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ORIGINAL PAPER

Does the increase in ambient CO_2 concentration elevate allergy risks posed by oak pollen?

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Abstract

Oak pollen is a major respiratory allergen in Korea, and the distribution of oak trees is expected to increase by ecological succession and climate change. One of the drivers of climate change is increasing CO_2, which is also known to amplify the allergy risk of weed pollen by inducing elevated allergenic protein content. However, the impact of CO_2 concentration on tree pollen is not clearly understood due to the experimental difficulties in carrying out extended CO_2 treatment. To study the response of pollen production of sawtooth oak trees (*Quercus acutissima*) to elevated levels of ambient CO_2, three open-top chambers at the National Institute of Forest Science in Suwon, Korea were utilized with daytime (8 am–6 pm) CO_2 concentrations of ambient (× 1.0, ~ 400 ppm), × 1.4 (~ 560 ppm), and × 1.8 (~ 720 ppm) treatments. Each chamber had three sawtooth oak trees planted in September 2009. One or two trees per chamber matured to bloom in 2016. Five to six catkins were selected per tree and polyethylene bags were attached to collect pollen grains. The total number of catkins per tree was counted and the number and weight of pollen grains per catkin were measured. Oak allergen—*Que a* 1 (Allergon Co., Uppsala, Sweden)—was extracted and purified to make an ELISA kit by which the antigen levels in the pollen samples were quantified. Total pollen counts per tree of the × 1.4 and × 1.8 treatments showed significant increase of 353 and 1299%, respectively, from the × 1.0 treatment ($p < 0.001$). Allergenic protein contents at the × 1.4 and × 1.8 treatments also showed significant increase of 12 and 11%, respectively ($p = 0.011$). The × 1.8 treatment induced significant difference from the × 1.0 treatment in terms of pollen production and allergenic protein content, whereas the × 1.4 treatment showed mixed significance. In summary, the oak trees under the elevated CO_2 levels, which are expected in the changing climate, produced significantly higher amount of pollen and allergenic protein than under the present air conditions.

Keywords Open-top chamber · CO_2 enrichment · Allergenicity · *Que a* 1 · Pollen production · Oak pollen

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Introduction

Climate change is induced by an increase in greenhouse gases, and carbon dioxide is one of the important gases that have a greenhouse effect. The monthly mean CO_2 concentration in May 2016 observed at the Mauna Loa Observatory (MLO), Hawaii, reached 407.70 ppm, and the annual mean value in 2016 was 404.21 ppm (see ftp://aftp.cmdl.noaa.gov/products/trends/co2/co2_annmean_mlo.txt). This is an increase of more than 120 ppm from the pre-industrial concentration of 280 ppm. The concentration has increased 1.55 ppm $year^{-1}$ since the start of the observation at MLO in 1959, and it has accelerated to 2.16 ppm $year^{-1}$ since 2000 (NOAA/ESRL 2017). Because of the effects of increased greenhouse gases, the mean global air temperature increased 0.85 °C between

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1880 and 2012 [Intergovernmental Panel on Climate Change (IPCC) 2013]. The increase of CO_2 as well as the air temperature in temperate regions, where the growing period is limited by temperature, can induce increased carbon uptake and prolonged growing periods. In general, plants are expected to grow more rapidly during the period of global warming.

There have been several reports on the potential increase of risks by allergenic pollen in relation to the temperature increase by climate change. Allergenic plants subjected to warmer temperatures may grow for longer periods of time, thus producing more pollen grains and increasing the length of the pollen season (Rogers et al. 2006). The flowering period or pollen production of oak species has been modeled based on growing degree hour (GDH) and observational data (García-Mozo et al. 2002; Zhang et al. 2014). The relationship between temperature, precipitation, and oak pollen season length were utilized in predicting the oak pollen season and its impact in the future (Anenberg et al. 2017). The authors estimated increases in the length of the pollen season per 20-year period of as much as 0.5–2% for RCP 4.5 and 1.4–2.9% for RCP 8.5 scenarios. They also estimated the increased growth rate of oak trees to be 1–5% for RCP 4.5 and 2–8% for RCP 8.5. Temperate oaks are expected to have an expedited start of pollen season, increased yearly pollen production, and higher peak pollen concentration as a result of climate change (Zhang et al. 2014).

The increase in CO_2 concentration can also affect the pollen production directly. For example, ragweed yielded more pollen grains per plant in CO_2-enriched growth chambers (Ziska and Caulfield 2000). However, the direct impact of CO_2 increase on tree pollen has not been demonstrated yet (see Fann et al. 2016). The amount of allergen protein in a pollen grain can increase because of increased CO_2 concentration, such that the same number of pollen grains differ in their allergenicity (Beggs 2004; Reid and Gamble 2009; Sheffield et al. 2011a). Previous studies revealed an increased number and allergenic protein level of pollen grains of weed species, such as common ragweed and Timothy grass under high CO_2 concentrations (Albertine et al. 2014; Ziska and Caulfield 2000; Singer et al. 2005). However, the impact of increased CO_2 on tree species has not been reported much because it requires longer periods of CO_2 exposure and thus more resources.

A quantitative increase in allergenic pollen will affect patients with allergic rhinitis, who are an estimated 10–30% of the global population, and 300 million patients with asthma (Schmidt 2016). Airborne pollen was found to escalate the sale of allergy medicine by 28.7% after 2 days of high pollen concentrations in New York City (Sheffield et al. 2011a). Generally, allergy-related disorders are anticipated to increase in children especially (Sheffield et al. 2011b). To develop a proper adaptation plan, it would be valuable to provide quantitative changes in the amount and allergenicity of pollens in future climate-change scenarios.

Sawtooth oak (*Quercus acutissima*) is one of the representative oak trees of Korea, along with *Q. aliena*, *Q. dentata*, *Q. mongolica*, *Q. serrata*, and *Q. variabilis*. It is one of the hardwood species used for the production of lumber because it grows up to 25 m in height and 1.5 m in diameter. In Korea, the annual plantation area for *Q. acutissima* is about 1500 ha. Oak trees are expected to replace the present red pine (*Pinus densiflora*) forests because they can be regenerated by seeds, especially in steep and infertile sites; they can also be easily regenerated by sprouts after disturbances, such as forest fires. Acorns are useful as a food resource not only for wildlife but also for people, and so oak is a good candidate for use in North Korean reforestation in the future. In this study, the impact of increased CO_2 concentration, one of the drivers of climate change, on the exposure risk of allergenic oak pollen is quantified.

Materials and methods

Open-top chamber

CO_2 enrichment experiments used open-top chambers (OTCs) to provide stable long-term exposure to elevated CO_2 concentrations with near-natural conditions, such as air temperature, relative humidity, and light. To study the response of oak pollen to varying CO_2 concentrations, OTCs located at the National Institute of Forest Science in Suwon, Korea (37° 15′ 04″ N, 136° 57′ 59″ E), were utilized (Fig. 1). The chambers are 10 m in diameter and 7 m in height (Lee et al. 2012). Three treatment levels of CO_2 concentration were applied to the OTCs: ambient (× 1.0, ~ 400 ppm), × 1.4 (~ 560 ppm), and × 1.8 (~ 720 ppm). A CO_2 concentration of × 1.4 is expected to be reached in the 2040s according to the RCP 8.5 scenario and in the 2060s by the RCP 4.5 scenario and a CO_2 concentration of × 1.8 is expected to be reached in the 2050s by the RCP 8.5 scenario and in the 2090s by the RCP 6.0 scenario (IPCC 2013). The OTCs in the current study were exposed to the respective CO_2 concentrations from 8 am to 6 pm from April to November, when photosynthesis and evapotranspiration take place actively. The OTCs are fully operational since September 2009. The performance of the chambers was evaluated by Lee et al. in 2011 (Lee et al. 2012) and following years. During the 2012–2014 period, the annual mean daytime CO_2 concentrations (mean ± standard deviation, %) were maintained at 110 ± 7.8%, 98 ± 2.4%, and 89 ± 3.1% of the target concentrations of ambient (× 1.0), × 1.4, and × 1.8, respectively. Although the air temperature inside was 1.2–2.0 °C higher than that of the outside, the temperature difference among the OTCs was less than 0.2 °C.

The yearly mean air temperature and total precipitation at nearby weather station (KMA Suwon, station #119) from

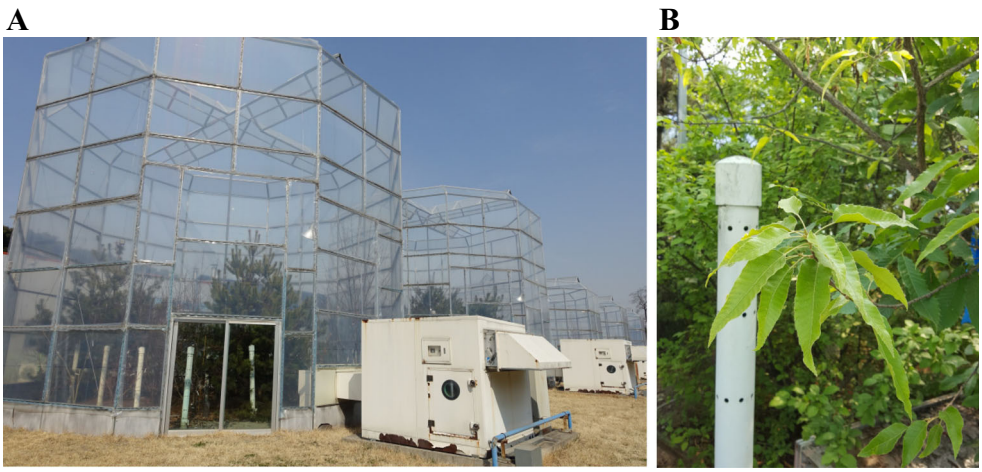

Fig. 1 **a** OTCs at the National Institute of Forest Science in Suwon, Korea and **b** its internal view showing a CO_2 injection nozzle and plants

2009 to 2016 were 12.7 ± 0.6 °C and 1347.5 ± 410.9 mm, respectively and those of climatology (1981–2010) were 12.0 °C and 1312.3 mm, respectively.

Oak plants

At the beginning of the OTC operation in 2009, soil to a depth of 1 m was replaced with soil from a forest to establish the same soil conditions among the chambers (Lee et al. 2012). Tree species of a representative Korean temperate forest were selected and planted in the OTCs (× 1.0, × 1.4, × 1.8) in September 2009. The planted species were *P. densiflora*, *Fraxinus rhychophylla*, *Sorbus alnifolia*, *Acer pseudosieboldianum*, *Crataegus pinnatifida*, and *Q. acutissima* (Ryu et al. 2014). Among them, *Q. acutissima*, *F. rhychophylla*, *A. pseudosieboldianum*, and *P. densiflora* are known to produce allergenic pollen. *Q. acutissima* was selected for further analysis of its allergenicity considering its natural abundance in Korea and its relative impact on respiratory allergies (Oh et al. 2012)

All of the oak plants (*Q. acutissima*) in this study were the same clone obtained from the National Forest Seed and Variety Center in 2008 (Seo et al. 2014). Out of three sawtooth oak trees planted, one (× 1.0) or two (× 1.4 and × 1.8) trees per chamber matured to bloom in 2016. The × 1.0 chamber had no oak bloom in 2017, and so the allergenicity comparison between the CO_2 treatments was conducted in 2016 only. The trunk diameter 5 cm above ground level and the height of the oak trees surveyed in May 2016 are shown in Table 1.

Pollen collection

A polyethylene bag was placed over each branch with 5–6 catkins (male flowers) from April 19 through May 15. After placement of the bag, the slit was taped. The bags were tapped gently each day until all catkins completely dehisced. Pollen grains were collected from *Q. acutissima* in the OTCs during the peak of the flowering season, on April 19 and 26, 2016. The number and weight of pollen grains from each catkin and the diameters of the grains were actually counted and measured. A hemacytometer microscope was used to count the number of pollen grains. Also, the number of catkins of each tree was observed to estimate total number of pollen grains per tree. Following pollen collection, all pollen collection bags from each mature plant for a given CO_2 treatment were kept frozen at − 70 °C until further analysis.

Protein extraction of pollen

After pollen removal from the catkins, the pollen was placed into microtubes filled with 50 ml of ethanol until protein extraction. Samples of pollen (10 mg) were suspended in Coca's solution—200 μL of 0.5% NaCl/0.25% $NaHCO_3$, *w*/*v*—in water. Pollen suspensions were sonicated at 0 °C with a Fisher Model 550 Sonic dismembrator (Fisher Scientific, Pittsburgh, PA) fitted with a micro-tip (3.2 mm diameter). The sonicated suspensions were kept on ice with occasional mixing for 2 h. Following the extraction period, particulates were removed by centrifugation for 10 min at 13,000 *g* and 6 °C. The supernatant fluid was carefully withdrawn from the pellet, and the volume of each extract measured. An equal volume of 50% (*w*/*v*) glycerol was added and the extracts were kept at − 20 °C. The Bradford method (Bradford 1976) was used for protein quantification.

Table 1 Mean and standard deviation of trunk diameter 5 cm above ground level and height of the oak trees observed in May 2016 at each OTC

CO_2 concentration in chamber	Diameter (mm)	Height (cm)
× 1.0 (control = ambient = 400 ppm)	38.9 ± 7.6	355.0 ± 106.1
× 1.4 (560 ppm)	38.5 ± 1.4	310.0 ± 14.1
× 1.8 (720 ppm)	41.5 ± 8.7	373.3 ± 64.3

Immunochemical quantification of *Que a* 1

The amounts of protein from the extraction processes were determined as the concentration of *Que a* 1 allergen (Allergon Co., Uppsala, Sweden) by an enzyme-linked immunosorbent assay (ELISA). Also, the pollen protein extracts were diluted to initial concentrations of 2.5×10^{-4} mg protein/ml in 0.05 M sodium carbonate buffer, pH 9.6; linear dilutions over a tenfold range were prepared in the same buffer as for ELISA analysis. Bovine serum albumin (Sigma-Aldrich, St Louis, MO) was used as a blocking agent to prevent non-specific protein binding. Plates were developed with the addition of 1 mM H_2O_2 and 2 mM 2.2′-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid, Sigma-Aldrich) in 0.2 M phosphate–0.10 M citrate (pH 5.0) buffer. The rate of color development at 450 nm was measured with a 96-well plate reader (Bio-Rad Laboratories, Hercules, CA). The concentration of allergen in a given sample was measured using arbitrary ELISA units. The final results are reported as ng protein/ml of pollen extract.

Statistical analysis

Differences in CO_2 treatment groups were compared with one-way ANOVA along with a post-hoc Tukey's test for the treatment groups (significance level of $p \leq 0.05$). The statistical analyses were conducted using a IBM SPSS Statistics ver. 23.0 (IBM Co., Armonk, NY, USA).

Results

The number and weight of pollen grains per male flower (catkin) showed clear difference with the CO_2 treatment ($p = 0.045$ and $p = 0.003$, Fig. 2). However, the increasing trend was not clear with the × 1.4 treatment: insignificant with either × 1.0 or × 1.8 (Tukey groupings, Fig. 2). The number of pollen grains per tree was estimated by multiplying the number of pollen grains per catkin by the number of observed catkins per tree. The number of total pollen grains per plant was significantly different with the CO_2 treatment ($p < 0.001$, Fig. 3a). The mean diameter of the pollen grains showed increasing trend with CO_2 concentration ($p < 0.001$, Fig. 3b). Obviously, higher CO_2 concentrations enhanced the number of pollen grains, weight, and size as well. The increasing number of pollen grains per tree with increasing CO_2 concentration was steeper than that per catkin because of the higher number of catkins under higher CO_2 concentrations.

The mean concentrations of allergen (*Que a* 1) in the pollen extracts between × 1.0, × 1.4, and × 1.8 were determined to be 419.7 ± 27.7, 472.2 ± 26.4, and 467.2 ± 30.7 ng/ml, respectively (Table 2). The mean concentrations among the × 1.0, × 1.4, and × 1.8 treatments were significantly different ($p = 0.011$), but those between the × 1.4 and × 1.8 treatments showed no significance (Fig. 4).

The impacts of elevated CO_2 concentration on pollen production are summarized in Table 3. The ratios are calculated based on the × 1.0 observations, and they can be converted to percent increases by subtracting 1.0 then multiplying by 100. All of the ratios of × 1.8 are significantly different from those of × 1.0 (see Tukey's test groupings, Figs. 2, 3, and 4). The present results demonstrate that the oak trees under 40 and 80% higher CO_2 concentration than ambient air produced 20 and 82% more pollen grains per catkin and 353 and 1299% more per tree with 11% more allergenic protein content at this early stage of reproductive life.

Discussion

Airborne pollen is one of the major causes of respiratory allergies. Allergenic protein in the pollen is the causal agent of

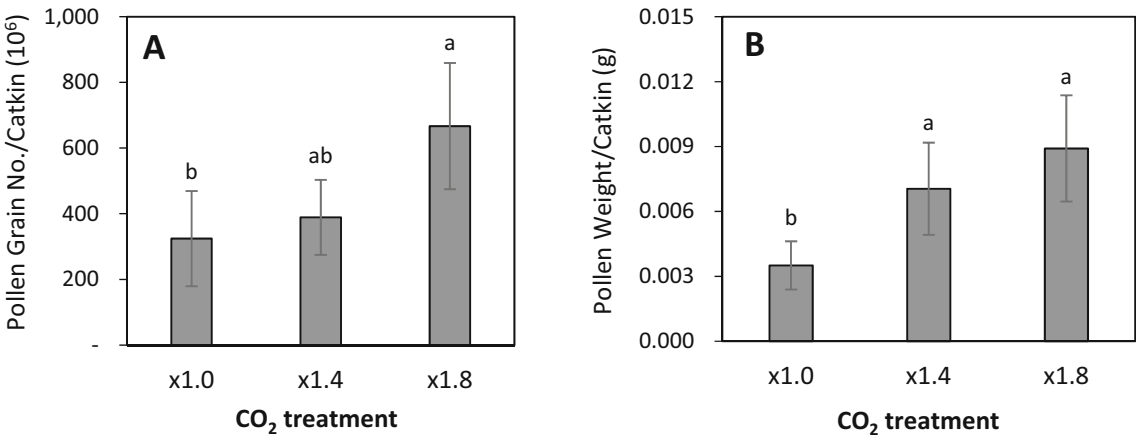

Fig. 2 **a** Number and **b** weight of pollen grains per male flower (catkin) from the CO_2 OTCs × 1.0, × 1.4, and × 1.8 CO_2 concentrations. × 1.0, × 1.4, and × 1.8 mean 400, 560, and 720 ppm CO_2 concentrations, respectively. Different lowercase letters on the bars indicate significant differences among treatments in OTCs at 5% significance level by Tukey's test. Data represent mean ± standard deviation

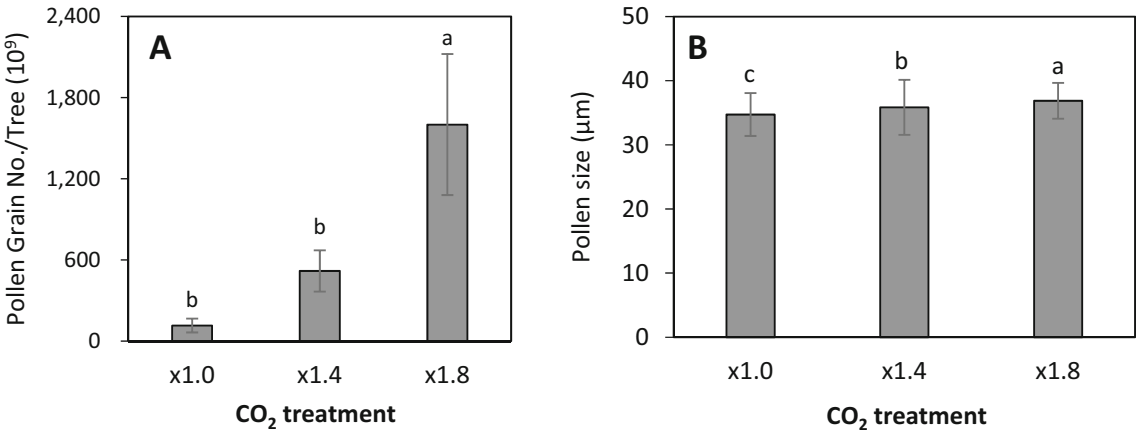

Fig. 3 **a** Total number of pollen grains and **b** mean diameter of the pollen grains collected from the CO_2 OTCs × 1.0, × 1.4, and × 1.8 CO_2 concentrations. × 1.0, × 1.4, and × 1.8 mean 400, 560, and 720 ppm CO_2 concentrations, respectively. Different lowercase letters on the bars indicate significant differences among treatments in OTCs at 5% significance level by Tukey's test. Data represent mean ± standard deviation

the allergy symptoms so that the amount of the protein is expected to determine the severity of pollen allergy. The protein production in this study was quantified as two steps: number of pollen grains in a tree and allergenic protein content in a pollen grain.

The number of pollen grains per tree increased by 353% (× 1.4) and 1299% (× 1.8) from the × 1.0 treatment. It was determined by the number of catkins per branch, the number of branches, and the number of pollen grains per catkin, which increased by 20% (× 1.4) and 82% (× 1.8). Because the oak plants at higher CO_2 treatments grew more vigorously during the 6 years (Table 1), they were able to produce many more pollen grains than the control. Weeds species, such as common ragweed and Timothy grass also showed increased pollen production per plant with doubled CO_2 concentrations but at a lower extent: 320% increase for common ragweed (CO_2 increase from 280 to 600 ppm) (Ziska et al. 2000) and 202% increase for Timothy grass (CO_2 increase from 400 to 800 ppm) (Albertine et al. 2014).

On the other hand, the allergenic protein content of the oak species studied here increased by 12% (× 1.4) and 11% (× 1.8) only. The common ragweed and Timothy grass showed 91 and 190% increase, respectively, in their allergenic protein content by the doubled CO_2 experiments (Albertine et al. 2014).

Because the oak seedlings we planted just entered the reproductive stage with sufficient space for branching, the impact of elevated CO_2 concentration on pollen productivity might have been over-estimated than in an old forest, where space for new branching is limited. Nevertheless, the higher productivity in their early life stages would be an important characteristic if the old forest was disturbed or if the trees are adapted/introduced to a new area. The overall exposure risk, which can be assessed as multiplying number of pollen grains per tree by allergen content, can increase higher with oak trees than weeds species because of the big increase of oak pollen production at elevated CO_2 concentrations.

Application to future climate conditions

In order to assess the future environment on pollen allergy, it is necessary to apply the present results to climate change scenarios. The influence of climate change on pollen allergy can be quantified in four categories: (1) allergenicity or allergenic protein content in each pollen grain, (2) productivity of the pollen grains per unit plant, (3) length of the pollen season, and (4) geographic distribution of the allergenic plant. The first two aspects were experimentally quantified in this study.

Allergenicity

Oak trees require a lot of space and the time from seedling to flowering is long, and so it has been difficult to experiment on the impact of CO_2 on the allergenicity of oak pollen. Sawtooth oak is widely distributed in eastern Asia and the eastern USA (Whittemore 2004), and cross-pollination of oak species with geographical overlapping is observed in general (Boavida et al. 2001; Kremer and Petit 1993). *Que a* 1, the major allergen from *Q. alba*, has shown strong cross-reactivity to the pollen of *Q. mongolica*, a dominant oak species in Korea (Lee et al. 2017). We therefore expect that the sawtooth oak shares common characteristics of genetics of pollen allergenicity and growth with other oaks in temperate climates. In this study, the expected increase in allergenic protein per unit weight of pollen under elevated CO_2 concentrations was revealed (Fig. 4). This information will reduce the uncertainty of pollen risk in the future and provide a better adaptation strategy to climate change.

Table 2 Allergen contents of oak pollen determined as ng protein/ml of pollen extract according to the CO_2 treatment

Sampling bag	Chamber treatment		
	× 1.0	× 1.4	× 1.8
1	431.8	462.5	425.2
2	392.9	481.1	438.8
3	393.2	457.5	467.2
4	401.1	498.2	492.3
5	441.4	501.1	474.2
6	457.9	432.5	505.4
Mean	419.7	472.2	467.2
SD	27.7	26.4	30.7

Productivity

The diameter and height of the trees in this study (Table 1) showed accelerated growth under higher CO_2 concentrations, which conforms to other studies. For example, Kuribayashi et al. (2017) modeled the ecological changes of oak and birch forests based on ecophysiological processes. Canopy phenology and carbon budget were simulated using various climate-change scenarios. The authors stated that by 2085, early budding and delayed shedding are expected to increase the growing period by approximately 10 days, and gross primary production (GPP), ecosystem respiration (ER), and net ecosystem production (NEP) are predicted to increase by 25.2, 23.7, and 35.4%, respectively. Therefore, the oak ecosystem will act as a stronger CO_2 sink and grow more vigorously as a result of increased CO_2 concentration and the growing period will be prolonged because of the increased temperature (Kuribayashi et al. 2017). Furthermore, the pollen productivity changes in this study will provide more useful information in direct relation to allergenicity.

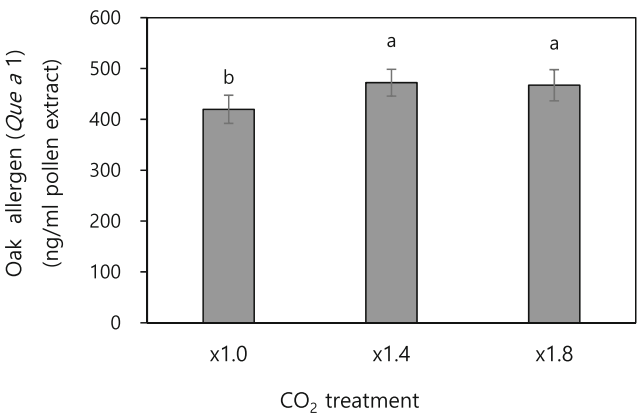

Fig. 4 Allergenic protein contents of the oak pollen according to the CO_2 treatment. Different lowercase letters on the bars indicate significant differences among treatments in OTCs at 5% significant level by Tukey's test. Data represent mean ± standard deviation

Season length

Phenological changes, such as expedited flowering and delayed defoliation may not be directly related to the allergenicity of oak pollen. However, in the other experiments with the same OTCs, Seo et al. (2014) found that the dates of budburst and unfolding of oak were expedited more by air temperature than by CO_2 concentration, whereas coloring and shedding dates were delayed by elevated physiological activities due to higher CO_2 concentrations (Seo et al. 2014). The longer growing period, induced by higher air temperature along with higher CO_2 concentration, might cause more yearly growth and subsequently more flowers. These compound effects of temperature and CO_2 need more investigation. Based on the historical observation of oak trees, an earlier start to pollen season and the production of more pollen are expected in the future (Anenberg et al. 2017, Jeong et al. 2012).

Geographic distribution

Pollen production is largely dependent upon vegetation area, and thus changes in spatial suitability of allergenic species are important in assessing the impact of climate change. There are several ecological studies regarding the spatial changes of allergenic pollen-producing species. In England, long-term analysis over 30 years of the temporal changes in grass pollen since 1962 revealed that the increase or decrease of pollen was different between different regions and that it depended on the variation of grassland area within a 40-km radius (Emberlin et al. 1999). In North America, it was predicted that the higher allergenic trees, such as oak and hickory would occupy larger areas as compared to less allergenic trees, such as pine, spruce, and fir trees due to climate change (Iverson et al. 2008). Oak is a native tree of East Asia, including Korea, and it is expected to dominate over other species during the vegetation transition (Choung et al. 2004; Shao 1996), so that its distribution will be broader. Overall, as the geographic distribution is predicted to increase in the future; the number of male flowers and pollen grains are also expected to increase. By developing phenological models based on weather conditions, such as air temperature, one can assess future oak species distribution and length of the pollen season under climate change scenarios and then quantify the exposure risks by the allergenic pollen based on this study.

Limitations of the OTC experiment

OTCs have intrinsic problems of elevated temperature, reduced wind speed, and probably low rainfall. Temperature effects on the speed of plant growth are well-known. Even though the wind speed inside an OTC is very low, the difference is generally ignored in CO_2 experiments. However, the amount and/or maturity of the oak pollen, including the

Table 3 Number of pollen grains per catkin, weight of pollen per catkin, number of pollen grains per tree, pollen size, and allergenic protein content and their ratio to the × 1.0 treatment. The *p* values from the one-way ANOVA are also indicated

Treatment	Grains/catkin		Weight/catkin		Grains/tree		Size		Allergen content	
	(10^6)	Ratio	(mg)	Ratio	(10^9)	Ratio	(μm)	Ratio	(ng/ml)	Ratio
× 1.0	324.2	1.00	3.50	1.00	114.4	1.00	34.7	1.00	419.7	1.00
× 1.4	388.8	1.20	7.05	2.01	518.3	4.53	35.8	1.03	472.2	1.12
× 1.8	590.2	1.82	8.91	2.54	1600.7	13.99	36.9	1.06	467.2	1.11
p value	0.045		0.003		< 0.001		< 0.001		0.011	

diameters of the grains, may have been affected by this low wind speed inside the OTCs because oak produces large quantity of anemophilous pollen and releases mature pollen grains by wind. Partial interception of rainfall by the OTC structure, along with the elevated temperature, may have induced more evapotranspiration and less soil moisture. Insufficient soil moisture can close stomata and then stop photosynthesis and growth (Moser et al. 2017). Further study is needed to assess the respective and combined impacts of various micrometeorological conditions on pollen production. The study by Bianchi et al. (1959) on the impact of temperature on anther formation and that of humidity on the opening of pollen sacs of ragweed is a good example.

Applicability to other pollens

The number and weight of oak pollen grains from the CO_2-enriched OTCs (× 1.4, × 1.8) per plant and per catkin increased from the ambient CO_2 OTC (× 1.0). Although there are few reports on the response of tree pollens to CO_2 concentrations, these results are supported by those of other studies on weed species. Wayne et al. (2002) and Ziska and Caulfield (2000) reported that ragweed produced 60–90% more pollen with high concentrations of CO_2 (700 or 600 ppm). Rogers et al. (2006) reported faster growth of spikes as a result of high CO_2 concentrations over a limited period of time. In general, CO_2 enrichment makes weeds grow faster so that they produce more pollen grains (Ackerly and Bazzaz 1995; Deng and Woodward 1998). The production of oak allergen also increased, similar to the increase of *Amb a* 1 from common ragweed with higher CO_2 concentrations (Singer et al. 2005). Ziska and Beggs (2012) suggested that the complex interaction of CO_2 concentration and temperature on pollen production and allergenicity makes the overall risk assessment of tree pollens a good challenge. Climate change may induce changes in the ecological suitability of allergenic plants, which may result in new allergy symptoms in groups with no previous problems (Reid and Gamble 2009). As a final stage, we will have to determine dose-response relationships between the allergenic proteins and allergy symptoms to advance the current results to more useful clinical prediction models on allergy risks in future climate conditions.

Conclusion

Oak trees that bloomed in OTCs in 2016 were exposed to elevated CO_2 concentrations for more than 6 years. The accumulated effects of the CO_2 treatments resulted in 353% (× 1.4) and 1299% (× 1.8) increases of yearly pollen production (number of grains per tree). The allergen protein *Que a* 1 also increased by 11% due to the increased CO_2, which is one of the major drivers of climate change. Allergy risk by oak pollen is a function of number of pollen grains and the allergenicity of the pollen proteins. From this study, the total exposure risk is predicted to increase in the future, and the results can be applied to a strategic plan for climate change for more effective adaptation.

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