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Pollen long-distance transport associated with symptoms in pollen allergics on the German Alps: An old story with a new ending?

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HIGHLIGHTS

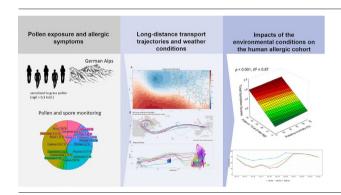
- Pollen exposure and allergic symptoms were monitored on low-vegetation German Alps.
- The possible origin of airborne pollen types was identified using back trajectory modelling.
- We found >1000 pollen grains m⁻³ of air on the Alps within only 4 days.
- Pollen originated most frequently from at least Switzerland, up to France and Canada.
- Far-transported pollen may explain allergic symptoms at a rate of 87 %.

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ABSTRACT

Pollen grains are among the main causes of respiratory allergies worldwide and hence they are routinely monitored in urban environments. However, their sources can be located farther, outside cities' borders. So, the fundamental question remains as to how frequent longer-range pollen transport incidents are and if they may actually comprise high-risk allergy cases. The aim was to study the pollen exposure on a high-altitude location where only scarce vegetation exists, by biomonitoring airborne pollen and symptoms of grass pollen allergic individuals, locally.

The research was carried out in 2016 in the alpine research station UFS, located at 2650 m height, on the Zugspitze Mountain in Bavaria, Germany. Airborne pollen was monitored by use of portable Hirst-type volumetric traps. As a case study, grass pollen-allergic human volunteers were registering their symptoms daily during the peak of the grass pollen season in 2016, during a 2-week stay on Zugspitze, 13–24 June. The possible origin of some pollen types was identified using back trajectory model HYSPLIT for 27 air mass backward trajectories up to 24 h.

We found that episodes of high aeroallergen concentrations may occur even at such a high-altitude location. More than 1000 pollen grains m $^{-3}$ of air were measured on the UFS within only 4 days. It was confirmed that the locally detected bioaerosols originated from at least Switzerland, and up to northwest France, even eastern American Continent, because of frequent long-distance transport. Such far-transported pollen may explain the observed allergic symptoms in sensitized individuals at a remarkable rate of 87 % during the study period.

Long-distance transport of aeroallergens can cause allergic symptoms in sensitized individuals, as evidenced in a sparse-vegetation, low-exposure, 'low-risk' alpine environment. We strongly suggest that we need cross-border pollen monitoring to investigate long-distance pollen transport, as its occurrence seems both frequent and clinically relevant.

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1. Introduction

Airborne pollen is a major trigger of respiratory allergies worldwide, adversely affecting the quality of life of those afflicted (Roger et al., 2016) and having a major negative impact on economies (Zuberbier et al., 2014). Several studies predict that warmer temperatures and later onset of frosts due to climate change will cause the pollen season to start earlier in the year (Kinney, 2008; Shea et al., 2008). Also, temperature increase will enable plants to survive in higher latitudes, changing the pollen types present, and may extend the pollen season for some plants (IPCC, 2007; Kinney, 2018). Within the last century, the vegetation period in temperate climates has already been prolonged, bringing about longer overall pollen seasons, higher abundances of some pollen types and the occurrence of new allergenic species such as Ambrosia, which together inflict a higher burden on allergy sufferers (Biedermann et al., 2019; Damialis et al., 2019b; Muzalyova et al., 2019; Nadeau et al., 2021). While there is strong evidence of increasing abundances of airborne pollen and prolonged pollen seasons, it becomes more difficult to predict the spatial and temporal occurrence of pollen (Rojo et al., 2021).

Airborne pollen monitoring networks usually reflect local conditions in urban environments and, consequently, the local biodiversity and mainly the nearby emission sources. But it is well known for decades now that pollen grains can travel in the air for long distances under specific weather conditions (Rantio-Lehtimäki, 1994). Having said the above, information on pollen seasons based exclusively on local vegetation is not enough and a quantitative assessment of the origin of atmospheric pollen from more distant sources due to long-distance transport (LDT) ought to be considered as well when predicting the exposure risk for allergic patients.

The occurrence and frequency of long-distance transport of different biological particles has been the topic of numerous papers in the recent years, including but not limited to airborne pollen (Grewling et al., 2019; Rosselli et al., 2015; Varga et al., 2021). To the same direction, new technological developments in monitoring, observation and source-orientated modelling techniques of atmospheric pollen have contributed to deeper elaborate on this (Maya-Manzano et al., 2021). Studies have shown that pollen could be transported by air masses moving in an altitude of >2000 m above sea level (Damialis et al., 2017; Rousseau et al., 2003) and travel distances of >500 km from their source (Bogawski et al., 2019; Grewling et al., 2022). As a matter of fact, for some decades now it has been documented that pollen may be transferred across the sea or among continents covering distances up to hundreds or thousands of kilometres away from the source (Campbell et al., 1999; Stix, 1975). While such incidents may not be as frequent in every pollen season (Frisk et al., 2022), this could be highly variable among localities and years. Such research and relevant findings are particularly important given the growing social and scientific concern about the effects that climate change will have on bioaerosol distributions worldwide and subsequent human health (IPCC, 2022). Therefore, in times when the biodiversity is dramatically altering (IPCC, 2022), we need to also start considering the term 'remote biodiversity'.

A study which examined the long-distance transport of *Ambrosia* artemisiifolia pollen indicated that the most likely sources of pollen arriving in south-eastern Poland are Ukraine and Slovakia (Kasprzyk et al., 2011) or to the UK and the Netherlands from Central and south Europe (de Weger et al., 2016). In another work, *Pinus* pollen was a common component of long-distance transported pollen (Szczepanek et al., 2017). However, these investigations were carried out at low altitudes, and there had been a lack of research on potential long-distance transport of pollen at higher elevations.

Generally, LDT will take place under certain atmospheric conditions (Skjøth et al., 2021). One possible mechanism was indicated for ragweed pollen by Šikoparija et al. (2013), who described how a high-pressure field around the European part of Russia and low pressure over northwestern Europe caused surface winds that, in association with sunny weather and orographic 'foehn' winds, forced *Ambrosia* pollen movement northward to Poland and Scandinavia. Such events may extend the regional pollen season and elicit respiratory symptoms in sensitized individuals

(Damialis et al., 2019a). So, what has not been thoroughly examined to date is the intensity of each one of such isolated events; particularly in terms of public health, such incidents may still trigger respiratory symptoms to sensitized individuals. To detect and quantify LDT incidents the use of numerical atmospheric dispersion models is usually employed, such as COSMO-ART (Pauling et al., 2020) or SILAM (Siljamo et al., 2007), which can support in simulating sources for atmospheric pollen concentrations measured at designated sites. Backward trajectory analysis and atmospheric circulation models have frequently been used to model air paths and to find possible source regions for airborne pollen (Celenk, 2019; Izquierdo et al., 2011; Menzel et al., 2021), mainly for allergenic taxa found in low altitudes (Bachert, 2004; Buters et al., 2012; Plaza et al., 2016; Riediker et al., 2000). However, little is known about the origin of pollen found in high altitudes. High altitude vegetation includes exceptional and endemic species particularly sensitive to global change, which makes it important to study allergenic plants and their pollen in such extreme environments (Lamprecht et al., 2018; Stanisci et al., 2016). Such sites are also excellent 'real-life labs' for the detection of LDT, as vegetation is scarce and pollen from woody plant taxa is highly likely to originate from far-away sources. Nonetheless, very few studies have focused on alpine ecosystems and have indicated potential long-term pathways of pollen transport from far-distant sources (Ghasemifard et al., 2020). What is still unanswered is how frequent such phenomena are and, mostly, if they are relevant clinically for sensitized individuals.

The aim of this study was to investigate the biodiversity and spatiotemporal abundance patterns of airborne pollen and fungal spores in a highaltitude alpine setup, attempting to identify the origin of bioaerosols. In parallel, an allergic human cohort was established there to potentially prove whether incidents or far-transported aeroallergens may indeed cause associated symptoms. Our original hypothesis was that LDT on such an extreme environment like the Alps would be rare, if any, and the effect of real-life symptoms on the allergic cohort would be negligible.

2. Methods

2.1. Sampling site

The research was carried out in 2016 on the Environmental Research Station UFS (Umweltforschungsstation Schneefernerhaus), located on the Zugspitze Mountain at an altitude of 2650 m (latitude: 47.42° N, longitude: 10.98° E) in Southern Bavaria, Germany. The study period was during 13 to 24 June.

This high-alpine region reflects an ecosystem highly sensitive to climate variability and change (Schirpke et al., 2017). The climate is tundra (Köppen climate classification: ET), preserving the single glacier existent in Germany, despite its decrease over the years, and with the Westerlies as the predominant wind.

As in the majority of alpine ecosystems, vegetation is sparse, however, differences exist between the northern and the southern part of the mountain. The shaded and moist northern slopes host pines, growing at elevations of up to 1800 m. The woods lower down consist mainly of spruce and fir, but honeysuckle, woodruff, meadow-rue and speedwell also occur here. To the south, the sight changes to larch and pine forests and into mixed woods of beech and sycamore. Here too, mountain pine grows at the higher elevations of over 2000 m. Relatively rare in the entire Zugspitze area are trees like birch, rowan, juniper and yew (Väre et al., 2003).

The vegetation found in the city of Augsburg, on the other hand, is very different (Fig. 1). The urban vegetation is almost entirely planted and is permanently disturbed by anthropogenic influence; nevertheless, it adopts certain characteristics of the plant community due to the temporal evolution of the plant composition (Schwandt and Friedmann, 2018). The tree layer generally consists of Fagus sylvatica, Carpinus betulus, Quercus robur, Betula spp., Corylus spp., Pinus sylvestris and Picea abies. In addition to frequently planted and maintained sites, there are also areas of low influence where classical spontaneous vegetation can spread, where most of the grasses, legumes and other herbaceous plants like Urticaceae family appear.

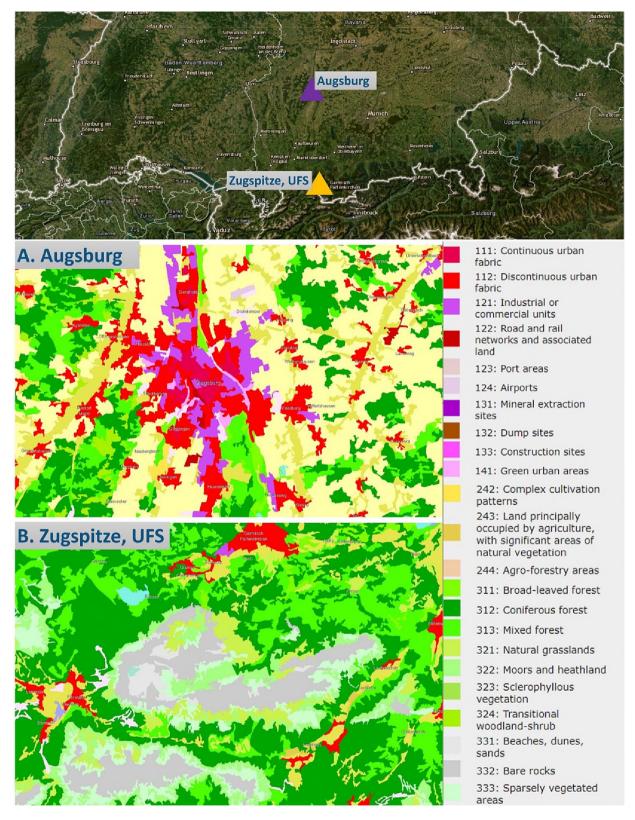


Fig. 1. Study sites location and land cover raster with vegetation coverage in Augsburg city and surroundings (A), and on Zugspitze Mountain (B) (CLC, 2020).

2.2. Pollen monitoring

Pollen monitoring in Zugspitze was conducted every 8 h (morning, afternoon, night) during half an hour each time using portable Burkard samplers (Burkard Manufacturing Co. Limited, Rickmansworth, Hertfordshire, England, UK) (Hirst, 1952) at ground level. Pollen grains were identified

under light microscope and were estimated per cubic metre of air, on two time resolutions, per day and per 8 h. The laboratory techniques including pollen identification, counting and the measurement units used were described in Damialis et al. (2019a). Briefly, the portable trap was equipped with a vacuum pump that sucked in 10 l of air min $^{-1}$. The air particles were trapped on an adhesive-coated slide (Burkard gelvatol). The slides

were stained with a solution of saffranin, gelatin, glycerol and phenol. All pollen grains were counted under a light microscope (Leica DM750) at \times 400 magnification. Counts were expressed as mean daily pollen concentrations (number of pollen grains per $\rm m^3~d^1$). Likewise, airborne pollen in the city of Augsburg was collected by use of a stationary, 7-day recording Burkard volumetric trap, located at the Bavarian Environmental Agency bureau, at ground level. Counts were made on a bi-hourly basis and expressed as mean daily pollen concentrations following the recommendation by European Aerobiology Society (Galán et al., 2014).

2.3. Human cohort characteristics

Five volunteers were recording their respiratory symptoms by means of an online symptom diary ('Pollen' app, Stiftung Deutscher Polleninformationsdienst). The participants were sensitized to grass pollen (sIgE >0,3 kU/L) with self-reported respiratory symptoms during the grass pollen season.

Monitoring of any symptoms and medication intake took place daily during the (expected low-altitude) peak of the grass pollen season. Symptoms were recorded on an 8-hourly scale regarding nasal, ocular, and pulmonary symptoms. A total nasal symptom and medication score was calculated from the data as previously described (Damialis et al., 2019a).

Before moving to the UFS, the participants had to record their symptoms in Augsburg, a Bavarian city located approx. $100~\rm km$ to the northwest of the Alps and 494 m of altitude. The stay on Zugspitze Mountain took $10~\rm days$, without interruption. The participants were allowed to go outside on the terrace at any time or take the cable car to the restaurant on the Zugspitze summit for lunch.

The study was approved by the local ethics committee (code: 19/15) and conformed to the guidelines of Helsinki. Study participants were enrolled after written informed consent.

2.4. Meteorological data

The meteorological analysis was based on selected parameters recorded by the Deutscher Wetterdienst (DWD), the Federal Ministry of Transport and Digital Infrastructure at UFS, which is located in the immediate vicinity of the aerobiological monitoring site. The following meteorological parameters were used: air temperature (°C), air pressure (hPa), precipitation (mm), sunshine duration (h), and wind vectors (speed and direction).

The prevailing regional atmospheric circulation patterns were derived from the Global Forecast System (GFS) provided by the National Oceanic and Atmospheric Administration (NOAA) with a spatial resolution of $1^{\circ}\times1^{\circ}$. The regional wind fields were calculated by using the U- and V-component of wind on the geopotential height of 500 hPa (mb) level. This height of 500hpa, according to the American Meteorology Society (https://glossary.ametsoc.org/wiki/Welcome) and the European Centre for Medium-Range Weather Forecasts (ECMWF; https://www.ecmwf.int/), is typically used in weather forecasting, and is considered as the 'level of non-divergence': the structures of the migratory cyclonic-scale weather systems are associated with this layer, while the strongest vertical movements are observed there.

2.5. Back trajectories

Backward air mass trajectories were calculated using the HYSPLIT model (Hybrid Single-Particle Lagrangian Integrated Trajectory) in order to investigate potential long-distance transport at the UFS. The trajectories were computed using the Global Forecast System (GFS) provided by the National Oceanic and Atmospheric Administration (NOAA) with a spatial resolution of 0.25° . Backward trajectory ensembles were calculated with vertical velocity motion, the maximum duration of 315 h and 3-times daily (07:00, 15:00, 23:00 CEST) within the study period, where each ensemble run contains 27 trajectories with one meteorological grid point in the horizontal and 0.01 sigma units in the vertical. All times are presented as Central European Summer Time (UTC + 2).

Both altitude and longitude of trajectories were analyzed. In the first step, by altitude, each track was ended when it first fell below 200 m above ground level (AGL). These shortened trajectories were then reduced to one with a total distance longer than 100 km and declared as trajectories with long distance transport, as defined by Rantio-Lehtimäki (1994). This analysis was also applied on other thresholds for altitude and distance (see Supplementary Material).

Later, the trajectories were calculated in different scenarios for the longitude. First, up to 24 h backwards, on four levels: 1500, 2000, 2500 and 3000 m above sea level and compared with wind field. Most probable trajectories were obtained with high spatial resolution data of 0.25° meteorological data sets. They take fluctuations into account that result from convection, friction forces and geomorphology. Normally, pollen grains detected at >1000 m height most likely do not originate from local sources but from long-distance transport. The level of 1000 m above sea level reveals the lower boundary of the free atmosphere where the friction phenomenon of the earth's surface vanishes (Oke, 2002). Pollen at this altitude (and above) can be transported over long distances (LTD) (de Weger et al., 2016; Damialis et al., 2017), as they might be lifted up to this height by air convection and turbulences.

Second, we chose a 315-h backwards period, for which we calculated trajectories of potentially higher length. Such trajectories need a longer time (minimum 3 days) for the air mass transformation to take place and to possibly observe changes in atmospheric circulation that might result in airborne pollen transport. Moreover, the frequency of every trajectory exceeding or equal to 10, 100 or 1000 km distance was calculated.

The backward trajectories were processed using R ("RStudio," 2020).

3. Results

3.1. Pollen and spore data

In total, 17 different pollen types (Fig. 2) and 11 fungal spore types (Agrocybe, Alternaria, Ascospores, Asperisporium, Botrytis, Cladosporium, Fusarium, Ganoderma, Leptosphaeria, Periconia, Ustilago) were detected on the Umweltforschungsstation Schneefernerhaus (UFS) in the German Alps, during the period 13–24 June 2016. Of the pollen types detected, 10 were the most abundant, namely Alnus, Betula, Carpinus, Cupressaceae, Cyperaceae, Fraxinus, Pinaceae, Poaceae, Quercus, and Urticaceae (Fig. 2). Since in this research the bioaerosol data of pollen only will be compared against pollen-associated symptoms, we do not further provide any details on the fungal spores' diversity or abundances.

The exposure varied significantly during the studied period. Even though the concentrations of pollen (and fungal spores) were most frequently low (comparably to the lower-altitude region of Augsburg; Fig. 2), still there were some episodes with high pollen concentrations. On June 16, >1000 *Carpinus* pollen grains per m³ were detected, and on the last two days (June 22, 23), Poaceae and Urticaceae concentrations were higher than 750 pollen grains/m³ (Fig. 2). Although 11 different pollen types were found on the UFS, only four types represented 84 % of the total spectrum, namely *Carpinus* (34 %), Pinaceae (26 %), Poaceae (15 %), and Urticaceae (9 %) (Fig. 2).

The pollen diversity collected at the UFS between 13 and 24 June was different from Augsburg pollen data, which was collected at the same time approx. 100 km north of UFS in an altitude of only 500 m a.s.l. The Augsburg data showed a greater variety of pollen types, strongly dominated by Poaceae (50 %) and Urticaceae pollen (39 %), all other 20 pollen types accounting for 0.5 % to 1 % of the entire spectrum. During the study period, no *Carpinus* pollen was detected in Augsburg, and Poaceae as well as Urticaceae pollen concentrations were lower in Augsburg than on UFS, with the seasons at the lower altitudes probably being already over, compared to the delayed high-altitude respective seasons.

In Fig. 3, the temporal distribution of the different pollen types may be seen. The maximum concentrations per day revealed distinct peaks for the biomonitoring period on the UFS, pointing out high pollen concentrations on the days of 14 June, 16 June, and 21–24 June 2016, which are ranked

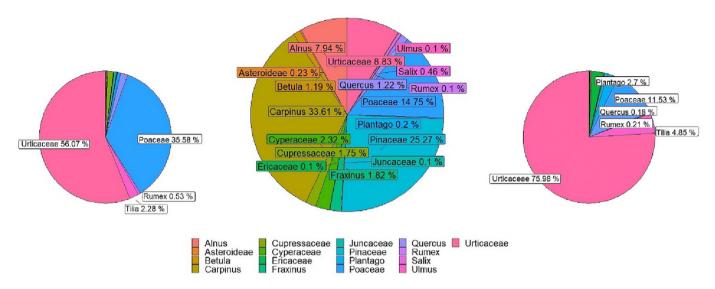


Fig. 2. Percentages of different pollen taxa during the study period (13–24 June 2016) at the UFS on Zugspitze Mountain (middle) and in Augsburg (left and right) 10 days before and 10 days after the stay in the Zugspitze station UFS.

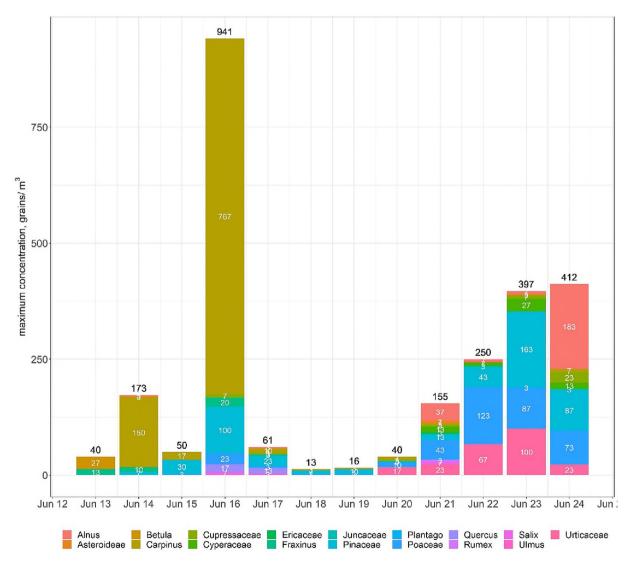


Fig. 3. Temporal distribution of airborne pollen concentrations (maximum per day, per pollen type) at the UFS during the period 13-24 June 2016.

on the highest percentile of all pollen concentrations during the whole period.

3.2. Long-distance transport trajectories and weather conditions

During the investigation period (13–24 June 2016), there were changing weather conditions at the UFS and therefore also changing long-distance transport trajectories (Fig. S1). Fig. 5A-E shows the resulting trajectories for the UFS during the highest pollen concentration days. To make sure that we do not observe and major carry-over effect because of delayed effects from recent pollen exposure, we did not consider the first day on the UFS, 13 June.

In the beginning, 14 June 2016 (Fig. 4A), a low-pressure field over north-eastern Europe and the Barents Sea was leaving towards the north-east. From the west, a distinct depression, an Icelandic low-pressure field, was moving towards Europe from the northern Atlantic. This weather situation was causing low pressure, thunderstorms and moderate to strong prevailing west winds in Central Europe and at the UFS.

On 14 June 2016 (Fig. S1), 78 % of the ensemble trajectories had at least a distance of 100 km and an altitude of 200 m AGL, showing a clearly long-distance transport from the west and transporting aeroallergens from northern Switzerland, Western France, or even more remote areas. Mainly *Carpinus*, Pinaceae and Poaceae pollen where measured (Fig. 3). The analysis of this situation, maps and the back-trajectories suggest that on 16 June 2016 (Fig. 5A), *Carpinus* pollen grains could have been transported from Italy, France and even Spain, 500–750 km from UFS, due to the direction of airflow. Later, air masses approached from the southwest and then Italy and Switzerland before finally reaching the Zugspitze Mountain (Figs. 4A, 5A).

On 18 June 2016 (Fig. 4B), the prevailing low-pressure system started to turn into a cut-off low and moved south to the Mediterranean Sea causing changing wind directions at the UFS with fewer long-distance transport tracks (52 %). The air masses arrived mainly from southwest to southeast with significantly lower abundances of Pinaceae, Poaceae and Urticaceae pollen types (Fig. 3).

But on 21 June 2016, the weather conditions changed again to consistent westerly winds, which contributed to potentially transferring airborne pollen from France, even farther away, across the Atlantic (93 % of the trajectories; Fig. 5B). On 22 June 2016 (Fig. 4C), a high-pressure field from southwest moved to Central Europe, bringing warmer temperatures and longer sunshine duration, which lasted up to the end of the investigation period. This resulted in stable, prevailing winds from the west with long-distance transport trajectories (85 % of the trajectories; Fig. 5C). Primarily pollen from Pinaceae, Poaceae, Urticaceae and *Alnus* were measured (Fig. 3) and, again, trajectories from northern Switzerland, Western France, or even more remote regions were the expected pollen sources.

When the frequency and intensity of prevailing winds in the study area were examined, it was found that westerly winds are the commonest in Southern Bavaria, with stronger winds coming from west and southwest (Fig. 6).

Wishing to quantify the incidents of long-distant transport of pollen, we visualised the frequency of tracks for each day with altitude $\geq\!200$ m and grouped for different potential long-distance transport distances. As seen in Fig. 7, always the highest probability (>50 %) was to observe midrange pollen transport (>100 km trajectories). But even for the longest-distance pollen transport (>2000 km trajectories), the highest frequency (>50 %) was also observed in most of the days, on 14/06/2016 and particularly during 20–24 June 2016. Over the last period, the probability of observing long-distance pollen transport of >2000 km was on average accounting for 81.5 % (Fig. 7).

3.3. Impacts of the environmental conditions on the human allergic cohort

Having gathered evidence for long-distant transport of pollen for approximately half of the days of the stay on the German Alps (Fig. 7), we studied whether the potentially far-transported pollen concentrations

might have had an impact on the allergic rhinitis patients present on the UFS. Using GLM models (General Linear Models) and a backward stepwise variable elimination procedure, we found that the most significant variables contributing to higher overall symptoms scores are the pollen concentrations over the previous three days, air temperature and relative humidity (p < 0.001 for all variables). Based on the GLM performed, airborne pollen concentrations during the previous three days could explain a remarkable fraction of 87 % of the overall variability, along with relative humidity, as seen in Fig. 8.

4. Discussion

4.1. Urban vs. natural ecosystem pollen exposure

Following our original research hypothesis, bioaerosol exposure on the Zugspitze Mountain was significantly lower compared to that in Augsburg, resulting in overall lower allergic symptoms. However, our results here show that under specific weather conditions, mainly rising temperatures, higher atmospheric pressure and low precipitation, systematic episodes of high aeroallergen concentrations can occur even in a high-altitude and low-vegetation environment. We hypothesised that we would not record high pollen concentrations and therefore allergic symptoms, if any, would be negligible. In contrast, what was evidenced was >1000 pollen grains per cubic meter of air during approximately half the stay on the Zugspitze. Per pollen type and for most different taxa, this amount is reached during the entire season in a typical urban environment (Bastl et al., 2016; Simoleit et al., 2017; Ziello et al., 2012). Because of these pollen amounts on the study area and the observed study period, our participants not only exhibited allergic symptoms, but the symptom scores registered correlated with the airborne pollen with a 3-day lag, and in synergy with relative humidity. Based on our GLM model, these two parameters can explain a remarkable 87 % of the total symptom score of our patients. Ultimately, our original hypothesis not only is rejected, but it is evident that longdistance transport of pollen is frequent (more than half of the days and instances) and clinically relevant (highest symptoms days observed on the longest-distance transport cases).

4.2. Remote biodiversity and long-distance transport of pollen

While it is common sense that airborne pollen largely reflect the local and regional vegetation, the occurrence of longer-range transport of pollen has been documented several times, thus, setting grounds for the term 'remote biodiversity'. During a dramatically changing global climate and with biodiversity being greatly affected, it seems that the phenomenon of 'remote biodiversity' is underestimated, if not neglected. Studies on longdistance transport of airborne pollen have been quite numerous, including the exact same study area quite recently: Ghasemifard et al. (2020), also documented consistent trajectories from distant (from the west) sources and transfer of Alnus pollen from Switzerland or farther away into the region of the German Alps. In Alpine environments the phenological stages are later and shorter than at lower altitudes (Preite et al., 2015; Willemse and Furger, 2016) and some of the pollen types are not present in the flora of the Alps (Bohn and Neuhäusl, 2004). Back trajectories and weather patterns clearly suggested that the origin of these pollen was long distance transport (at least 100 km). In our attempt to quantify this incident, we assessed that pollen on such environments and at higher altitudes, both above sea level and above ground level, may transfer the aeroallergens for several hundreds, even thousands of kilometres away from the source. Based on our data here, to the extent that pollen may be transferred from distant sources, the latter would then be most probably Switzerland, France, Spain, up to the northeast coast of the American Continent.

The meteorological conditions that cause transport of pollen over long distances depend on the pollen type and the area under study. Already in 1991, it was reported that wind direction and -speed, temperature, relative humidity, precipitation, air pressure and solar radiation as well as microand macro-topography within the area- influence *Betula* pollen dispersion

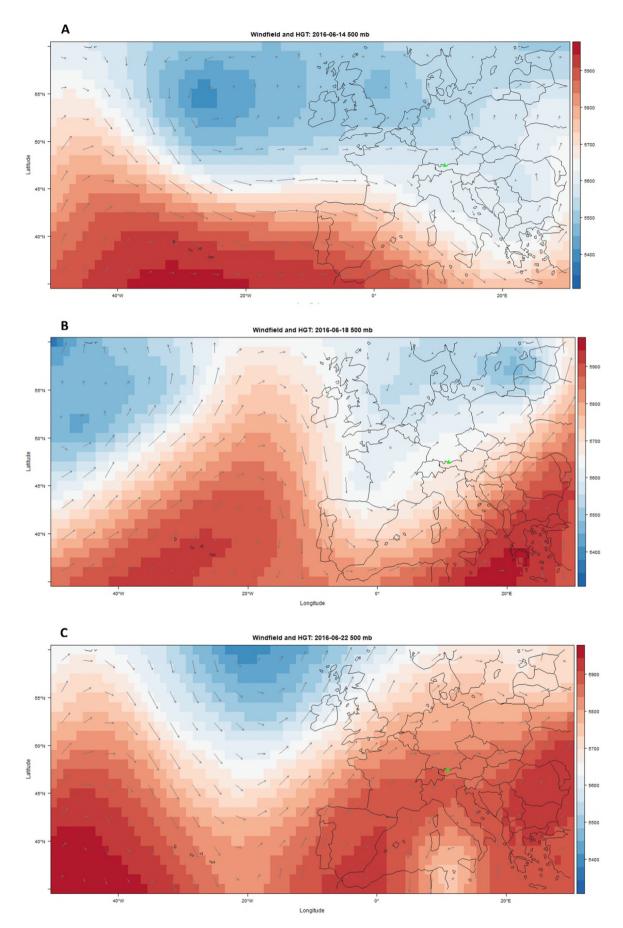


Fig. 4. Maps with the meteorological conditions over Europe at 500 hPa geopotential height and wind field, A) on 14 June 2016, B) on 18 June 2016, C) on 22 June 2016.

(Hjelmroos, 1991). Pollen transport to Poland in air masses over the Carpathian Mountains was found to be related to the depth of the planetary boundary layer, hot and dry weather and winds from the south (Smith et al., 2008). A 23-year observation study on Pinaceae pollen distribution showed a correlation of pollen with high temperatures during summer before the onset of the local Pinaceae flowering season (Huusko and Hicks, 2009).

In our study, regarding the question where the pollen grains come from, back trajectories were processed and the frequency and type, along with weather patterns, were combined. The back-trajectory analysis provided a prominent source consistently from Northern Italy, Spain and France during most of the days of the study period. One could therefore hypothesise that they might comprise a source of *Carpinus* pollen in southern and western Germany, leading to extended *Carpinus* pollen seasons in these regions.

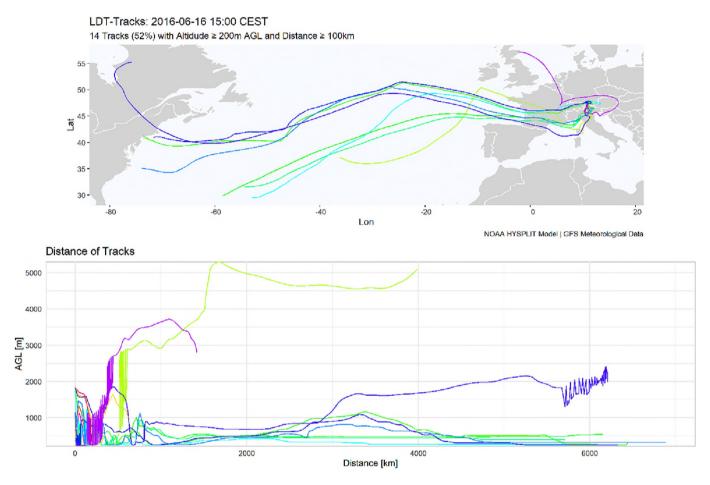


Fig. 5. A. HYSPLIT back trajectories at the UFS, on 16 June 2016.

Upper graph: 14 Trajectories with altitude ≥ 200 m AGL and distance ≥ 100 km (out of a total combination of 27 tested tracks). Y-axis shows the latitude and X-axis the longitude.

Lower graph: Y-axis shows the height profile of HYSPLIT back trajectory with altitude ≥ 200 m AGL, and X-axis shows the trajectory distance ≥ 100 km. (Note the different Y- and X-axis values).

B. HYSPLIT back trajectories at the UFS, on 21 June 2016.

Upper graph: 25 Trajectories with altitude ≥ 200 m AGL and distance ≥ 100 km (out of a total combination of 27 tested tracks). Y-axis shows the latitude and X-axis the longitude.

Lower graph: Y-axis shows the height profile of HYSPLIT back trajectory with altitude ≥ 200 m AGL, and X-axis shows the trajectory distance ≥ 100 km. (Note the different Y- and X-axis values).

C. HYSPLIT back trajectories at the UFS, on 22 June 2016.

Upper graph: 23 Trajectories with altitude ≥ 200 m AGL and distance ≥ 100 km (out of a total combination of 27 tested tracks). Y-axis shows the latitude and X-axis the longitude.

Lower graph: Y-axis shows the height profile of HYSPLIT back trajectory with altitude ≥ 200 m AGL, and X-axis shows the trajectory distance ≥ 100 km. (Note the different Y- and X-axis values).

D. HYSPLIT back trajectories at the UFS, on 23 June 2016.

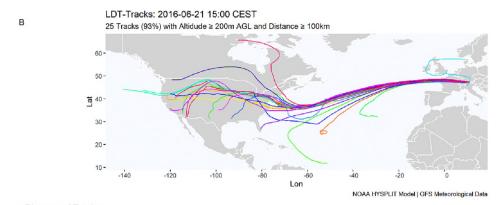
Upper graph: 21 Trajectories with altitude ≥ 200 m AGL and distance ≥ 100 km (out of a total combination of 27 tested tracks). Y-axis shows the latitude and X-axis the longitude.

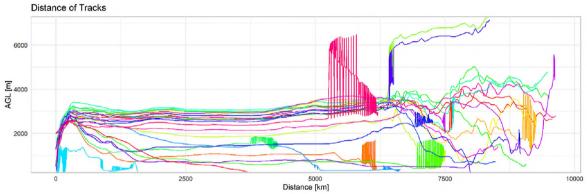
Lower graph: Y-axis shows the height profile of HYSPLIT back trajectory with altitude ≥ 200 m AGL, and X-axis shows the trajectory distance ≥ 100 km. (Note the different Y- and X-axis values).

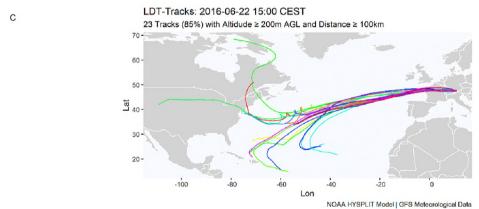
E. HYSPLIT back trajectories at the UFS, on 24 June 2016.

Upper graph: 15 Trajectories with altitude ≥ 200 m AGL and distance ≥ 100 km (out of a total combination of 27 tested tracks). Y-axis shows the latitude and X-axis the longitude.

Lower graph: Y-axis shows the height profile of HYSPLIT back trajectory with altitude ≥ 200 m AGL, and X-axis shows the trajectory distance ≥ 100 km. (Note the different Y- and X-axis values).







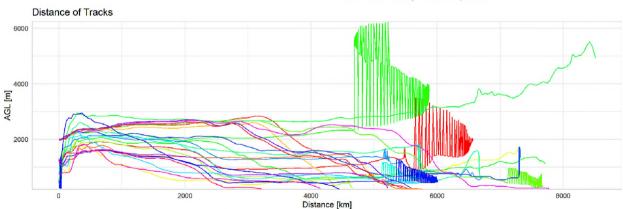
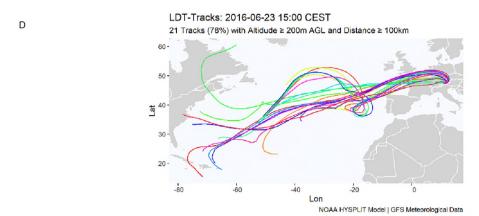
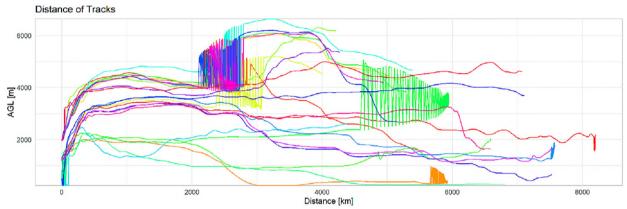
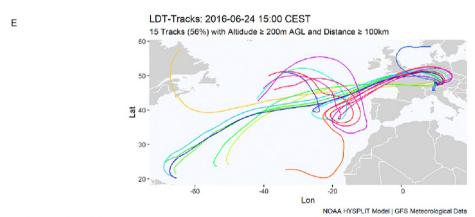


Fig. 5 (continued).







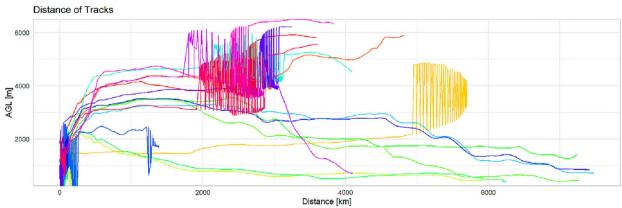


Fig. 5 (continued).

Frequency Wind Rose

June 14 - June 23, 2016

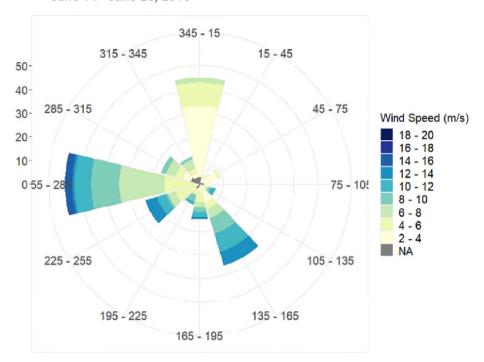


Fig. 6. Average wind direction during 13–24 June in UFS. Minute values with a given wind direction in degrees are sorted into eight bins. The length of bars indicates the percentage of (minute-) measurements in which the respective wind direction occurred.

Since Carpinus and Betula had ended their flowering in Central Europe by end of May (e.g. Kolek et al., 2021), the pollen grains detected at UFS in June must have originated from medium- or even from long-distance transport and have either derived from plants that flowered later than the local plants, or from plants with significantly prolonged flowering, or from resuspension of local pollen released months earlier. However, in a more conservative concept, we must note that back-trajectory modelling does not provide evidence per se of long-distance transport incidents. They mark the probability of having such incidents, but only the combination with vegetation and regional flowering phenology data may lead to determinative conclusions.

It is also well known that altitude affects the seasonal exposure to allergens. However, pollen and fungal spore abundances depend on complex environmental parameters, such as prevailing winds, local vegetation, land use, topography and local meteorology (Davies, 1969a, 1969b; Weber, 2002). It has therefore to be considered that Alpine regions might host

unpredictable environmental regimes, both temporally and spatially (Menzel et al., 2003; Scheifinger et al., 2013). A general rule of lower pollen concentrations in high altitude locations can therefore not easily be deducted (Gehrig and Peeters, 2000).

4.3. Association between airborne pollen long-distance transport and allergic symptoms

Our data suggest that even in environments with scarce vegetation, such as an Alpine summit, low pollen concentrations are not always guaranteed, and thus, high-altitude stays might not reliably result in lower symptoms in allergic patients. The analysis of pollen concentrations at high altitudes is, however, essential to improve our understanding of basic aerobiological patterns. Of note, recent studies on vertical aeroallergen abundance have shown that there is an increase in the concentration of pollen grains (and fungal spores) with increased altitude (Damialis et al., 2017; Pitari et al.,



Fig. 7. Probability (%) of occurrence of HYSPLIT trajectories with long-distance transport (>100 km), with altitudes ≥ 200 m AGL per day.

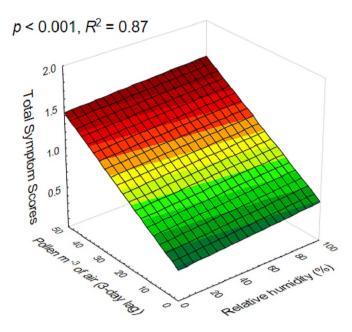


Fig. 8. Surface plot of total symptom scores, airborne pollen concentrations and relative humidity.

Pollen concentrations refer to lagged concentrations of -3 days before.

2014). Damialis et al. (2017) have strongly suggested the existence of pollen and spore clouds towards the boundary layer, which points out the need for three-dimensional atmospheric modelling, instead of the common belief that bioaerosols are mostly prevalent near the vegetation sources.

To confirm the origin of long distance transported pollen, information on both, pollen source locations and local flowering phenology are suggested (Hernandez-Ceballos et al., 2014), since phenological observations alone do not suffice to determine the timing of the main pollen because of LDT (Ranta et al., 2006). Latest studies use satellite-based remote sensing data such as Normalized Difference Vegetation Index (NDVI) or Enhanced Vegetation Index (EVI) as alternatives for flowering times (Bogawski et al., 2019). While this is a limitation of our study, the lack of vegetation data for our analysis, this was not feasible as the human cohort was allergic to grass pollen and such information is not available at a cross-national scale, to the best of our knowledge.

Regarding the symptoms related with high elevation, there is no previous evidence that altitude contributes to the severity of seasonal allergy symptoms. If we assume that due to the higher altitudes (where the humidity is lower and there are fewer deciduous trees), pollen grains and fungal spore concentrations are lower, the allergic symptoms should also be lower. This has been also supported by previous results on the same study area (Damialis et al., 2019a, 2019b), where it was highlighted that pollen exposure and associated symptoms (before and after a stay in an Alpine region) were higher in a nearby urban location and this reflected in the actual symptom scores, as well in several immune responses. Of note, highaltitude sites have been recently suggested as optimal treatment locations for severe and uncontrolled asthma in sensitized individuals (Fieten et al., 2022). While there is some basis on this assumption, it is still not quite clear if this would be applicable to allergenic pollen as well, since our results here clearly indicate that the German Alps most frequently are not a 'safe' environment, because of far-transported pollen. Symptoms occurred even under lower-grade exposure, in a presumed 'low-risk' environment. We showed for the first time that potential incidents of long-distance transported pollen could indeed cause allergic symptoms in sensitized individuals, namely at a remarkable rate of 87 %. Finally, given that on such high-altitude environments thunderstorm occurrence is also more frequent, it is possible that thunderstorm asthma incidents may also be responsible for increased respiratory symptoms, as has been previously documented (Thien et al., 2018), which also has to be considered when visiting Alpine regions, especially for treating severe asthma patients (Fieten et al., 2022).

High elevation could also put individuals under environmental stress, and, concomitantly, they may become symptomatic even under occasional high pollen exposure during short intervals. Especially since repeated incidents of long-range transport of pollen take place beyond and out of the pollen season, we suggest that more thorough research on similar setups and for entire seasons have to be elaborated in order to clarify the associated mechanisms. Particularly if one takes into account that here we have evidenced a high probability of pollen being transported from even some thousands of kilometres away and such incidents causing allergic symptoms, a global and consistent cross-border monitoring of bioaerosols has to be organised and operated so as to inform, warn and predict high-allergy-risk intervals.

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CRediT authorship contribution statement

Conceptualization, A.D.; methodology, D.B., M.P.P. and A.D.; formal analysis, D.B., M.P.P and A.D.; investigation, D.B., M.P.P., F.K., V.L.-W., C.T.-H., S.G. and A.D.; data curation, D.B., F.K., M.P.P., A.D. and V.L.-W.; writing—original draft preparation, S.G., M.P.P., D.B. and A.D.; writing—review and editing, D.B., S.G., M.P.P., F.K., V.L.-W., C.T.-H. and A.D.; visualization, M.P.P., D.B. and A.D.; supervision, A.D.; funding acquisition, C.T.-H. All authors have read and agreed to the published version of the manuscript.

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Data availability statement

The associated to the manuscript datasets may be available by the corresponding author upon reasonable request.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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