

Effects of extreme spring temperatures on urban phenology and pollen production: a case study in Munich and Ingolstadt

Susanne C. Jochner^{1,*}, Isabelle Beck², Heidrun Behrendt²,
Claudia Traidl-Hoffmann², Annette Menzel¹

¹Department of Ecology and Ecosystem Management, Ecoclimatology, Technische Universität München,
Hans-Carl-von-Carlowitz-Platz 2, 85354 Freising, Germany

²ZAUM Center for Allergy and Environment, Department of Dermatology and Allergy Biederstein, Technische Universität München, Biederstein Str. 29, 80802 Munich, Germany

ABSTRACT: Extreme temperatures have a notable effect on phenology, much greater than expected from the general rule that low temperatures lead to a later—and high temperatures to an earlier—onset of phenological phases. The latter phenomenon can be seen when comparing urban areas with their rural surroundings: plants flower earlier in cities due to the urban heat island effect that contributes to higher temperatures. We investigated the effects of extreme temperatures on differences between urban and rural phenology and on human health (considering allergenic plants) in 2009 using phenological observations of flowering and leaf unfolding of birch *Betula pendula* Roth and flowering of horse chestnut *Aesculus hippocastanum* L. in the cities of Munich and Ingolstadt, Germany. Temperatures recorded in Munich during April 2009 were the second highest since records began in 1955 and led to rapid plant development whereas differences between urban and rural phenology were diminished. Laboratory examination of birch pollen grains revealed that the amount per catkin did not differ significantly between the city of Munich and the surrounding countryside. Long-term observations (1951/1955 to 2008, German Meteorological Service) were used to study the differences in flowering onset times between Munich and its surroundings. We found that weather conditions lasting only a few days can influence phenological behaviour, especially at the micro- and mesoscale. High temperatures, mainly extreme warm spells, were more likely to result in simultaneous flowering in urban and rural environments; low temperatures resulted in a longer delay in phenological onset times for flowering in Munich.

KEY WORDS: Phenology · Extreme events · Urban heat island · Pollen amount · Allergy · *Betula pendula* · Temperature sums · Germany

— Resale or republication not permitted without written consent of the publisher —

1. INTRODUCTION

Climate change is likely to have an impact not only on mean temperatures, but also on temperature variability (Schär et al. 2004). Extreme events (e.g. warm spells) are expected to increase in the future, both in frequency and intensity (IPCC 2007), and are likely to influence ecosystems more than any change in mean temperatures (Jentsch et al. 2007). Research in

extreme event ecology is therefore increasing and further investigations in the field of phenology–human health interactions are still needed.

Since temperature is the most important driver of plant phenology in temperate and moist regions, warm spells and their influence on plant development is of great interest. The heat wave during summer 2003 was a first opportunity to study such impacts (Fink et al. 2004, Meehl & Tebaldi 2004,

*Email: jochner@wzw.tum.de

Schär et al. 2004, Stott et al. 2004). The situation of 2003 is likely to be repeated: every second summer could be as warm as that summer or even warmer by 2100 (Schär et al. 2004). Thus, we should learn from recent extremes and study their consequences which are likely to be more frequently experienced in the future.

As well as showing current extremes, cities can also be regarded as a mirror of future climate. There is plenty of evidence that plants in urban areas flower earlier than those in the surrounding countryside (Franken 1955, Rötzer et al. 2000, White et al. 2002, Zhang et al. 2004, Mimet et al. 2009). A major feature of cities is the urban heat island effect (UHI) (Landsberg 1981, Oke 1987) that is associated with higher temperatures. The UHI can be observed throughout the year, but exhibits a daily and annual variability and is most distinctive during anticyclonic weather conditions with weak winds, clear skies and a marked diurnal temperature range (Oke 1987). The magnitude of the UHI also depends on the size of a city (Oke 1973). In general, the mean UHI varies between 1 and 3 °C (Landsberg 1981). In Munich, for example, this effect averages 1.9°C and can even reach values of 8.2°C (Matzarakis 2001). The temperature range of projected warming according to different scenarios is 1.8 to 4.0°C by 2100 (IPCC 2007) and can therefore be compared with mean UHI. Consequently, studying urban ecosystems allows estimation of changes that may occur in the near future. Understanding phenological events is crucial for assessing the impacts of temperature increases on the life cycle of plants and thus their survival and distribution (Chuine 2010). Since 2008, more than half of the world population has lived in cities (Grimm et al. 2008), and these areas are thus fundamentally interesting.

There is strong evidence that people living in cities are more affected by pollinosis incidence than those living in the countryside—an effect that is probably related to urbanization, increased vehicle emissions and westernised lifestyle (Braun-Fahrlander et al. 1999, D'Amato 2000, Ring et al. 2001). Further features of urban phenology are that earlier onset dates for allergenic plants in the city imply earlier appearance of symptoms. Due to the temporal delay in flowering onset between the city and the countryside, wind transport may also transfer urban pollen to the countryside where the flowering season of allergenic plants has not started. Subsequent to the end of urban flowering, pollen from the countryside can also be transported to the city, implying a protracted pollen season. Therefore, wind transport of airborne

pollen is an important cause for unsynchronised starting dates in phenology and aerobiology (Estrella et al. 2006).

In Europe, the incidence of asthma, allergic rhinitis, allergic conjunctivitis and eczema presents an important human health problem (Traidl-Hoffmann et al. 2003, WHO 2003). Pollen-related allergic diseases are influenced by generally earlier and longer pollen seasons, higher total pollen counts, stronger allergenicity and altered plant and pollen distribution (e.g. reviewed by Beggs 2004, Traidl-Hoffmann et al. 2009); thus, further investigations are needed to improve climate risk assessment.

We therefore aimed to explore the phenological behaviour of plants and the pollen production of *Betula pendula* Roth (birch) in urban and rural areas in and around the Bavarian cities of Munich and Ingolstadt during the extreme warmth of April 2009. Long-term phenological and temperature data (1951/1955 to 2008) from the German Meteorological Service (DWD) were used to study the reasons for diminished differences in onset dates between the city and the countryside. We hypothesised that higher temperature sums, i.e. warmer periods, lead to less pronounced differences in flowering phenology between the city and the countryside. Consequently, lower temperature sums, i.e. cooler periods, could imply a remarkable temporal delay between urban and rural flowering dates. We also hypothesised that the magnitude of the UHI (the urban–rural temperature difference) could have an influence on the differences in urban–rural phenology: smaller differences in temperature could also lead to smaller differences in phenological onset dates and vice versa. To describe the association between climate variability and allergenic disorders, we address the impacts of extreme warm periods on the phenological behaviour of (allergenic) plants and subsequently on human health.

2. MATERIALS AND METHODS

2.1. Study area

The regions of interest (Fig. 1, see Table 1) were 2 German cities located in the state of Bavaria, and their surroundings: Munich (48.14°N, 11.58°E; Fig. 1a) and Ingolstadt (48.77°N, 11.43°E; Fig. 1b). To examine urban and rural differences in phenology, the study areas were restricted to a maximum distance of 35 km from the city centres of Munich and Ingolstadt.

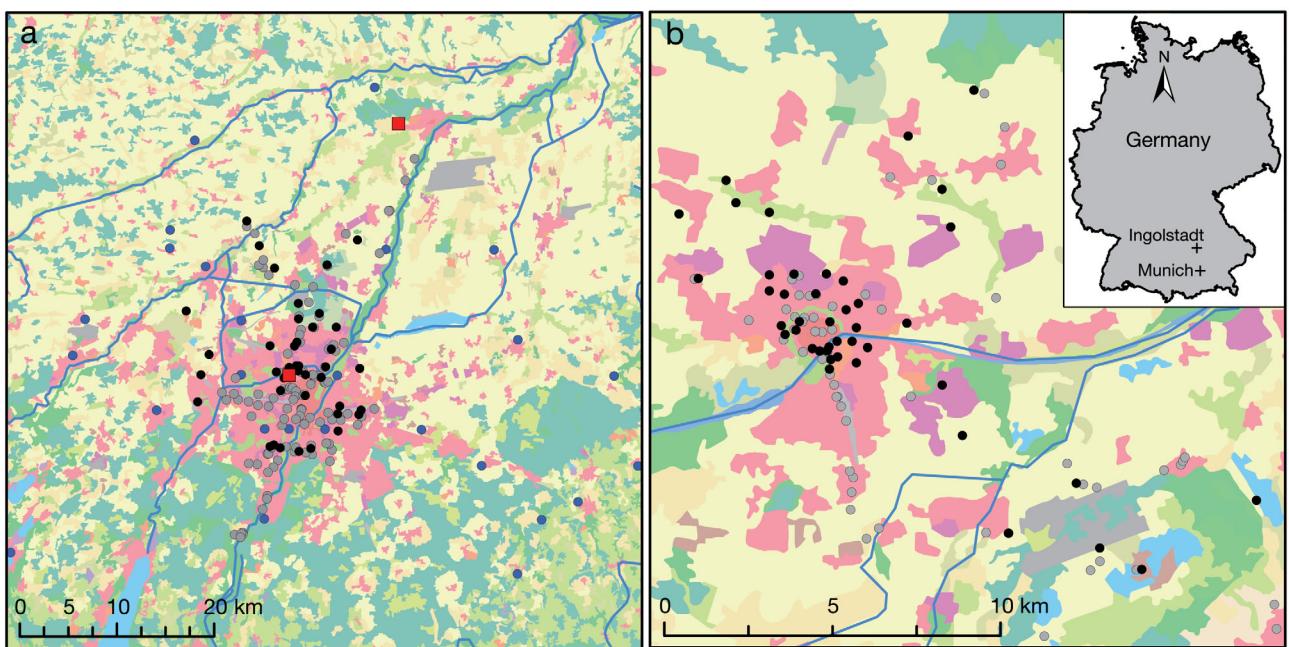


Fig. 1. Studied cities: (a) Munich and (b) Ingolstadt, located in southern Bavaria, Germany. (a) Red squares: German Meteorological Service (DWD) climate stations in Munich City (urban) and Weihenstephan (rural); blue circles: long-term phenological observations; (a,b) Black and grey circles: field observations in 2009 for *Betula pendula* Roth (birch) and *Aesculus hippocastanum* L. (horse chestnut), respectively; background: CORINE Land Cover 2000 (EEA 2000), major classes: red = urban fabric, green = forest and pastures, yellow = arable land, blue = rivers, lakes (see www.eea.europa.eu/themes/landuse/interactive/clc-download for a complete legend)

The classification of sites as either urban or rural was based on CORINE Land Cover 2000 data (EEA 2000). Components of built up areas (e.g. continuous and discontinuous urban fabric, industrial or commercial units) were extracted from the land cover dataset to calculate the proportion of urban area within a radius of 2 km of the phenological recording stations. This radius was chosen to ensure that stations within a large built up area were identified as such. A radius of 2 km is also used by the DWD to define the observation area for their phenological stations. We denoted urban sites as those with at least 50% urban cover and the remainder as rural sites. A subjective comparison of the observed sites in 2009 with our definition of urban and rural areas confirmed the classification, with agricultural areas, forests and natural areas being correctly classified as

rural sites. Urban areas were conversely characterised by a high building density and a large proportion of impervious surfaces. The characteristics of the 2 cities are summarised in Table 1. The amount of urban land use as specified above varied owing to the physical size of the city itself.

2.2. Phenological data

2.2.1. Phenological observations in 2009

In spring 2009, phenological observations were conducted according to the BBCH (Biologische Bundesanstalt, Bundesortenamt und Chemische Industrie) code (Meier 2001) for leaf unfolding and flowering of *Betula pendula* Roth (birch, see black circles in

Table 1. Details of the cities of Munich and Ingolstadt. Values for latitude, longitude and altitude are for the city centre. No. of inhabitants are given for 2008. Land use: survey area. City: within city administrative boundaries

	Latitude (°N)	Longitude (°E)	Altitude (m a.s.l.)	Inhabitants (no.)	Land use (%) Urban	Rural	City area (km ²)	City land use (%) Urban	Rural
Munich	48.14	11.58	515	1 326 807	12.8	87.2	308.9	62.5	37.5
Ingolstadt	48.77	11.43	370	123 925	4.9	95.1	133.4	26.7	73.3

Fig. 1a,b) as well as for flowering of *Aesculus hippocastanum* L. (horse chestnut, see grey circles in Fig. 1a,b) in Munich and Ingolstadt. Observations in each city were done by 2 to 4 trained recorders every third day. The observation period lasted from March 27 to May 8 (Table 2). In Ingolstadt, observations always started and ended a little bit earlier as phenological development stages were reached earlier than in Munich. Since both genetic variation among individuals and vigour influence phenology (Baumgartner 1952), dates for each phenological site were the means for 2 to 6 plants per species. Moreover, we excluded trees of <50 cm diameter as plant age alters phenological behaviour, i.e. older trees tend to have later onset dates of flowering and leaf unfolding than younger trees (Rosenzweig et al. 2008).

We calculated descriptive statistics (mean, maximum and minimum date of onset, range, SD and SE) by city, species, phase, and land use (urban and rural).

We defined the difference in phenological onset dates between urban and rural areas as the urban phenology effect (UPE) since this deviation is supposed to be an expression of the UHI. Statistical comparison of means was done using 2 sample *t*-test unless the data were not normally distributed or failed the homogeneity of variance test (Levene's test), in which case a Mann-Whitney *U*-test was used.

2.2.2. Long-term phenological data (1951 to 2008)

Long-term observations of flowering (1951 to 2008) were provided by the DWD. We focused on 3 plant species that are frequently found in cities: *Corylus avellana* L. (hazel), *Forsythia suspensa* L. (forsythia) and horse chestnut. We selected stations that were within a radius of 35 km from the city centre of Munich. To avoid problems with phenological lapse rates, the altitudes of phenological stations should be similar to the selected climate stations (Weihenstephan: 477 m, Munich City: 512 m). Therefore, only phenological stations ranging in altitude from 450 to 560 m a.s.l. were chosen. Since observations within cities were sparse and often had gaps of observation years, we constructed composite time series using the R pheno package of Schaber (2003) from all available phenological data in urban areas. For rural areas, the time series were derived from those stations that had at least 15 observation years within the study period 1951 to 2008. This resulted in the following number of series (urban/rural): hazel (5/18), forsythia (5/19) and horse chestnut (5/20) (see blue circles in Fig. 1a).

Table 2. Number of sites and trees per land use type for birch *Betula pendula* and horse chestnut *Aesculus hippocastanum* in Munich and Ingolstadt, and duration of the observation period. Urban (rural) = sites with >50% (<50%) urban land use within a 2 km radius

Type	Sites	Trees	Observation (d.mo.yr.)
Birch			
Munich			04.04.2009–16.04.2009
Urban	28	66	
Rural	17	48	
Total	45	114	
Ingolstadt			27.03.2009–15.04.2009
Urban	28	85	
Rural	13	50	
Total	41	135	
Horse chestnut			
Munich			20.04.2009–08.05.2009
Urban	87	317	
Rural	26	74	
Total	113	391	
Ingolstadt			19.04.2009–04.05.2009
Urban	31	111	
Rural	31	85	
Total	62	196	

2.3. Temperature data

2.3.1. Long-term temperature data (1955 to 2008)

The selected climate stations—Munich City (48.17°N, 11.55°E; urban station, 1955 to 2008) and Weihenstephan (48.4°N, 11.7°E; rural station, 1955 to 2008) (see red squares in Fig. 1a)—are operated by the DWD. In this study, the difference between the temperature measured at Munich City and that at Weihenstephan climate stations was defined as the UHI of Munich.

A common evaluation of extreme events focuses mostly on percentiles (e.g. 10th percentile), threshold, or duration indices (Alexander et al. 2006, IPCC 2007). Here we used the temperature deviation from the standard reference period 1961–1990 of the respective station as a proxy for extremes of temperatures. Correlation analyses incorporating these deviations and the UHI were used to identify relationships with the magnitude of the UPE.

2.3.2. Short-term temperature sums

Warm spells of relatively short duration may trigger UPE at a finer temporal resolution than a calen-

dar month. Therefore, an additional analysis considering short-term temperature sums was calculated using the method of Ring et al. (1983):

$$T_{\text{sum}} = \sum_{i=1}^n T_{\text{mean}} - x \quad (1)$$

where $i = 1, 2, 3, \dots, n$, and n is the number of days that are considered, $T_{\text{mean}} = (T_{\text{max}} + T_{\text{min}})/2$ = mean daily temperature, T_{max} = daily maximum temperature, T_{min} = daily minimum temperature, x = base temperature, i.e. 0°C ; negative values of T_{mean} were set to zero. Applying the zero method of Snyder et al. (1999), the base temperature was chosen to be zero for all phases, since this approach is believed to lead to quite reliable results.

The temperature sums were calculated for periods of different lengths (i.e. 1, 2, 3, 4, 5, 10, 15, 20, 25, 30, 35, 40, 45 and 50 d before the start of flowering in the city of Munich). The mean regional temperature sums (T_{sum}) were calculated using the daily temperatures averaged for Munich City and Weihenstephan.

The second summation was performed for the magnitude of UHI:

$$UHI_{\text{sum}} = \sum_{i=1}^n UHI \quad (2)$$

where i and n are as in Eq. (1), $UHI = (T_{\text{urban}} - T_{\text{rural}})$, T_{urban} = mean daily urban temperature, T_{rural} = mean daily rural temperature; negative values of UHI were set to zero.

These 2 temperature sums describing the short-term temperature conditions before urban flowering times of the plants were used for correlations with UPE.

2.4. Laboratory examination of pollen grains

Common methods for collecting airborne pollen include continuous volumetric pollen traps, e.g. the Burkhard trap or Hirst type trap (see Giesecke et al. 2010 for a complete overview of the pollen trapping methods). Nevertheless, due to variable atmospheric conditions, one cannot be sure of the origin of the pollen. Moreover, the phenological development stage (e.g. start of flowering or full flowering) of the tree that released the pollen is not known. Therefore, we harvested catkins of birch in 2009 during the start of flowering and at full flowering. Catkins were dried for 24 h and pollen was extracted by sieving catkins first with a $100 \mu\text{m}$ sieve followed by a $72 \mu\text{m}$ sieve. The amount of pollen per catkin was then weighed. Due to rapid growth in April, it was hard to investigate the same number of birches in urban and rural areas of Munich and Ingolstadt. Therefore, meaning-

ful analyses were available only for Munich: 10 samples for start of flowering (n: urban = 5, rural = 5); 26 samples for full flowering (n: urban = 12, rural = 14).

3. RESULTS

3.1. Field observations in 2009

3.1.1. Phenology of birch and horse chestnut

Table 3 shows the descriptive statistics of the observed phenophases in Munich and Ingolstadt in spring 2009. For leaf unfolding and flowering of birch, the dates of onset occurred on average between 9 April (day of year DOY: 99) and 16 April (DOY: 106) in Munich and ~2 d earlier in Ingolstadt, which can be explained by the lower elevation (370 m) of Ingolstadt compared to Munich (515 m). The temporal delay between the start of flowering (BBCH 61) and full flowering (BBCH 65) was <2 d, indicating rapid plant development in April 2009.

UPE for flowering and leaf unfolding of birch did not differ very much (UPE range: -1.1 d [flowering, BBCH 61, Ingolstadt] to $+0.6$ d [leaf unfolding, BBCH 11, Munich]). Furthermore, differences in Munich were not significant, although the differences for the start of flowering (UPE: -1.1 d, $p < 0.001$) and full flowering (UPE: -0.9 d, $p = 0.007$) in Ingolstadt were significant, albeit relatively small.

The start of flowering and full flowering of horse chestnut in Munich occurred from 23 April (DOY: 113, urban, BBCH 61) to 3 May (DOY: 123, rural, BBCH 65). In the city centre of Ingolstadt, first flowers had already opened on 21 April (DOY: 111) and the last day of full flowering was recorded on 29 April (DOY: 119) in the countryside.

In contrast to birch, differences in the mean dates of onset for the observed phenophases of horse chestnut were comparatively larger and ranged from -1.4 d (BBCH 61, Ingolstadt) to -3.0 d (BBCH 65, Munich). The UPE was greater for Munich than for Ingolstadt. Mean onset dates for flowering phenophases of horse chestnut for each city were significantly earlier than in their matching rural areas.

3.1.2. Pollen amount

Table 4 shows the mean amount of pollen per birch catkin (g) for different development stages and urban and rural areas in Munich. For the urban areas, the amount of pollen during the start of flowering was

Table 3. Descriptive statistics and tests of equality of means for dates of leaf unfolding and flowering of birch *Betula pendula* Roth and flowering of horse chestnut *Aesculus hippocastanum* L. expressed in DOY (day of the year) in urban and rural areas of Munich and Ingolstadt. BBCH (Biologische Bundesanstalt, Bundessortenamt und Chemische Industrie) codes—11: first leaves unfolded, 61: start of flowering, 65: full flowering. UPE: urban phenology effect, difference between urban and rural phenological onset dates (d); tests: *t*-test (for normally distributed data), Mann-Whitney (for data with non-normal distribution); significance (in UPE column): *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$, ns: not significant ($p > 0.05$)

BBCH	Type	n	Mean	Min.	Max.	SE	SD	UPE	Test
Birch									
Munich									
11	Urban	28	101.9	99.0	106.0	0.3	1.5	+0.6 ns	<i>t</i> -test
	Rural	13	101.3	99.0	103.0	0.4	1.3		
61	Urban	28	101.2	99.2	104.0	0.2	1.1	0 ns	<i>t</i> -test
	Rural	13	101.2	99.5	102.9	0.3	1.0		
65	Urban	28	102.8	100.5	105.0	0.2	0.9	0 ns	<i>t</i> -test
	Rural	13	102.8	101.5	104.3	0.2	0.9		
Ingolstadt									
11	Urban	28	99.4	96.8	102.0	0.3	1.5	-0.1 ns	<i>t</i> -test
	Rural	17	99.5	98.2	100.7	0.2	0.7		
61	Urban	28	98.8	97.5	100.5	0.2	0.9	-1.1 ***	<i>t</i> -test
	Rural	17	99.9	97.8	101.0	0.2	1.0		
65	Urban	28	100.6	99.0	102.0	0.2	1.0	-0.9 **	<i>t</i> -test
	Rural	17	101.5	99.5	103.5	0.3	1.1		
Horse chestnut									
Munich									
61	Urban	69	113.4	110.8	120.5	0.3	2.4	-2.5 ***	<i>t</i> -test
	Rural	24	115.8	111.0	120.9	0.6	2.7		
65	Urban	87	120.0	114.5	128.0	0.3	2.9	-3.0 ***	Mann-Whitney
	Rural	26	123.0	117.5	128.0	0.7	3.5		
Ingolstadt									
61	Urban	24	110.6	109.8	112.0	0.1	0.7	-1.4 ***	<i>t</i> -test
	Rural	31	112.0	110.0	115.0	0.3	1.5		
65	Urban	31	116.5	113.0	119.0	0.3	1.5	-2.9 ***	<i>t</i> -test
	Rural	31	119.3	117.0	123.0	0.3	1.6		

Table 4. Mean (\pm SD) amount of pollen per birch catkin (g) in April 2009 in urban and rural areas in Munich for different flowering phases (BBCH 61: start of flowering, BBCH 65: full flowering), ns: not significant ($p > 0.05$)

Phenophase	Urban	n	Rural	n	Difference	p
BBCH 61	0.013 ± 0.005	5	0.016 ± 0.013	5	-0.002	ns
BBCH 65	0.015 ± 0.004	12	0.007 ± 0.002	14	0.009	ns

slightly smaller than in rural areas (not significant). At full flowering, the differences were greater, with a higher amount of pollen per catkin in the city (not significant).

3.1.3. Climatic conditions

Fig. 2 (left y-axis) shows the mean daily temperatures from December 2008 to April 2009 for Munich City and the corresponding average for the standard

reference period 1961 to 1990. Temperatures for December 2008 to March 2009 were lower (by an average of 0.7°C) than the 1961 to 1990 mean for the same months. However, temperatures for April were 4.3°C higher and were the second highest April temperatures since the beginning of records in 1955. Therefore, this warm spell (especially during the first half of April 2009) with constantly high temperatures close to 15°C , can be seen as an extreme event.

For Munich, the average yearly temperature difference between Munich City (urban) and Weihenstephan (rural) for 1961 to 1990 was 1.6°C , and was greater for minimum (2.9°C) than for maximum temperatures (1.0°C) (not shown). A less pronounced UHI effect was observed in Munich from December 2008 to April 2009 (mean = 0.9°C) than during the reference period 1961–1990 (1.5°C). The highest deviation from the reference period was observed in December 2008: the UHI was only 0.6°C , which was 0.8°C smaller than the December 1961–1990 average (1.4°C).

3.2. Analyses of long-term phenological and temperature data

3.2.1. Urban phenology effect in Munich (1951 to 2008)

The mean differences in phenological onset dates between urban and rural areas (UPE) are shown in Table 5. Hazel had the most pronounced UPE for the period 1951 to

2008: first flowering averaged 4.7 d earlier in urban Munich. The smallest UPE was for flowering of forsythia (-3.5 d). The sub-period 1981 to 2008, when most of the recent climate warming occurred (IPCC 2007), had very similar UPE except for hazel where the mean UPE was -7.6 d. Nevertheless, temporal trends (b) of UPE were only significant for horse chestnut in 1981 to 2008, indicating a reduction in the difference between urban and rural dates. Note that the temperature trends, e.g. of February to April, for Munich City (1955 to 2008, $b = 0.039$, $p = 0.044$) and

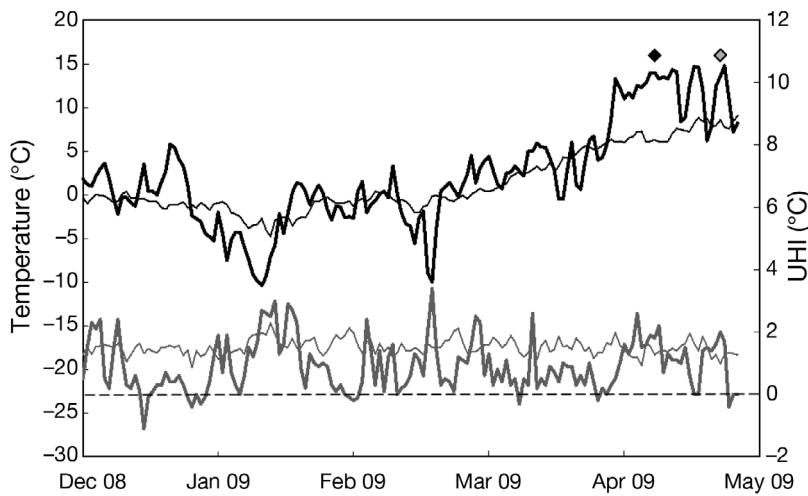


Fig. 2. Left axis (black curves): mean daily temperature in Munich City. Bold line: December 2008 to April 2009, thin line: standard reference period 1961 to 1990. Right axis (grey curves): mean daily urban heat island effect (UHI, temperature difference between Munich City [urban] and Weihenstephan [rural]). Bold line: December 2008 to April 2009, thin line: standard reference period 1961 to 1990, dashed line: UHI = 0. Diamonds: start of flowering (BBCH 61) for *Betula pendula* Roth (birch, black) and *Aesculus hippocastanum* L. (horse chestnut, grey) in the greater area of Munich in 2009

Table 5. Mean UPE (urban phenology effect; difference between urban and rural phenological onset dates in d), temporal trends (*b*) of the UPE and R^2 for flowering of hazel *Corylus avellana*, forsythia *Forsythia suspensa* and horse chestnut *Aesculus hippocastanum* (1951 to 2008 and 1981 to 2008) in the greater area of Munich. *** $p < 0.001$, ns: not significant ($p > 0.05$)

Flowering of	Period	Mean UPE	SD	SE	<i>b</i> (d yr ⁻¹)	R^2 (%)	<i>p</i>
Hazel	1951–2008	-4.71	10.91	1.53	-0.098	2.1	ns
	1981–2008	-7.63	13.50	2.88	0.240	1.8	ns
Forsythia	1951–2008	-3.47	4.46	0.64	0.014	0.3	ns
	1981–2008	-3.86	4.30	0.90	0.087	2.4	ns
Horse chestnut	1951–2008	-4.36	3.58	0.51	0.049	5.1	ns
	1981–2008	-3.82	3.50	0.75	0.319	50.2	***

Weihenstephan (1951 to 2008, *b* = 0.025, p = 0.006) did not differ significantly (p = 0.432), as tested using analysis of covariance (ANCOVA). The same was true for the period 1981 to 2008 (Munich City: *b* = 0.063, p = 0.071; Weihenstephan: *b* = 0.072, p = 0.039; ANCOVA: p = 0.844).

3.2.2. Long-term temperature data

Correlation analyses of the UPE for flowering of hazel, forsythia and horse chestnut with the temperature difference between Munich City (urban) and Weihenstephan (rural) (UHI) are shown in Table 6. The results show that for single months and 2 or 3 mo

periods, no significant correlation between these variables existed.

Similar findings were obtained from the correlations between UPE-values and the temperature differences from the 1961 to 1990 means for the selected climate stations (as a proxy for extreme temperatures) (see also Table 6). Only 2 significant correlations, for forsythia (February r = -0.329, p = 0.029) and horse chestnut (April r = 0.381, p = 0.009), were revealed. High temperatures in April were associated with a less pronounced UPE for horse chestnut. However, higher temperatures in February led to a more pronounced UPE for forsythia.

3.2.3. Short-term temperature sums

The analysis of short-term (1 to 50 d) temperature data indicated significant correlation coefficients for Tsum, especially between 10 to 30 d before the first flowering in the city. The temporal pattern of the correlation coefficients is shown in Fig. 3 (black lines). Flowering of hazel (Fig. 3a) had the weakest relationships, with most of the coefficients being nonsignificant; however, they were significant although weak for Tsum at 20 to 25 d before urban flowering (highest r = 0.345, p < 0.05, Tsum20). For forsythia (Fig. 3b) and horse chestnut (Fig. 3c), the magnitude of the coefficients were

notably higher and often significant, with respective maximum *r* values of 0.584 ($p \leq 0.001$) and 0.726 ($p \leq 0.001$) occurring around the 20th day before urban onset dates. The strongest correlation between UPE and Tsum was noted for horse chestnut at 20 d before urban flowering, as can be seen in Fig. 4: the higher the Tsum in the respective 20 d period, the less pronounced (close to 0 or even positive) was the UPE. The data for 2009 derived from the field study in Munich were added to Fig. 4 and are indicated by a diamond.

For UHI sums, the results (see Fig. 3, grey lines) were quite mixed: whereas no significant correlation was found for forsythia, the 15 d period before urban onset of hazel flowering showed a significant but

Table 6. Correlations of UPE (urban phenology effect; difference between urban and rural phenological onset dates in d) (1955 to 2008) for hazel *Corylus avellana* ($n = 47$), forsythia *Forsythia suspensa* ($n = 45$) and horse chestnut *Aesculus hippocastanum* ($n = 46$) with UHI (temperature difference between Munich City and Weihenstephan) and Tdev (temperature deviation from the standard reference period 1961 to 1990). ** $p < 0.01$, * $p < 0.05$, ns: not significant ($p > 0.05$)

	UHI cf. UPE				Tdev cf. UPE							
	Hazel		Forsythia		Horse chestnut		Hazel		Forsythia		Horse chestnut	
Dec–Feb	r -0.029	p ns	r -0.010	p ns	r -0.149	p ns	r -0.154	p ns	r -0.258	p ns	r 0.240	p ns
Jan–Mar	r 0.080	p ns	r 0.025	p ns	r -0.153	p ns	r -0.121	p ns	r -0.149	p ns	r 0.186	p ns
Feb–Apr	r 0.252	p ns	r 0.112	p ns	r -0.060	p ns	r -0.095	p ns	r -0.247	p ns	r 0.217	p ns
Jan–Feb	r -0.039	p ns	r -0.081	p ns	r -0.215	p ns	r -0.148	p ns	r -0.229	p ns	r 0.274	p ns
Feb–Mar	r 0.214	p ns	r 0.123	p ns	r -0.128	p ns	r -0.086	p ns	r -0.204	p ns	r 0.107	p ns
Mar–Apr	r 0.263	p ns	r 0.151	p ns	r 0.076	p ns	r -0.051	p ns	r -0.057	p ns	r 0.167	p ns
Dec	r 0.006	p ns	r 0.136	p ns	r 0.083	p ns	r -0.110	p ns	r -0.181	p ns	r 0.059	p ns
Jan	r -0.138	p ns	r -0.122	p ns	r -0.111	p ns	r -0.123	p ns	r 0.044	p ns	r 0.225	p ns
Feb	r 0.083	p ns	r -0.002	p ns	r -0.228	p ns	r -0.109	p ns	r -0.326	*	r 0.189	p ns
Mar	r 0.248	p ns	r 0.206	p ns	r 0.052	p ns	r -0.030	p ns	r 0.071	p ns	r -0.067	p ns
Apr	r 0.158	p ns	r 0.037	p ns	r 0.065	p ns	r -0.046	p ns	r -0.197	p ns	r 0.381	**

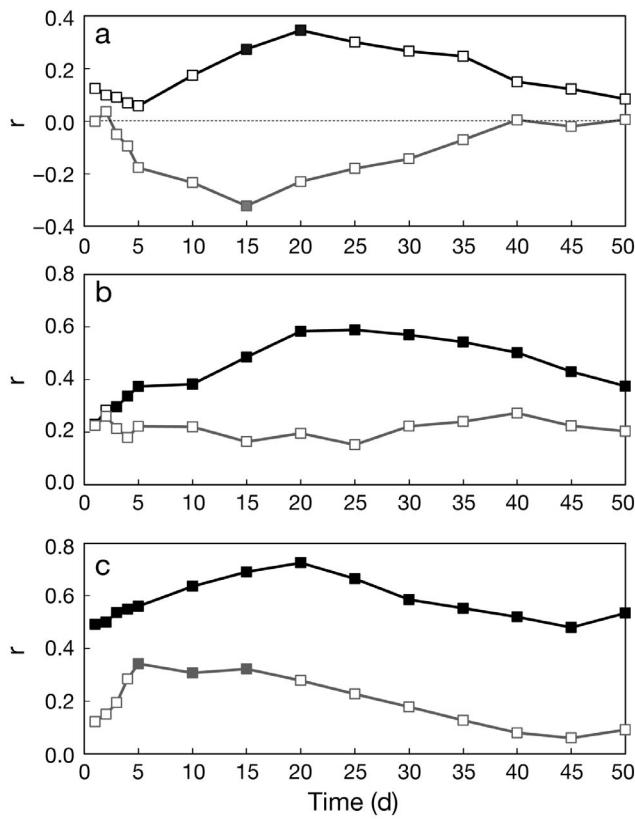


Fig. 3. Temporal development (from 1 to 50 d before urban flowering) of correlation coefficients (r) of temperature variables (temperature sums [T_{sum} , black line] and urban heat island effect sums [UHI_{sum} , grey line]) with UPE (urban phenology effect) for (a) hazel *Corylus avellana*, (b) forsythia *Forsythia suspensa* and (c) horse chestnut *Aesculus hippocastanum*. UPE is the difference between urban and rural phenological onset dates (in d). Filled squares: significant correlation coefficients ($p \leq 0.05$)

weak relationship with UPE ($r = -0.323$, $p \leq 0.05$). For horse chestnut, there were 3 significant coefficients ($p \leq 0.05$), with the highest values being calculated at 5 d before urban flowering (UHI_{sum5} : $r = 0.343$, $p \leq 0.05$). For hazel, the significant coefficient was negative, i.e. a greater difference in temperature between the urban and rural stations induced a more pronounced UPE (increasing negative values, Fig. 3a). In contrast, horse chestnut showed a positive relationship, i.e. a greater temperature difference led to a less pronounced difference in flowering times (i.e. UPE less negative or even positive).

4. DISCUSSION

4.1. Field observations in 2009

We considered April 2009 as an extreme event since it had the second warmest temperature on record. This fact implied rapid plant development, with trees flowering almost simultaneously and differences in phenology between urban and rural sites being reduced. This was especially true for the early spring phenophases of birch. The later spring phenophases of horse chestnut were not affected in this way. However, the observation data of 2009 shows a smaller UPE (compared to other years) with high temperature sums, e.g. for the 20 d before urban onset (see Fig. 4). Therefore, we suggest that differences in flowering dates between urban and rural environments are strongly affected by annual weather patterns.

4.2. Differences in urban–rural phenology (1951 to 2008)

The mean differences between urban and rural phenology (UPE, 1951 to 2008) showed earlier flowering of hazel, forsythia and horse chestnut in the city. However, temporal trends showed that these differences did not change significantly, apart from a reduction in the UPE for horse chestnut in 1981 to 2008. The increases in temperature measured at the urban and rural stations did not differ significantly. With climate warming advancing spring phenology (Fitter & Fitter 2002, Parmesan & Yohe 2003, Root et al. 2003, Badeck et al. 2004, Menzel et al. 2006) in both urban and rural areas (see ANCOVA in Section 3.2.1), one could hypothesise that the UPE should be more or less stable as long as the UHI is not increasing. Consequently, we conclude that phenological differences between the city and its surroundings cannot adequately be explained by temperature means and their trends in spring. Weather conditions of only a short duration (extremes) may often be overlooked when using mean temperature data of a longer period (monthly or longer), although they can influence the timing of flowering (Jentsch et al. 2007).

4.3. Long-term vs. short-term temperature data

The results of the phenological observations for April 2009 showed little difference in flowering

dates (especially in flowering and leaf unfolding of birch) between urban and rural areas of Munich and Ingolstadt. To evaluate these results, we explored the suitability of the UHI and the temperature deviations from 1961 to 1990 in explaining variations in urban and rural flowering dates using DWD time series (1955 to 2008) with correlation analyses. However, a dependency of the variables calculated on a monthly or longer resolution could not be proven.

To overcome the limitations of using crude monthly data, temperature sums of shorter periods (1 to 50 d before urban onset) were used for correlation with UPE; results showed that shorter periods are more suitable in explaining the temporal variation in urban and rural phenology. However, our results also indicated that temperature differences between the city and its rural surroundings are not sufficient to explain the variations in urban–rural phenology. The contrasting directions of the relationship between UHI and UPE for hazel (significant negative correlation coefficients) and for horse chestnut (significant positive correlation coefficients) suggest that differences in temperature may not fully explain temporal patterns in urban and rural phenology. Therefore, only the correlations between UHI and UPE for hazel showed the expected direction: the greater the temperature difference between the city and the countryside, the greater the difference in phenology (i.e. UPE values more negative). This could be attributed to the temperature sensitivity of plants flowering early in the year (Ahas et al. 2002, Fitter & Fitter 2002). One reason for the inconclusive results might be that the urban and rural temperatures respectively recorded at Munich City and Weihenstephan do not adequately reflect the variations within the sites used in this study. However, we specifically discarded stations located at higher elevations and thus suggest that the compact nature of the study area (within a maximum of 35 km from Munich city centre) should result in only minor latitudinal or longitudinal effects on temperature. In addition,

we investigated gridded temperature data from the DWD to compare urban and rural temperature differences (results not shown) and found that the mean temperature data of the

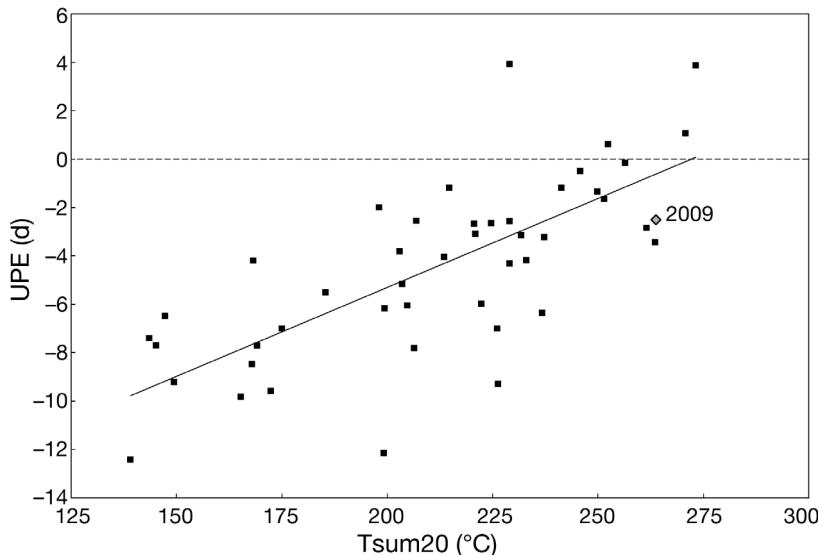


Fig. 4. Scatterplot of UPE (urban phenology effect; difference between urban and rural phenological onset dates in d) against temperature sums for a 20 d period before urban onset (Tsum20) for horse chestnut *Aesculus hippocastanum* in Munich (1955 to 2008). $R^2 = 0.5268$, regression equation: $y = 0.0735x - 20.001$. Grey diamond: data for the field study in 2009

urban and rural sites matched the temperatures recorded at the 2 climate stations. It is thus more likely that short warm spells could be more explanatory than the UHI which is mainly affected by general atmospheric circulation patterns. Temperature sums provided often significant and larger correlations than UHI, especially at 20 to 25 d before urban onset dates. Therefore, we suggest that warm spells during this period could result in negligible differences in flowering phenology ($UPE \approx 0$) or even earlier flowering in the countryside ($UPE > 0$).

Subsequently, we suggest that weather conditions lasting only a few days can effectively influence phenology, especially at the micro- and mesoscale. Extremely high temperatures are more likely to result in simultaneous flowering in urban and rural environments. Low temperatures, e.g. cold spells, can contribute to a longer delay of phenological onset times in the countryside compared to the city.

4.4. Linkage to climate variability

In response to previous temperature extremes, the biological effects of the heat wave of summer 2003 in the Swiss Alps were analysed by Jolly et al. (2005). Higher elevations were affected by increased growth and longer effective growing season; however, lower elevations exhibited reverse effects on plants, mainly due to an enhanced evaporative demand. Luterbacher et al. (2007) assessed the impacts of the exceptional warmth of autumn 2006 and winter 2007 in Europe. They observed that some plant species had a partial second flowering or an extended flowering season that lasted until the beginning of winter. Moreover, the first spring phases in 2007, e.g. flowering of snowdrop *Galanthus nivalis* L. or hazel, occurred distinctively earlier. Manipulation experiments conducted by Jentsch et al. (2009) showed that a 32 d period of drought advanced mid-flowering dates of grassland and heath species by 4 d on average and extended the flowering season by 4 d. Conversely, heavy rainfall led to a shortened flowering season.

However, these studies focused on weather conditions prevalent in summer or autumn and winter, but not on atypical high spring temperatures that we explored in our study. Analyses of phenological onset dates of birch and horse chestnut in 2009 suggested that anomalous warm periods in spring could lead to synchrony in plant development between urban and rural areas, with various effects on pollen and human health.

4.5. Pollen and human health

Besides the consistency in phenological onset dates of birch, no significant variation in birch pollen amount per catkin could also be found between Munich and its surrounding countryside in 2009.

Several studies reported a correlation between temperature and airborne pollen amount: Spieksma et al. (1995) found increased trends of annual sums of daily airborne pollen concentration in Basel, Leiden, London, Stockholm and Vienna. Rising birch pollen concentrations were also attributed to climate change by Frei & Gassner (1998) for Switzerland and Rasmussen (2002) for Denmark. Teranishi et al. (2000) demonstrated the existence of a significant relationship between mean temperatures of the previous July and total pollen amount on Japanese cedar for urban areas of Japan. Ziska et al. (2003) found that higher atmospheric pollen amounts of the herbaceous common ragweed in North American urban areas were associated with increasing CO_2 concentrations and air temperatures, both of which are related to urbanization and climate change.

These findings suggest that higher temperature could be a primary cause of greater pollen concentrations. Small differences in temperatures ($UHI \approx 0$), as observed in winter 2008 and spring 2009 between urban and rural areas of Munich, would be expected to lead to an equality of pollen grain abundance. However, the method for pollen sampling was used for the first time in this study, thus the influence of the warm spell in April 2009 cannot be definitely assessed. Having pollen information for 'normal' years with UHI and temperatures that are close to average values would allow a better understanding of the effect of extreme events on pollen production.

Since the development of both urban and rural catkins was rapid, pollen was released almost simultaneously in urban and rural Munich. The start of flowering in urban and rural sites in Ingolstadt only had a temporal lapse of 1.1 d. Therefore, allergic people in both the city and the countryside were affected starting almost the same day. Moreover, the temporal development from the start of flowering to full flowering was also rapid. Thus, the symptoms of birch allergy probably did not last long compared to other years when colder temperatures inhibit rapid development of catkins.

In general, asthmatic and pollen allergic individuals can benefit from the countryside by being exposed later to pollen. However, when warm spells lead to urban–rural synchrony, people do not have the option to be less exposed in the nearby country-

side. In contrast, during cold spells, there was a larger difference in onset dates between urban and rural areas. This would imply that people probably have the opportunity to spend pollen-free time in the countryside but might also suffer for longer due to a steady development and transport of pollen in the air from the city to the countryside and vice versa.

Altered onset dates of flowering and pollen season durations of certain allergenic plants could enhance disorders; e.g. an overlapping with the peak of another allergenic plant (Doi et al. 2010) could result in strengthening of symptoms and a decrease in allergy-free time. Earlier onsets of flowering could also imply interrupted pollen seasons due to colder and more humid weather conditions in late winter or early spring (D'Amato & Cecchi 2008), leading to perpetual minor and major ailments.

Few studies focused on the impacts of extreme weather conditions: analyses of grass pollen production and airborne pollen loads during summer 2003 (Gehrig 2006) showed that the pollen season in Switzerland started remarkably early, but was also shorter than average. Allergic people experienced several days with high pollen concentration and therefore had more intense symptoms. In addition, the usually less affected higher regions of the Alps were also characterised by a more severe pollen season. In addition, precipitation plays an important role in pollen release. While dry conditions support the release of pollen from anthers, high humidity or rainfall events may interrupt this process (D'Amato & Cecchi 2008). However, Gehrig (2006) found that extremely long periods of negative water balance in southern Switzerland in 2003 led to exceptionally low concentrations of pollen from *Rumex* spp., *Urtica* spp. and *Artemisia* spp. Climate variability is just one factor influencing the relationship between pollen and allergy but seems to have a great effect on allergic disorders, especially in terms of urban–rural comparison.

Since allergies are prevalent worldwide and affect both young and old people, phenological observations of allergenic plants and the effects of extremes are necessary to understand possible future risks and trends concerning this important human health issue. We have demonstrated homogeneity of flowering dates of allergenic plants due to warm spells at the micro- and mesoscales. For allergic people, the favourable consequence of shorter pollen seasons is contrasted with the loss of opportunity to escape to the countryside to experience a lower allergen load. Further investigations should examine a combination of pollen amounts produced by single trees and air-

borne pollen counts from populations, as this will make a valuable contribution for assessing human health impacts of climate change.

Acknowledgements. We thank our phenological observers D. Rottler, H. Seifert, K. Katheder, S. Gubo, Y. Munich and R. Behrendt for their efforts during spring 2009. S.C.J. gratefully acknowledges the support of the TUM Graduate School's Faculty Graduate Center Weihenstephan at the Technische Universität München. The research conducted in this study was supported by grant ME 179/3-1 of the Deutsche Forschungsgemeinschaft (DFG).

LITERATURE CITED

- Ahas R, Aasa A, Menzel A, Fedotova VG, Schleifinger H (2002) Changes in European spring phenology. *Int J Climatol* 22:1727–1738
- Alexander LV, Zhang X, Peterson TC, Caesar J and others (2006) Global observed changes in daily climate extremes of temperature and precipitation. *J Geophys Res* 111:D05109. doi:10.1029/2005JD006290
- Badeck FW, Bondeau A, Böttcher K, Doktor D, Lucht W, Schaber J, Sitch S (2004) Responses of spring phenology to climate change. *New Phytol* 162:295–309
- Baumgartner A (1952) Zur Phänologie von Laubholzern und ihre Anwendung bei lokalklimatischen Untersuchungen. *Ber Dtsch Wetterd US-Zone* 42:69–73
- Beggs PJ (2004) Impacts of climate change on aeroallergens: past and future. *Clin Exp Allergy* 34:1507–1513
- Braun-Fahrlander C, Gassner M, Grize L, Neu U and others (1999) Prevalence of hay fever and allergenic sensitization in farmer's children and their peers living in the same rural community. SCARPOL team. Swiss study on childhood allergy and respiratory symptoms with respect to air pollution. *Clin Exp Allergy* 29:28–34
- Chuine I (2010) Why does phenology drive species distribution? *Philos Trans R Soc Lond B* 365:3149–3160
- D'Amato G (2000) Urban air pollution and plant-derived respiratory allergy. *Clin Exp Allergy* 30:628–636
- D'Amato G, Cecchi L (2008) Effects of climate change on environmental factors in respiratory allergic diseases. *Clin Exp Allergy* 38:1264–1274
- Doi H, Takahashi M, Katano I (2010) Genetic diversity increases regional variation in phenological dates in response to climate change. *Glob Change Biol* 16:373–379
- EEA (European Environment Agency) (2000) CORINE land cover 2000. Available at www.eea.europa.eu/themes/landuse/clc-download
- Estrella N, Menzel A, Krämer U, Behrendt H (2006) Integration of flowering dates in phenology and pollen counts in aerobiology: analysis of their spatial and temporal coherence in Germany (1992–1999). *Int J Biometeorol* 51: 49–59
- Fink A, Brücher T, Krüger A, Leckebusch G, Pinto J, Ulbrich U (2004) The 2003 European summer heatwaves and drought—synoptic diagnosis and impacts. *Weather* 59: 209–216
- Fitter AH, Fitter RSR (2002) Rapid changes in flowering times in British plants. *Science* 296:1689–1691
- Franken E (1955) Der Beginn der Forsythienblüte 1955 in Hamburg. Ein Beitrag zur Phänologie der Großstadt. *Meteorol Rundsch* 8:113–114

- Frei T, Gassner E (2008) Climate change and its impacts on birch pollen quantities and the start of the pollen season: an example from Switzerland for the period 1969–2006. *Int J Biometeorol* 52:667–674
- Gehrig R (2006) The influence of the hot and dry summer 2003 on the pollen season in Switzerland. *Aerobiologia* 22:27–34
- Giesecke T, Fontana SL, van der Knaap WO, Pardoe HS, Pidek IA (2010) From early pollen trapping experiments to the Pollen Monitoring Programme. *Veg Hist Archaeobot* 19:247–258
- Grimm NB, Stanley HF, Gobubiewski NE, Redman CL, Wu J, Bai X, Briggs JM (2008) Global change and the ecology of cities. *Science* 319:756–760
- IPCC (2007) Climate change 2007. Impacts, adaptions and vulnerability. Contributions of Working Group II to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge University Press, Cambridge
- Jentsch A, Kreyling J, Beierkuhnlein C (2007) A new generation of climate change experiments: events, not trends. *Front Ecol Environ* 5:365–374
- Jentsch A, Kreyling J, Boettcher-Treschkow J, Beierkuhnlein C (2009) Beyond gradual warming: extreme weather events alter flower phenology of European grassland and heath species. *Glob Change Biol* 15:837–849
- Jolly WM, Dobbertin M, Zimmermann NE, Reichstein N (2005) Divergent vegetation growth responses to the 2003 heat wave in the Swiss Alps. *Geophys Res Lett* 32: L18409. doi:10.1029/2005GL023252
- Landsberg HE (1981) The urban climate. Academic Press, New York
- Luterbacher J, Liniger MA, Menzel A, Estrella N and others (2007) Exceptional European warmth of autumn 2006 and winter 2007: historical context, the underlying dynamics, and its phenological impacts. *Geophys Res Lett* 34:L12704. doi:10.1029/2007GL029951
- Matzarakis A (2001) Die thermische Komponente des Stadtklimas. Berichte des Meteorologischen Institutes der Universität Freiburg
- Meehl GA, Tebaldi C (2004) More intense, more frequent, and longer lasting heat waves in the 21st century. *Science* 305:994–997
- Meier U (ed) (2001) Entwicklungsstadien mono- und dikotyler Pflanzen. BBCH-Monograph. Biologische Bundesanstalt für Land und Forstwirtschaft, Berlin
- Menzel A, Sparks TH, Estrella N, Koch E and others (2006) European phenological response to climate change matches the warming pattern. *Glob Change Biol* 12: 1969–1976
- Mimet A, Pellissier V, Quénol H, Aguejdad R, Dubreuil V, Rozé F (2009) Urbanisation induces early flowering: evidence from *Platanus aceriflora* and *Prunus cerasus*. *Int J Biometeorol* 53:287–298
- Oke TR (1973) City size and the urban heat island. *Atmos Environ* 7:769–779
- Oke TR (1987) Boundary layer climates, 2nd edn. Methuen, London
- Parmesan C, Yohe G (2003) A globally coherent fingerprint of climate change impacts across natural systems. *Nature* 421:37–42
- Rasmussen A (2002) The effects of climate change on the birch pollen season in Denmark. *Aerobiologia* 18: 253–265
- Ring DR, Harris MK, Jackman JA, Henson JL (1983) A Fortran computer program for determining start date and base temperature for degree-day models. The Texas Agricultural Experiment Station Bull MP-1537, The Texas University System, College Station, TX
- Ring J, Krämer U, Schäfer T, Behrendt H (2001) Why are allergies increasing? *Curr Opin Immunol* 13:701–708
- Root TL, Price JT, Hall KR, Schneider SH, Rosenzweig C, Pounds AJ (2003) Fingerprints of global warming on wild animals and plants. *Nature* 421:57–60
- Rosenzweig C, Karoly D, Vicarelli M, Neofotis P and others (2008) Attributing physical and biological impacts to anthropogenic climate change. *Nature* 453:353–357
- Rötzer T, Wittenzeller M, Häckel H, Nekovar J (2000) Phenology in central Europe—differences and trends of spring phenophases in urban and rural areas. *Int J Biometeorol* 44:60–66
- Schaber JA (2003) The R pheno package: auxiliary functions for phenological data analysis. Available at <http://cran.r-project.org/web/packages/pheno/index.html>
- Schär C, Vidale PL, Lüthi D, Frei C, Häberli C, Liniger MA, Appenzeller C (2004) The role of increasing temperature variability in European summer heatwaves. *Nature* 427: 332–336
- Snyder RL, Spano D, Cesarcic C, Duce P (1999) Determining degree-day thresholds from field observations. *Int J Biometeorol* 42:177–182
- Spieksma FTM, Emberlin JC, Hjelmroos M, Jäger S, Leuschner RM (1995) Atmospheric birch (*Betula*) pollen in Europe: trends and fluctuations in annual quantities and the starting dates of the seasons. *Grana* 34:51–57
- Stott PA, Stone DA, Allen MR (2004) Human contribution to the European heatwave of 2003. *Nature* 432:610–614
- Teranishi H, Kenda Y, Katoh T, Kasuya M, Oura E, Taira H (2000) Possible role of climate change in the pollen scatter of Japanese cedar *Cryptomeria japonica* in Japan. *Clim Res* 14:65–70
- Trajdíl-Hoffmann C, Kasche A, Menzel A, Jakob T, Thiel M, Ring J, Behrendt H (2003) Impact of pollen on human health. More than allergen carriers? *Int Arch Allergy Immunol* 131:1–13
- Trajdíl-Hoffmann C, Jakob T, Behrendt HJ (2009) Determinants of allergenicity. *J Allergy Clin Immunol* 123: 558–566
- White MA, Ramakrishna R, Nemani R, Thornton PE, Running SW (2002) Satellite evidence of phenological differences between urbanised and rural areas of the eastern United States deciduous broadleaf forest. *Ecosystems* 5: 260–273
- WHO (World Health Organisation) (2003) Phenology and human health: allergic disorders. Report of a WHO meeting, 16–17 January 2003, Rome, Italy
- Zhang X, Friedl MA, Schaaf CB, Strahler AH, Schneider A (2004) The footprint of urban climates on vegetation phenology. *Geophys Res Lett* 31:L12209. doi:10.1029/2004GL020137
- Ziska LH, Gebhard DE, Frenz DA, Faulkner S, Singer BD, Straka JG (2003) Cities as harbingers of climate change: common ragweed, urbanization, and public health. *J Allergy Clin Immunol* 111:290–295