Dear Editor,

The recently published articles in the Iranian Journal of Biotechnology inspired us to write this short communication on nitrogen dioxide NO$_2$-philic plants (1).

To cope with environmental pollution, we thought (still think) that development of methods that mitigate pollution is an urgent issue, and that NO$_2$-philic plants that can grow with NO$_2$ as the sole nitrogen source are instrumental for the implementation of mitigation of air pollution.

The main point of this idea is that mitigation, rather than monitoring or observation, of environmental pollution is of primary importance. In other words, our point is that “plants that rely on pollution”, rather than “plants that are resistant to pollution”, are more meaningful to implement air remediation by plants. In the very beginning of this study, we attempted screening of such plants among naturally occurring plants (2). Screening study revealed that several plant species have a high ability to uptake and assimilate NO$_2$ (2). But it was not high enough for these species to grow with NO$_2$ as the sole nitrogen source (3). We also performed genetic engineering of nitrite reductase (NiR), which is a second enzyme of nitrate reduction pathway and a key enzyme of assimilation of NO$_2$. We have produced genetically modified plants of Arabidopsis thaliana (a model plant species) (4) and Rhaphiolepis umbellata (a roadside tree species) (5) that significantly bear higher NiR activities compared with non-transformed control plants. However, their ability to uptake NO$_2$ was less than two times higher than that of non-transformed plants, and again this was not enough for them to be NO$_2$-philic (3).

We also investigated the growth of plants in the presence of NO$_2$ as the sole nitrogen source, using Nicotiana plumbaginifolia as a test species. The Plants were grown in the complete absence of root nitrogen under exposure to 0, 0.15, 1.0 and 4.0 ppm NO$_2$ and natural light at 22°C for 9–13 weeks. Control plants were grown with 10 mM KNO$_3$ in ambient air containing < 0.05 ppm NO$_2$. Plants grown at 0 ppm NO$_2$ died after 6 weeks. The relative growth rate in biomass (RGR) of plants grown at 1.0 ppm NO$_2$ and that of those grown at 4.0 ppm NO$_2$ were very similar to the RGR of control plants (about 0.1.day$^{-1}$). However, plants grown at 1.0 and 4.0 ppm NO$_2$ exhibited some morphological abnormalities in leaves (Figure 1, Nakagawa, Takahashi and Morikawa, unpublished results), that indicates the toxic effects of NO$_2$. Interestingly, plants grow at 0.15 ppm NO$_2$ did not show such damages and grew normally up to 13 weeks. Furthermore, we also noticed that NO$_2$ at 0.15 ppm is somewhat stimulatory for the growth of plants.

![Figure 1. An Attempt to Produce a NO$_2$-Philic Plant](image)

Five-week-old Nicotiana plumbaginifolia plants that were grown either with 1 ppm NO$_2$ as the sole nitrogen source (right), or with 10 mM KNO$_3$ in ambient air (<0.05 ppm NO$_2$) (left).

Clearly, the effect of NO$_2$ on plant growth changes as a function of its concentrations; it was inhibitory at high concentrations but stimulatory at low concentrations. Thus, clarification of these toxic and stimulatory effects of NO$_2$ is necessary for the selection of NO$_2$-philic plants as bioremediation systems.
NO₂ may provide an important key to produce NO₂-philic plants. The investigation on the stimulatory effect of NO₂ led to the finding of a plant-hormone like effect of NO₂ on the growth and development of plants (6). Ambient concentrations of NO₂ stimulated growth and biomass production of plants that were well fed root nitrogen. Uptake per plant of nutrients was also stimulated. Carbon and nitrogen metabolisms also were stimulated by the presence of NO₂. This effect was confirmed in several plant species including Arabidopsis and horticultural species (3).

The investigation on the toxic effect of NO₂ led us to the investigation on metabolic fate of NO₂ in plants, and to the finding of novel oxidized nitrogen compounds including nitro- and nitroso-organic compounds formed from NO₂ (7, 8). We also found that fumigation of Arabidopsis plants with a high concentration (4 - 40 ppm) of NO₂ induces nitration of specific proteins involved in photosystem II in chloroplasts (Takahashi and Morikawa, submitted). Protein tyrosine nitration is an important posttranslational protein modification, and thus protein nitration induced by NO₂ may be of important biological relevance. Currently, which one(s) of such nitro- and nitroso-organic compounds including nitrated proteins is responsible to the toxic effect of NO₂ has yet to be identified. Nonetheless, the abovementioned results on these two effects of NO₂ implicate that exogenous NO₂ plays a distinct role in cellular signaling. Clarification of the mechanisms and identification of genes involved in both toxic and stimulatory effects of NO₂ will pave the way for production of NO₂-philic plants for air remediation.

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Authors’ Contribution

Misa Takahashi developed the original idea and the protocol, abstracted and analyzed data, wrote the manuscript, and are guarantor. Hiromichi Morikawa developed the idea, abstracted, and wrote the manuscript together with M.T. Makiko Nakagawa mainly contributed to the analysis of data.

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No relevant financial interests or financial conflicts within the past 5 years and for the foreseeable future.

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