Elevated CO_2 causes earlier flowering in an alpine medicinal herb *Aconitum heterophyllum* Wall.

Flowering is a crucial stage in plant life, and any kind of alteration in flowering time can bring about changes in the species, community as well as ecosystem levels[1]. An understanding of the responses of phenological events and flowering to global change factors, e.g. elevated atmospheric carbon dioxide (CO_2) levels, is therefore essential to determine the implications of these changes on natural as well as agricultural ecosystems.

Alpine plant species are dominated by perennial life forms and are expectedly more sensitive to climate change than other lowland species due to limited growing season and low ambient partial pressure of CO_2, which may change their relative abundance[2]. *Aconitum heterophyllum* Wall. (Ranunculaceae) is an important native medicinal plant of Western–Central Himalaya, distributed between 3000 and 4000 m amsl. The International Union for Conservation of Nature and Natural Resources considers this species as critically endangered[3,4]. Only a few studies have been carried out in the alpine regions to understand the impact of elevated CO_2 (ECO_2) on vegetation, although the flowering pattern of alpine species under ECO_2 has not been explored previously[5–8].

The present study was conducted at Tungnath (N30°14′, E79°13′, 3600 m amsl), Uttarakhand, India using open-top chambers (OTCs), controlled by supervisory control and data acquisition (SCADA) automatic system (Figure 1). Seed-grown plants of *A. heterophyllum* were treated with elevated (650 ppm) level of CO_2 and tested against ambient CO_2 (400 ppm) for three growing seasons (2017–2019). At Tungnath, the temperature remains above 5°C during the middle of May to the first week of October, which is considered favourable for growth of alpine plant species[9]. Under ECO_2, plant height and leaf area were comparatively lower than ambient CO_2 (ACO_2); this may be due to decreased growth rate under ECO_2. Under ECO_2, flower-bud initiation took place in the second week of August with the first flower opening on 21 August, while under ACO_2 flower initiation started during the fourth week of August with the first flower opening on 5 September in the third growing season (2019). The flowering time under ECO_2 was advanced by 17–20 days compared to that reported by Nautiyal *et al.*[10]. The mature fruiting bodies (pods) were $26.3 \pm 2.6\%$ smaller in plants grown under ECO_2. The pod length under ambient CO_2 was 19.4 ± 1.8 mm, while under ECO_2 it was 15.3 ± 2.0 mm. The plants grown under ECO_2 showed decreased nitrogen content in leaves (–34%) and roots (–19%).

An earlier flowering response under ECO_2 has been reported in several studies on lowland species, and certain species expressing expedited flowering reach the minimal size required for early floral initiation as a result of an amplified growth rate under elevated CO_2 levels[11]. An early flowering in *A. heterophyllum* could be responsible for earlier senescence as observed in the present study, since in most species reproductive phase marks the beginning of senescence[12]. Altered flowering in alpine conditions may change the phenophases in the next generation and an earlier flowering followed by reduced fruiting bodies may be the result of decreased plant N, which plays an important role during fruit-setting. ECO_2 may affect plant

Figure 1. The installed open-top chambers at Tungnath (3500 m amsl) to study the impact of climate change on alpine plant species.

competence by altering its flowering time and size of the plant at the time of flowering, influencing the amount of available resources[13]. Stearns[14] also postulated that any alterations in the flowering time of perennial species may affect the availability of nutrient resources required for the production of seeds in the course of peculiar reproductive events. The ECO_2 causes accumulation of sugars in plants, but due to some limitation these are not transported to the sink and these non-structural carbohydrates may lead to a decrease in total N content due to dilution effect[15], further hampering plant growth.

A long-term exposure to ECO_2 levels can affect multiple facets of plant growth and metabolism by influencing nutrient availability in plants. In the present study, it has been found that ECO_2 levels induce an early flowering response followed by reduced reproductive growth and early senescence. A lack of long-term studies on the effects of rising atmospheric CO_2 levels on different types of alpine vegetation widens the gap for understanding the exact nature of adaptability in these ecosystems. Therefore, further studies are needed in these fields for better understanding the impact of climate change on alpine plants, which may be useful in formulating policies for the conservation of these species.

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