 \mu\text{mol}\cdot\text{mol}^{-1} \text{NO}_2$  treatment also was lower than the control (data not shown). However, the respiration rate was not different between  $0.45 \mu\text{mol}\cdot\text{mol}^{-1} \text{NO}_2$  treatment and the control. The growth of lettuce, to some extents, was not affected by  $\text{NO}_2$  treatment with respect to the photosynthesis and respiration rates.

**Other growth factors.** The leaf area of the lettuce plants was not significantly different between  $\text{NO}_2$  treatment and the control during the 5 days (Fig. 6). Leaf areas were  $426$  and  $440 \text{ cm}^2$  at 5th day, respectively. However, the leaf areas from the image were  $97$  and  $99 \text{ cm}^2$  for treated and the control leaves, respectively. The difference between the destructive and non-destructive methods might be due to the two-dimensional images.



**Fig. 5.** Photosynthetic rate of lettuce under air exchange condition of 10 min-ON and 20 min-OFF between plant and human modules at  $\text{NO}_2$  treatments of 130 (A), 300 (B), and 450 ppb (C). Symbols and error bars represent the means of three replications and standard error, respectively.



**Fig. 6.** Change in leaf area of lettuce at NO<sub>2</sub> treatments of 130 (A), 300 (B), and 450 ppb (C). Symbols and error bars represent the means of three replications and standard errors, respectively.

There are limits to calculating the exact leaf area of the lettuce with the two-dimension images because the leaf of the lettuce grows at about 60° angle from the horizontal plane and lies one upon another. Color of an object can be described by several color coordinate systems. Some of the most popular systems are RGB (red, green, and blue), Hunter L a b, CIE L\* a\* b\*, CIE XYZ, CIE L\* u<sup>v</sup>: v\*, CIE Yxy, and CIE LCH (Abbott, 1999). L (lightness), a (green-yellow), and b (red-blue) value, and tension force used to see the quality of lettuce were not significantly different between 0.45 μmol·mol<sup>-1</sup> NO<sub>2</sub> treatment and the control ( $P < 0.05$ ) (Table 2). The reason for this result is that no visible injury was revealed between the treatments. After the experiment, no differences in leaf width, leaf length, leaf area and fresh weight of the plants also were found (Table 3).

With this result, the lettuce growth was not different between NO<sub>2</sub> treatment and the control and showed no visible injury in either condition. The lettuce in the plant module could purify NO<sub>2</sub> gas generated from the human

**Table 2.** Mean values of L, a, b and maximum values of tension force of leaves at 0.45 μmol·mol<sup>-1</sup> NO<sub>2</sub> treatment and control for 5 days.

| NO <sub>2</sub> concentration (μmol·mol <sup>-1</sup> ) | L <sup>x</sup> | a <sup>x</sup> | b <sup>x</sup> | Tension force (g) <sup>y</sup> |
|---|----------------|----------------|----------------|--------------------------------|
| Control   | 48.9 ±0.37     | -22.0 ±0.24    | 33.4 ±0.55     | 178.0±2.45                     |
| 0.45  | 49.6 ±0.45     | -22.3 ±0.20    | 34.2 ±0.56     | 179.5±2.68                     |

<sup>x</sup>Mean±SE, n=20.

<sup>y</sup>Max ±SE, n=20.

**Table 3.** Leaf width, length, weight, and area of lettuce grown at different NO<sub>2</sub> concentrations for 5 days.

| NO <sub>2</sub> concentration (μmol·mol <sup>-1</sup> ) | Leaf width (cm)       | Leaf length (cm) | Leaf weight (g) | Leaf area (cm <sup>2</sup> ) |
|---|-----------------------|------------------|-----------------|------------------------------|
| Control   | 8.2±0.20 <sup>x</sup> | 14.2±0.28        | 15.1±1.77       | 424±35.9                     |
| 0.15  | 8.4±0.10              | 14.7±0.39        | 14.4±0.76       | 413±12.1                     |
| Control   | 9.0±0.62              | 15.7±0.53        | 15.1±0.68       | 459±11.1                     |
| 0.30  | 8.6±0.65              | 15.0±0.56        | 13.8±1.43       | 413±25.6                     |
| Control   | 8.9±0.33              | 17.9±0.40        | 14.2±1.66       | 449±41.4                     |
| 0.45  | 9.3±0.12              | 16.7±0.20        | 14.6±0.29       | 452±9.9                      |

<sup>x</sup>Means±SE, n=3.

module. It is therefore concluded that the UPF can accomplish the function of plant production and ambient air purification at the same time.

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## 부분 밀폐형 식물생산시스템에서 상추의 $\text{NO}_2$ 흡수 및 생육반응

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### 적 요

인간거주 모듈과 식물생산 모듈로 구성된 밀폐형 식물생산시스템에서 시스템의 환경특성, 상추의  $\text{NO}_2$  흡수 및 생육반응이 조사되었다. 잎이 4-5개인 상추는 10 cm 직경의 포트에 정식된 후,  $\text{NO}_2$  농도가 제어되는 식물모듈에서 재배되었다.  $\text{NO}_2$ 가 포함된 인간 모듈의 공기는 다이어프램에 의하여 10분 ON, 20분 OFF 형태로 인간모듈과 식물모듈 사이로 순환되었다.  $\text{NO}_2$  농도가 0.13, 0.30 및 0.45  $\mu\text{mol} \cdot \text{mol}^{-1}$ 로 일정하게 유지되는 조건에서 10분 동안  $\text{NO}_2$ 는 각각 0.040, 0.109, and 0.149  $\mu\text{mol} \cdot \text{mol}^{-1}$  감소되었다. 상추의  $\text{NO}_2$  흡수율은 처리조건중 0.45  $\mu\text{mol} \cdot \text{mol}^{-1}$ 에서 0.078  $\mu\text{mol} \cdot \text{mol}^{-1} \cdot \text{m}^{-2} \cdot \text{min}^{-1}$ 로 추정되었으며, 이 경우에도 상추의 가시적인 장애는 나타나지 않았다. 정식후 4-5일째에  $\text{NO}_2$  0.45  $\mu\text{mol} \cdot \text{mol}^{-1}$ 에 노출된 상추의 광합성 속도는 노출되지 않은 대조구에 비하여 상대적으로 낮았지만, 호흡율에는 큰 변화는 없었다. 또한  $\text{NO}_2$  0.45  $\mu\text{mol} \cdot \text{mol}^{-1}$ 에 노출된 상추의 생육은 대조구에 비하여 엽폭, 엽장, 엽면적, 생체중, 건물중과 품질 등에서 큰 차이가 없었다.

주제어 : 식물생산, 공기정화,  $\text{CO}_2$  이용, 사무실, 도시형 식물공장