Chapter 4
Monitoring, Modelling and Forecasting of the Pollen Season

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Abstract The section about monitoring covers the development of phenological networks, remote sensing of the season cycle of the vegetation, the emergence of the science of aerobiology and, more specifically, aeropalynology, pollen sampling instruments, pollen counting techniques, applications of aeropalynology in agriculture and the European Pollen Information System. Three data sources are directly related with aeropalynology: phenological observations, pollen counts and remote sensing of the vegetation activity. The main future challenge is the assimilation of these data streams into numerical pollen forecast systems. Over the last decades

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consistent monitoring efforts of various national networks have created a wealth of pollen concentration time series. These constitute a nearly untouched treasure, which is still to be exploited to investigate questions concerning pollen emission, transport and deposition. New monitoring methods allow measuring the allergen content in pollen. Results from research on the allergen content in pollen are expected to increase the quality of the operational pollen forecasts.

In the modelling section the concepts of a variety of process-based phenological models are sketched. Process-based models appear to exhaust the noisy information contained in commonly available observational phenological and pollen data sets. Any additional parameterisations do not to improve model quality substantially. Observation-based models, like regression models, time series models and computational intelligence methods are also briefly described. Numerical pollen forecast systems are especially challenging. The question, which of the models, regression or process-based models is superior, cannot yet be answered.

Keywords Aerobiology • Aeropalynology • Phenology • Pollen modelling • Phenological modelling

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List of Acronyms

AFEDA	French Association for Ragweed Study
ANN	Artificial Neural Networks
ARIMA	Autoregressive Integrated Moving Average
AVHRR	Advanced Very High Resolution Radiometer
CART	Classification and Regression Trees
COST725	COST Action 725: Establishing a European Phenological Data Platform for Climatological Applications
CAgM	Commission for Agrometeorology
DEM	Digital Elevation Model
DWD	Deutscher Wetterdienst
EAN	European Aeroallergen Network
ELISA	Enzyme-linked Immunosorbent Assay
EUMETNET	The Network of European Meteorological Services
GIMMS	Global Inventory Modeling and Mapping Studies
IC	Computational Intelligence
IAA	International Association for Aerobiology
IBP	International Biological Programme
INERIS	Industrial Environment and Risks National Institute
IPG	International Phenological Garden
Landsat TM	Landsat Thematic Mapper, Satellite
LUT	Look Up Table
MODIS	Moderate Resolution Imaging Spectroradiometer
NDVI	Normalised Difference Vegetation Index
NHMS	National Hydrometeorological Services
NOAA	National Oceanic and Atmospheric Administration
PCR	Polymerase Chain Reaction
PEP725	Pan European Phenological Database
PM	Particulate Matter
RMSE	Root Mean Square Error
SPOT	Satellite Pour l'Observation de la Terre
SOM	Self-Organising Maps
SVMs	Support Vector Machines
TSM	Temperature Sum Model
UM	Use and Management of Biological Resources
WCDMP	World Climate Data and Monitoring Programme
WCP	World Climate Programme
WIBS	Wide-Issue Bioaerosol Spectrometer
WMO	World Meteorological Organisation

4.1 Introduction

Input for the aeropalynological core topics of monitoring, modelling and forecasting of the pollen season have been drawn from an array of disciplines and cast into this review chapter. History, current state, recent developments and future prospects of phenological and pollen counting networks have been reviewed in the first section. Both, phenological observations and pollen counts collected by various networks form the observational basis of any quantitative description of the relationship between the seasonal cycle of plants and their atmospheric environment. The various modelling strategies and their applications are extensively elucidated in the second section.

Although phenology and aeropalynology experienced separate historical developments, they meet here and share the same models, which forecast the beginning of flowering and the beginning of pollen shedding, respectively. Links between aeropalynology and phenology are scattered throughout this review, but are explicitly summarised under the headings of "Phenological observations" and "Process-based phenological models":

- The natural relationship between phenology and aeropalynology may be expressed in the assumption that the beginning of flowering equals the beginning of pollen shedding into the atmosphere. Pollen emission modelling can benefit much from the knowledge, observations and modelling of flowering phenology.
- The effort to maintain a phenological network is less than to maintain a pollen observing network. Therefore in many regions the spatial density of phenological networks is higher than that of pollen traps and phenological time series are longer than pollen concentration time series. Thus it is possible to infer something about the pollen problem from phenology with a higher spatial density and/or further back in time than it would be possible based on pollen data alone.
- Phenology has made substantial progress during the last decade in various aspects like phenological modelling, satellite observation of the vegetation cycle, relation with climate variability and others, so that the problem of pollen allergenicity now can benefit from that progress in phenological research.

The recent boost in the interest in phenology as climate impact factor has been motivated by the discussion about human influence on climate, which became manifest in an increasing flood of publications with phenological background and an extended chapter of the 4AR about the role of phenology in climate impact research (Rosenzweig et al. 2007). Aeropalynology benefits a great deal from the enhanced interest in phenology within the frame of the climate impact discussion. Both fields of interest have more in common than it appears at first glance, a factor, which has still to be exploited.

4.2 Monitoring

4.2.1 Phenological Observations

4.2.1.1 Monitoring Networks

Systematic phenological observation can now look back on a history, which reaches back as far as the eighteenth century, when Carolus Linnaeus started the first phenological network in Sweden and Finland 1750–1752 (Nekovar et al. 2008). A few decades later phenological observations were also included in the first pan European meteorological network of the Societas Meteorologicae Palatinae (1781–1792). In the mid-nineteenth century the first national networks began their operation in the USA and Europe, although in most cases only for a limited time period. Ihne and Hoffmann managed to run their phenological network in a number of European countries over 1883–1941 (Fig. 4.1). During the 1950s the idea of International Phenological Gardens with a cloned set of plants was born, which resulted in a still operating and expanding phenological network in Europe. During the same period most national phenological networks began collecting phenological observations systematically, as recommended by the Commission for Agrometeorology (CAgM) of the World Meteorological Organisation (WMO).

A detailed global overview about phenological networks can be found in Schwartz (2003) and Koch (2010), whereas Nekovar et al. (2008) summarise the current situation in Europe.

National Monitoring Networks

Phenological research relies on phenological observations, collected mostly by national meteorological and hydro-meteorological services (NMHS). Phenological data collection with its rather small data volume has been usually running unobtrusively alongside the main stream collection of meteorological and climate data and thus survived in many NMHSs the ups and downs of the interest in phenological science through time. Another advantage of NMHSs is their experience in running station networks, quality controlling the incoming data, digitising and storing them on appropriate devices. Due to the efforts of COST Action 725 and the growing concern about climate change impacts, the Commission for Climatology (CCl) of the WMO now recommends the NHMS to organise phenological observations, whereas the World Climate Data and Monitoring Programme (WCDMP) and World Climate Programme (WCP) are working to stimulate and coordinate climate and climate impact monitoring activities around the world, which include phenological observations (www.omm.urv.cat/media/documents/WMO.pdf).

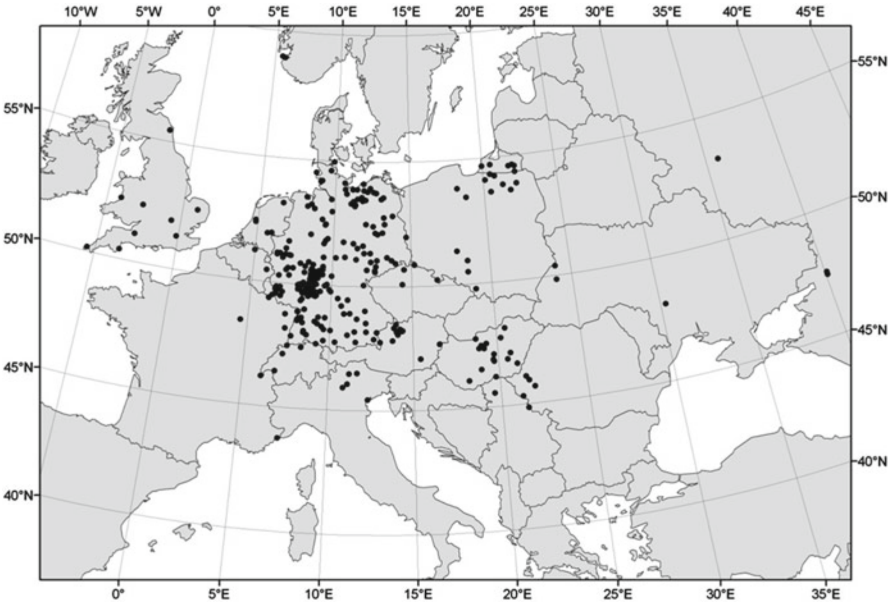

Fig. 4.1 Stations of the Hoffmann–Ihne phenological network from 1883 to 1941. Only stations with a minimum number of five observations are being displayed. The historical phenological database HPDB is maintained by the DWD (after Koch et al. 2008)

The recently published report of the COST Action 725 summarises the information about national European phenological networks (Nekovar et al. 2008). A few "phenophases", which are relevant for pollen allergies, entered the COST Action 725 data base. Here is a list of plants shedding allergenic pollen with a useful number of observations of phenological events (beginning of flowering) in this data base: Norway maple (*Acer platanoides),* horse chestnut (*Aesculus hippocastanum*), black alder (*Alnus glutinosa*), meadow foxtail (*Alopecurus pratensis*)*,* mugwort (*Artemisia vulgaris*), birch (*Betula pendula*), hazel (*Corylus avellana*), forsythia (*Forsythia suspensa*), ash (*Fraxinus excelsior*) and goat willow *(Salix caprea*).

An increasing number of national weather services and other organisations interested in nature observation have been creating internet-based observation networks, where volunteers can enter their georeferenced phenological observations (Table 4.1). Some weather services merge the data from their traditional network and the entries from the web.

International Monitoring Networks and Data Collection Initiatives

Contrary to national networks, the distribution of plants is not influenced by national borders. There exist two basic strategies to overcome the problem of national differences in observational methods, either by an international network in the first place or by merging national networks.

Table 4.1 List of web-based phenological observational networks

Number	Country	Name	Web address
1	The Netherlands	Natuurkalender	http://www.natuurkalender.nl
2	UK	Nature's Calendar	http://www.naturescalendar.org.uk/
3	Ireland	Nature's Calendar	http://www.biology.ie
4	USA	National Phenology Network	http://www.usanpn.org/?q=home http://www.usanpn.org/participate/observe
5	USA	Appalachian Mountain Club	http://www.outdoors.org/conservation/mountainwatch/index.cfm
6	USA	Project Budburst	http://www.windows.ucar.edu/citizen_science/budburst/results.php
7	Canada	Alberta Plantwatch	http://plantwatch.fanweb.ca/
8	Canada	PlantWatch	http://www.naturewatch.ca/english/plantwatch/intro.html
9	Austria	Phänologie	http://zacost.zamg.ac.at/phaeno_portal/
10	Sweden	Svenska fenologi-nätverket	http://www.blommar.nu/index.php
11	Australia	ClimateWatch	http://www.climatewatch.org.au/

The International Phenological Gardens (IPGs), for instance, were thought to obtain comparable and standardised large-scale phenological observations across Europe (Chmielewski 2008). With the same idea in mind, the Global Phenological Monitoring has been launched (http://www.agrar.hu-berlin.de/struktur/institute/pfb/struktur/agrarmet/phaenologie/gpm).

National data collection initiatives achieve their final value only after a number of national networks get merged over larger areas. Unfortunately, such merging efforts are only few because of a number of difficulties. COST Action 725 "Establishing a European Phenological data Platform for Climatological Applications" (2004–2009) aimed at creating a European reference data set of phenological observations that can be used for climatological purposes (Koch et al. 2005; Nekovar et al. 2008). The proposal for a follow up of COST725 so called "PEP725" (Pan European Phenological Database) was accepted by EUMETNET, has started in 2010 and will run over 5 years. PEP725 will maintain and update the COST initiated phenological database. Additionally, it will incorporate phenological data from before 1951 and develop better quality checking procedures. PEP725 will ensure an open access to the database for research and education. An attractive webpage will make phenology and climate impacts on vegetation more visible for the public, enabling a real time monitoring of vegetation development.

The European Phenology Network (http://www.pik-potsdam.de/~rachimow/epn/html/frameok.html) represents a broad based compilation of meta-information on phenological and related networks across the world.

Monitoring for Special Scientific Studies

For some research projects, special phenological observational data sets are required, because observations from ordinary networks are insufficient, not applicable or not available. Special networks are operated at a limited number of observational points and over a short period of time. For instance, this was the case for the larch phenological study in the Western Alpine Aosta valley by Migliavacca et al. (2008), where the influence of elevation and topography on the phenology of larch (*Larix decidua)* was studied. Ziello et al. (2010) linked phenological, meteorological and palynological data along an altitudinal gradient in the German Alps. The study of the flowering phenology of herbaceous plants in a lawn community required a special observational setup (Marletto et al. 1992) as did the observation of the beginning of male flowering of trees of the cypress family (*Cupressaceae)* for pollen modelling purposes (Torrigiani Malaspina et al. 2007).

Monitoring for Pollen Forecasting Purposes

In the traditional phenological monitoring setup the observational sheets are returned to the network operator at the end of each year. For more immediate information on the state of the vegetation, some network operators introduced a rapid information system (e.g. Sofortmeldenetz of the German and Swiss weather services). Such immediately transmitted phenological information supports the pollen forecast system of the German weather service, for instance. Assuming that the observers enter their observed entry dates immediately, information on the current state of the vegetation can be derived from the web-based networks (Table 4.1). Remote sensing systems, like satellites and real time digital cameras, can also serve the same purpose, but are still to be included into the operational procedures. Likewise, assimilation systems, which consistently merge the observational real time data into phenological and pollen dispersion models, still have to be developed (Stöckli et al. 2008).

4.2.1.2 Remote Sensing

Normalised Difference Vegetation Index (NDVI)

Live green plants absorb solar radiation in the photosynthetically active spectral region (400–700 nm), which they use as a source of energy for photosynthesis. At the same time leaf cells do not absorb but reflect and transmit solar radiation in the near-infrared spectral region. This large contrast in reflectance properties between red and near-infrared spectral regions is unique for photosynthetically active plants, and can be used by remote sensing sensors to distinguish them from other land cover types as soil, bare rock and snow.

Accordingly, phenological changes during the growing season can be studied by examining changes in the remote sensing-based Normalised Difference Vegetation index (NDVI) value. The NDVI is defined as:

$$\mathrm{NDVI} = (\mathrm{Ch2} - \mathrm{Ch1}) / (\mathrm{Ch2} + \mathrm{Ch1}), \tag{4.1}$$

where Ch1 and Ch2 represent reflectance measured in the near infrared and red channels, respectively (Lillesand and Kiefer 1994). NDVI quantifies the contrast between red surface reflectance, which decreases with chlorophyll content, and near infrared surface reflectance, which increases with leaf area index and crown density. NDVI of an area containing a dense green vegetation canopy will tend to have high positive values (typically 0.6–0.8); more sparsely vegetated areas will have lower values while clouds and snow fields will be characterised by negative values of the NDVI index.

Atmospheric noise in the NDVI caused by clouds, dust and aerosols is generally negatively biased. This is due to the additive path radiance, which causes an increase in red reflectance, while lower atmospheric transmission reduces near infrared reflectance (Guyot et al. 1989). Maximum value compositing (Holben 1986) is a common method of minimising such noise. In this method, only the highest NDVI value in a predefined compositing period (typically 15–16 days) is retained. This results in fewer but more reliable NDVI values representing the time series.

Satellite Sensors

Maximum value composite NDVI datasets with global coverage and bi-monthly compositing period have been created using data from sensors with large swath widths as the National Oceanic and Atmospheric Administration (NOAA) Advanced Very High Resolution Radiometer (AVHRR) and the VEGETATION sensor aboard Satellite Probatoire d'Observation de la Terre (SPOT). The starting dates of these time series are 1981 and 1998, respectively, and the products are available at spatial resolutions of 8 and 1 km, respectively.

Since 2000, NDVI products have been available from the Moderate Resolution Imaging Spectroradiometer (MODIS) aboard the Terra and Aqua satellites. Compared with the NOAA AVHRR GIMMS dataset, MODIS NDVI data have improved calibration and atmospheric correction, a spatial resolution of 236 m and compositing intervals of 16 days. This medium-resolution dataset opens new possibilities within global phenological monitoring.

Recently, satellite series as Formosat-2, Komsat-2, and RapidEye were launched. This new generation of satellite sensors, with both high temporal and spatial resolution (<10 m), opens new possibilities for local phenological monitoring.

Phenological Observations by Satellites

Satellite image-aided analysis of phenology of natural vegetation provides spatially complete coverage that can be used to interpolate traditional ground-based phenological observations, and NDVI has evolved as the primary tool for monitoring changes in vegetation activity. Probably the most commonly used long-time dataset is the Global Inventory Modeling and Mapping Studies (GIMMS) dataset based on the AVHRR instrument (Tucker et al. 2005). It has been used by many researchers to study the effect of global climate change on phenological timing and primary production (e.g. Myneni et al. 1997; Walker et al. 2003; Stöckli and Vidale 2004).

Karlsen et al. (2006, 2007) mapped the onset of the growing season of the whole of Fennoscandia by applying the GIMMS-NDVI dataset, where the onset of season was well correlated with the phenophase "onset of leafing of birch". They compared the NDVI-defined start of growing season with registrations of the onset of leafing of birch at 15 phenological registration sites across Fennoscandia. Most of the stations (13 out of 15) showed a moderately high correlation ($r^2=0.22$–0.65) between field data and the NDVI-defined start of growing season. Four of the stations had 20- or 21-year-long time-series. For these stations, the mean coefficient of determination (r^2) between the start of growing season and the onset of leafing of birch was 0.39 ($p<0.05$). For all stations except one, the mean time span between the NDVI-defined start of growing season and the onset of leafing of birch was less than 1 week, and the root mean square error between field data and NDVI data was less than 10 days for all stations. For bi-monthly maximum value composited NDVI time series this is probably as close as it is possible to get. To decrease the difference it is necessary to use daily NDVI data.

Birch Flowering Measured by Satellites

Onset of flowering of birch and leaf-bud burst of birch are well correlated. Linkosalo (1999, 2000) found in southern Finland that the difference in time from male flowering to the first date of bud burst is only 1.1 days with male flowering occurring first. This indicates that the phenophase observed as leaf-bud burst could be used to determine the timing of local birch pollen release. Also, since bud burst of birch is accurately measured by remote sensing, measurements of NDVI could be used to determine the timing of local birch pollen release. Høgda et al. (2003) used the GIMMS-NDVI dataset, correlated it with birch pollen measurements from five stations, and found correlation values (r) in the range from 0.55 to 0.85. They also found trends in the timing of the start of pollen seasons, consistent with effects of climate change.

Because of its mountainous topography, deep fjords, and long distance from north to south, Norway is climatically and ecologically very diverse. The number of pollen traps is also relatively low so developing pollen forecasts in Norway is a challenging task. Karlsen et al. (2009a) used MODIS-NDVI satellite data with 16-days time resolution and 236 m spatial resolution to map the average onset of birch flowering in Norway for the 2000 to 2007 period (Fig. 4.2).

In those studies, they found high correlation with phenological field data of onset of leafing of birch, as well as with the date when the annual birch pollen sum reaches 2.5% of the annual total from the ten Burkard traps across Norway. Accordingly, the satellite data can be used to determine the best location of the pollen traps and define the area with similar timing for start of birch pollen seasons as each trap.

Karlsen et al. (2009a) also identified the NDVI threshold for each pixel when the onset of birch flowering occurred. On this basis they developed a model for

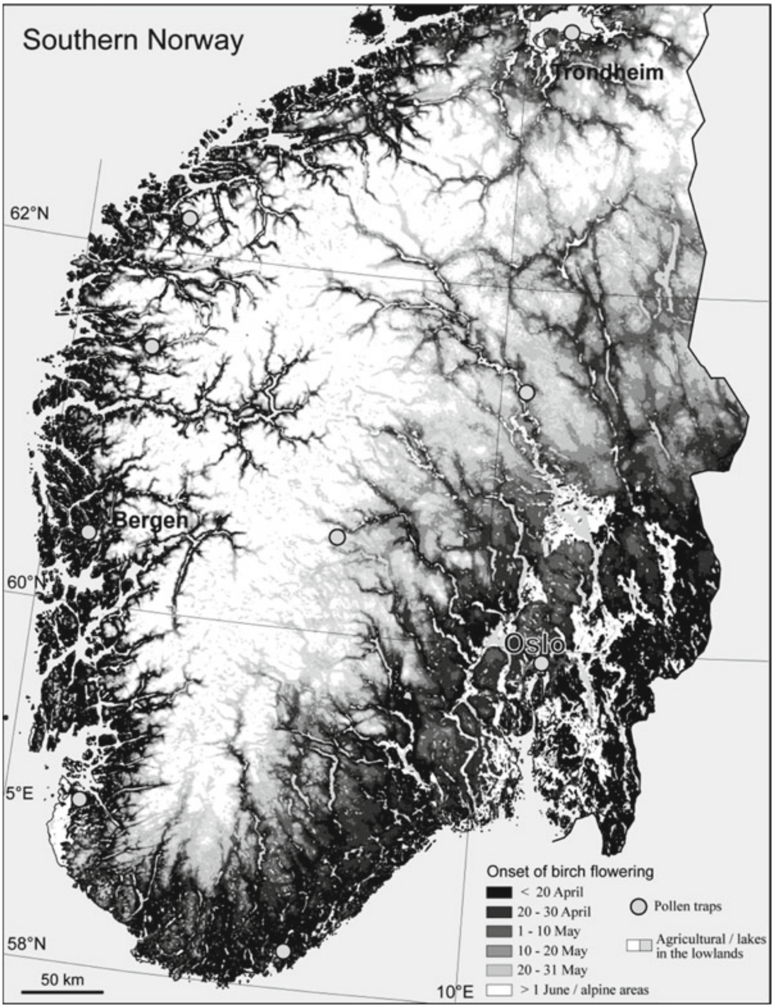

Fig. 4.2 Onset of birch flowering in southern Norway, based on mean values from the MODIS-NDVI dataset for the period 2000–2007, after Karlsen et al. (2009a). The map also shows the position of the pollen traps used in the study

real-time monitoring of the birch flowering. In the model they used additional geo-data, where the most important was a Landsat TM based vegetation map of all of Norway used to identify vegetation types where birch occurs. The model was applied to monitor the onset of birch pollen season in near real-time during spring 2009 (Karlsen et al. 2009b). The method showed in most cases good agreement with data from the pollen traps. However, the model does not give any information about the amount of birch pollen or about long-distance transported pollen. This will be a limitation for forecasting in areas where long-distance transport is an important factor.

Future Prospects

The next step to consider would be to apply the model, which was developed for the start of birch pollens season in Norway, at other places. In areas with complex topography, as the Alps, where atmosphere-based models are less reliable, remote sensing-based methods would be particularly useful. The same procedure would have to be followed as was applied in Norway, where at first the average onset of birch flowering had to be mapped and afterwards a model for real-time measuring was installed. It is also believed to be possible to further develop the model to map the onset of the grass pollen seasons as well, but with slightly less accuracy. Since the release of e.g. grass pollen occurs after the peak of the seasonal NDVI curve, simple threshold based methods as used for birch pollen will not be sufficient. Comparing time integrated NDVI and Growing Degree Days Karlsen et al. (2006) found a very high correlation. Accordingly, one method to apply satellite monitoring for estimating release dates of different pollen types could be to use time integrated NDVI as an additional data source.

The new generation of satellite sensors, with both daily data and high resolution (as Formosat-2, Komsat-2, and RapidEye), provide an opportunity to monitor the onset of the pollen season at a local scale. Due to the high data costs, it is only realistic to use these data for relatively small areas (<~1,000 km^2) and only some years. However, this new scale of observation creates a link between field observation of phenology/data from pollen traps and medium resolution sensors as MODIS. This would increase our understanding of the seasonal dynamics of the vegetation and improve the up-scaling from pollen traps to large regions, for instance, by the use of MODIS-NDVI data.

In the years to come there will be an increasing number of satellites with a range of scales in spatial resolution suitable to be used for near real time phenological monitoring (Ward 2008). Our conclusion is that satellite based monitoring of phenology is rapidly developing and observations will be assimilated into phenological models.

4.2.2 Aerobiological Observations

There is a wide spectrum of micro-organisms and biological particulate matter present in the atmosphere, which is investigated with a similarly wide range of methods and instruments (Cox and Wathes 1995). Biological aerosol sources are located in any place where biological activity exists. Many small organisms use the atmosphere as a means to be transported or to transport their own propagules. Bacteria, microalgae, microfungi, protozoa, insects and sometimes viruses are among the organisms that change their geographic location along their life cycle through the air. Fungal spores, lower plant spores or higher plant pollen grains are propagules transported by air. Fragments of fungi, animal or vegetal origin are also present in the atmosphere.

Aerobiology studies the release of biological particulate matter into the atmosphere, its transport through the atmosphere, and its deposition and re-suspension. In order to be taken up and transported, biological particles have to be released into the atmosphere, which is achieved by different mechanisms: (a) active, explosive or turgid; (b) passive, from an external agent. For example, in the case of most lower plants, i.e. bryophytes and pteridophytes, one finds active mechanisms for spore emission. In some cases, special sporangia and other devices enable an active emission by means of a catapult like discharge of the spores. However, these spores are usually large and heavy, which limits their dispersion through the atmosphere. In contrast, most of higher plants possess passive mechanisms for pollen emission. The role of pollen grains is to transfer the male gametophytes to the female reproductive organs. This is called pollination and is achieved via different mechanisms depending on the plant, which may be classified as anemophilous, entomophilous or hygrophilous. In order to guarantee an effective pollination, the entire structure of pollen grain is subject to selection pressure. However, this review will concentrate mainly on airborne pollen from wind-pollinated plants that are primarily responsible for seasonal allergies, the prevalence of which has been increasing substantially during the last decades.

4.2.2.1 Pollen Monitoring History

Aerobiology is a young scientific discipline that made great advances in the second half of the twentieth century, largely due to the introduction of advanced methods of monitoring. This brought a larger number of devotees to the subject and witnessed the rise of networks monitoring pollen and fungal spores on a national scale. Though aerobiology is related par excellence to ecology, it grew up following the major advancements in "allergology". The term "Aerobiology" was defined in the 1930s by Fred Campbell Meier (1893–1938), but Aerobiology did not become a recognised discipline until the 11 September 1974, when the International Association for Aerobiology (IAA) was founded at the 1st International Congress of Ecology, which was held at The Hague, in The Netherlands. Prior to this, in 1964, aerobiology had become a theme when the International Biological Program (IBP) was established. The major objective of the IBP was to study the biological basis for productivity of the world's ecosystems. NASA supported the Atmospheric Biology Conference, with the idea that the atmospheric dispersion of biological materials might be given attention by the IBP. An aerobiological programme was subsequently established in 1968, through the efforts of Benninghoff & Gregory, under the IBP section Use and Management of Biological Resources (UM). The IBP officially finished in 1974, when it was recognised that the studies on Aerobiology at international level should continue, and the IAA was formed 4 years later. Aerobiology is currently considered an experimental and multidisciplinary science that includes workers from botany, palynology, mycology, agronomy, microbiology, acarology, bioclimatology, meteorology, allergology and ecology. Aerobiology is made up of many different scientific disciplines and so it is not easy to trace the most significant milestones in its history.

The Origin of the Aerobiology

In ancient times, we find several references to the idea of micro-organisms and to the hypothesis that air can be a vector for diseases. For example, the Greek physician Hippocrates (460–377 BC) argued in “De Flatibus Corpus Hippocraticum” that people fall ill with fever after having inhaled infected air, although he was unaware of the nature of the infection. The author M. Terentius Varro (116–27 BC) cited invisible animals, which penetrated the body through mouth and nostrils thus causing disease. This concept was taken up in 1546 by Girolamo Fracastoro (1478–1553), who realised that some diseases were caused by “life seeds” that contaminate man, assuming that the body is reached by these particles by direct contact or by breathing in infected air. The Latin poet and philosopher Titus Lucretius Caro (98–55 BC) also mentioned small particles that can infect man.

Precursors of Aerobiology

Another important step was accomplished at the end of the fifteenth century by the invention of the microscope, the instrument that gave green light to explore the unknown world of the infinitely small and that allowed the investigation of aerobiological particles. The natural philosopher Robert Hooke (1635–1703) made a number of accurate microscopic observations in the book Micrographia (1665). This work inspired the Dutch biologist Antonie van Leeuwenhoek (1632–1723), who between 1673 and 1683 first described bacteria, the animalcules (protozoa) or diatomaceous, as well as some yeasts and moulds, and assumed that they were transported by the wind along with floating dust. In 1682 Nehemiah Grew published his book “Anatomy of Plants”, which contains the first known description of pollen (Fig. 4.3).

Birth of the Experimental Aerobiology

Around 1860, French biologist Louis Pasteur (1822–1895) began to study the bioaerosols in the atmosphere. He built a series of specially designed glass bottles with a long curved neck (“a swan neck”) with a spout at their end that could be sealed. The bottle was positioned in a way that the dust containing spores and germs were deposited on infusions that were sterilised by boiling in the bottle. When the air had been filtered or heated at a temperature high enough to kill all germs, the infusion remained sterile, but exposure to dust instead of air caused the deposition and growth of microorganisms on the infusion. Thanks to these experiments, Pasteur was able to demonstrate the heterogeneity of aerospora and the dispersion of germs in the atmosphere. Therefore, Pasteur is also considered a pioneer in aerobiology, the one who designed the first aerobiological experiments to examine the biological contents of dust in the air of Paris.

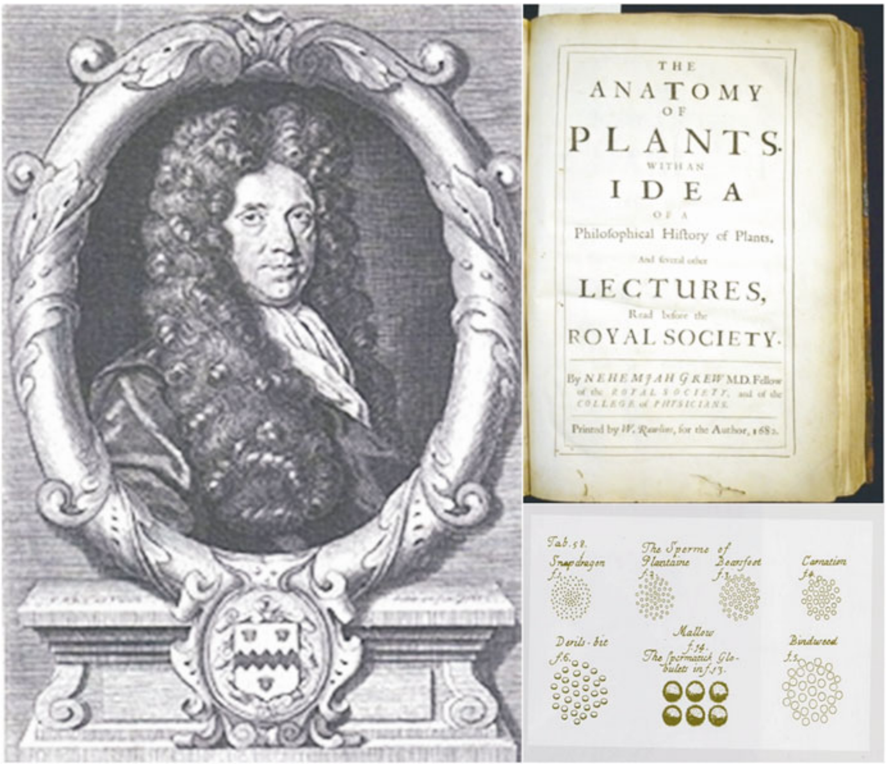

Fig. 4.3 Nehemiah Grew (*left*), who published the "Anatomy of plants" in 1682 (*top right*). His book contains a plate depicting the first description of pollen (*bottom right*)

His most direct descendant was without doubt the French physician Pierre Miquel (1850–1922), who continually monitored pollen and fungal spores in the atmosphere for years with various types of samplers of his own design. The results of his research were published in "Les organismes vivants de I'atmosphere" (1883), which presents interesting data and many graphs on the amount of fungal spores and bacteria in the air correlated with some meteorological parameters such as temperature, rainfall, humidity and wind speed.

During the nineteenth century, the German physician and naturalist Christian Gottfried Ehrenberg (1795–1876) worked as one of the founders of the science dealing with micro-organisms transported by the atmosphere. He examined samples of soil, water, sediments, atmospheric dust and rocks, describing hundreds of new species of bacteria, protozoa, diatoms, rotifers and fossils in about 400 publications. Irish physicist John Tyndall (1820–1893) became famous for his studies on light (the Tyndall effect) and sterilisation (Tindalizzazione). He also conducted aerobiological studies investigating the physical aspects of atmospheric particles and physiological growth of micro-organisms. Indeed, in essays on the floating matter of the air in relation to putrefaction and infection (1881), which represents his most important contribution to aerobiology, Tyndall studied the organic nature of dust in the atmosphere and claimed that epidemic diseases and putrefaction were caused by germs, thus refuting the abiogenesis. He was among the first to observe that seeds are abundantly present in and transported by the atmosphere. In the same century, Florentine Giorgio Roster (1843–1927), professor of biological chemistry and hygiene at the Royal Institute for Advanced Studies in Florence, can certainly be called one of the first experts on urban air pollution. He was the first true Italian aerobiologist.

Birth of “Aeropalynology” from “Aerobiology”

During the second half of the nineteenth century the study of micro-organisms expanded greatly. This stimulated an interest in studying also the pollen grains in the air, which led to the development of the field of aeropalynology. During this time, a number of hypotheses emerged, which tried to explain the seasonal appearance of respiratory allergy in humans. It should be recalled that in 1819 the English physician John Bostock (1773–1846) set forth in detail the clinical picture of the disease.

Origins of Pollen Monitoring

In 1873, another English physician, Charles H. Blackley (1820–1900), went down in history as the father of aerobiology. He was the first to show that pollen was present in large quantities in summer and was the cause of breathing problems (described as Catarrhus æstivus or summer catarrh), demonstrating the direct relationship between the amount of pollen in the air and the severity of symptoms. From 1866, he compiled pollen calendars of Manchester, UK, having counted the pollen that he collected with a self-made sampler under a microscope.

During the same period, Morrill Wyman (1812–1903) described the autumnal catarrh in the United States of America, which appeared each year in August and September. He attributed it to the flourishing of ragweed (*Ambrosia)*. Elias Marsh (1835–1908), who created the first pollen calendars for ragweed pollen in 1875 for Paterson (New Jersey), must also be mentioned.

The factor that contributed most to the increase of knowledge about aerospora, after the invention of the microscope, was the introduction of air sampling devices. Starting from the first sampler of Pasteur (1860) and Blackley (circa 1866), during the second half of the nineteenth century, many scientists devised their own equipment to conduct aerobiological investigations. Most famous among them were the aeroscopes of Maddox (1870), Cunningham (1873) and Miquel (1878), the aeroscope recorders for bacteria and moulds (France, circa 1884) and the electric suction pump of Roster (1885). These instruments relied on the state of the art technology of their time, but more rudimentary samplers were in operation between the late 1800s and the early decades of the twentieth century. For instance, samplers were built with clothes pegs and a glass slide covered with glycerine fixed to a backing of wood and often covered with a small roof to shield them from the elements. These low precision instruments enjoyed some success primarily for economic reasons and they aroused the interest of many scholars for aerobiology and pollen monitoring. In 1946, O.C. Durham introduced his gravimetrically standardised sampler, which became the most frequently used instrument around the world for many years. It was also adopted by the "Pollen and Mold Committee of the American Academy of Allergy" as standard equipment. In 1967, the American Botanists Ogden and Raynor devised the rotary impact sampler "Rotoslide". In 1952, the Englishman Jim Hirst designed a sampler with a suction pump, which was the ancestor of the modern Burkard and Lanzoni traps. Hirst-type volumetric samplers are still operated by the majority of monitoring centres throughout the world.

Aerobiology in 1800–1900

Without doubt the discovery of numerous micro-organisms responsible for infectious diseases have to be listed among the most significant achievements of biomedical science between the second half of the 1800s and the first decades of the 1900s. German bacteriologist and hygienist Carl Flügge (1847–1923), a colleague of Robert Koch who devised many bacteriological techniques and established the bacterial causes of a number of infectious diseases, proved around 1890 that coughing and sneezing releases small droplets, defined as "droplets of Fluggi", which contain numerous pathogenic micro-organisms that remain suspended in the air and are responsible for the transmission of infectious diseases to new hosts. Around 1912, the Czech-Austrian botanist and chemist Hans Molisch (1856–1937) coined the term "Aeroplankton" by including it in all sorts of particles and especially those of biological origin such as pollen, fungal spores, algae, etc.

Birth of Pollen Monitoring Networks

Pollen monitoring at national level started for the first time in the US by O.C. Durham (1889–1967) in an attempt to correlate patient and pollen data of *Ambrosia* in 1928. Within a few years, the American network had expanded to more than 50 stations scattered throughout the country and the measurements were extended to all types of pollen. The network spread in a short time over much of the continent including Canada, Mexico and Cuba. Durham continued to coordinate this pollen recording network until the 1950s. In 1955, Durham supervised the publication of the first report of the monitoring network, which constituted the first of its kind. The first conference to deal exclusively with aerobiological topics was held in 1942 under the auspices of the "American Association for the Advancement of Science". After 1970, national monitoring networks were established in most European countries and the "European Aerobiology Society" was founded in 2008.

Aerobiology is still developing as a discipline and considerable advances are expected in the coming years. For example, many aerobiologists still use sampling equipment that is based on a design from the 1950s (Hirst 1952) and the analysis of samples is by light microscopy, which is labour intensive and extremely time consuming. One of the main areas of future development is expected to be in the monitoring of airborne organic particles with automated detection and analysis technologies (Rogers 2006), such as monitoring the allergen content of the air, DNA analysis or image analysis. There are also likely to be improvements in atmospheric modelling. Many of these advances are expected to be fuelled by an increasing need for aerobiological data due to the effect of climate change on human health (Huynen et al. 2003).

4.2.2.2 Monitoring Instruments and Sampling Methods in Aerobiology

Conventional Pollen Monitoring Instruments

Mullins and Emberlin (1997) reviewed strategies applied in sampling airborne pollen. The authors stressed that airborne pollen samplers, if they are to be effective, should be able to obtain volumetric record of all particles (5–50 μm), irrespective of the wind velocity. Numerous strategies including cylinder traps, sedimentation traps, impactors and filters have been used in trapping pollen.

Volumetric pollen samplers based on the Hirst design (Hirst 1952) are used as the standard method in many national networks for measuring the pollen concentration in the atmosphere. Air is sucked in through a 2 mm × 14 mm nozzle at a rate of 10 l/min. The rate of 10 l/min was chosen for operation in the field because efficiency varied less at different wind speeds than at an alternative rate of 17.5 l/min. At a suction rate of 10 l/min there was also less danger of obscuring the spore deposit with fine dust particles, which are less efficiently impacted with a lower velocity in the orifice (Hirst 1952). Airborne particles are deposited on a tape mounted on a drum, which is slowly turned by clockwork (Levetin et al. 2000). The sticky tape is then mostly stained with suitable dye and analysed under an optical light microscope, whereby the pollen and spores of different taxa are determined and their numbers per surface area counted according to standardised procedures (Makinen 1981; BAF 1995; Galán et al. 2007). The Hirst type volumetric pollen trap supplies pollen concentration data at a temporal resolution of up to 2 h. Relating the pollen counts with the exposure time, the number of pollen grains per cubic meter and time can be calculated. In order to avoid the distortion of the pollen count by local emissions, the traps are located on the roof of buildings, often at 12 m above street level (Winkler et al. 2001).

In the US, whirling arm samplers, such as the Rotorod, are preferred. In whirling arm samplers, airborne particles impact on one side of translucent 6 cm long square rods (1.6 × 1.6 mm) that whirl through the air at 2,400 rpm resulting at a sampling rate of about 120 l/min. The duration of whirling period determines the sampling period.

The Cour's method (Cour 1974) is based on the principle of passive sampling, which has the advantage that it does not require any power supply. The traps are mounted in 3 m above the surface. The pollen is collected passively on a filter. It is a volumetric method, because the results are expressed as the number of pollen grains per m^3 of air. The method has been applied for agronomical questions and has exclusively been used by the French Association for Ragweed Study (AFEDA) for the last 29 years. A pair of 20×20 cm filters (400 cm^2) composed of six sterile cellulose gauzes impregnated with silicone oil are exposed over a week. The trap functions with a weather vane; it is always oriented perpendicular to the wind direction and retains pollen grains transported by air currents. Wind speed, measured by an anemometer at the level of the trap, is used to compute the volumetric quantity filtered and the number of pollen grains per cubic meter of air. In the laboratory, the gauze with the collected material is dissolved in hydrochloric acid, hydrofluoric acid, acetone and potassium hydroxide. Chemical treatment empties pollen grains of the nucleus and the cytoplasm. The pollen grains are concentrated in the residue which is diluted in glycerine and homogenised. A volume of 50 μl of this dilution is deposited between a slide and a 22×50 mm cover slip and examined using light microscopy.

Pollen Counting Methods for Conventional Instruments

Whatever method for airborne pollen sampling is used, further analysis requires identification and quantification of registered pollen types. The identification of pollen requires knowledge on basic palynology (primarily pollen morphology), and it is performed either based on a comparison with reference microscopic slides or by using pollen identification keys and atlases. Due to their small size, pollen grains are commonly analysed under light microscope. The magnification is chosen so that pollen can be safely identified according to the characteristics specific for each taxon. The most widely used magnification in aerobiological monitoring is ×400.

Because the Hirst-type pollen samplers are the most common ones today, the various quantification methods will be briefly described below. When performing the quantitative analysis of a sample collected by the Hirst-type volumetric sampling procedure, the most accurate method would be to count pollen on the entire surface of the 24 h sample. However, from a routine pollen monitoring point of view and in the context of producing data for forecasting and informing public on the prevalent allergy risk, this would be unacceptably time-consuming. Therefore, three sub-sampling methods, which analyse only a fraction of 24-h slide, are proposed:

a. The random field method (Makinen 1981) considers the examination of a certain number of fields chosen at random from the entire daily surface, and counting the pollen present in each single field. This is probably the quickest method for slide analysis but, although it is good at estimating the daily mean concentration, it is unable to estimate short term concentrations (bi-hourly) (Kapyla and Penttinen 1981). Furthermore, the application of the random field method can result in underestimates or overestimates of the pollen concentration, because their depositing is not uniform on the tape, but depends on the particular biological cycle, environmental conditions and the type of pollen (Tormo et al. 1996).

b. The transverse traverses method (Emberlin et al. 1994) considers either the examination of successive tangent fields in 12 transversal lines or the examination of complete 12 transversal lines, separated by 4 mm distance from one another. In this way a line is read for every 2 h, enabling an estimation of both daily and bi-hourly pollen concentrations. The choice of the position of the lines could influence the final result obtained by this sub-sampling method, because pollen deposited within a very short time on the tape might be missed.
c. The longitudinal traverses method considers either the examination of successive tangent fields (Mandrioli 1990) positioned on 3 or 4 or 5 horizontal lines or the examination of 3 or 4 or 5 complete horizontal (Dominguez et al. 1991) lines separated by a space of about 2 mm. Although this method enables the estimation of both daily and bi-hourly pollen concentrations, it was noted that overestimates can arise from counting only the central regions of the slide, where most of the pollen is deposited (Tormo et al. 1996).

All of these sampling methods produce the pollen count expressed as concentration in pollen grains/m^3, which is calculated having in mind the suction rate of the used sampler and the ratio between the total sample surface and the sub-sampled surface of the slide based on the formula: pollen grains/m^3 = (pollen count*total sample surface)/(sub-sample surface on which pollen are counted*total volume of air sampled) (BAF 1995).

Since the main disadvantage of sub-sampling in the airborne pollen monitoring is the analysis of only a small proportion of the daily sample, Comtois and his colleagues (1999) checked the effect of sub-sampling on the accuracy of the quantitative analysis. They found that, when comparing concentrations obtained by counting the total slide surface versus counting only a fraction of it, none of the sub-sampling methods was able to reproduce the counting result of the total slide nor did the fractional counting give exactly the same result. Furthermore, the sub-sampling error was much higher than what is commonly believed, and it was significantly correlated with the abundance of pollen taxa on the sampled slide. Although each method has its advantages and disadvantages, all proposed methods enable a fairly good estimation of the whole biological population contained in a certain volume of air (Comtois et al. 1999; Sterling et al. 1999; Carinanos et al. 2000).

Automated Pollen Counting Techniques

For forecasting purposes, a continuous delivery of pollen counts and most suitably in an hourly time resolution would be very valuable. This cannot be achieved by manual counting systems, but could be obtained with automated pollen counting systems. In recent literature several different methods for automated pollen detection have been described:

1. Systems that make use of multifocal optical microscopic images of air samples collected by a conventional Hirst-type pollen sampler. A first step in automated counting of the pollen is the discrimination of the pollen grains from other airborne material in the images (Landsmeer et al. 2009; Bonton et al. 2001).

For subsequent identification of the pollen grain, several characteristic pollen features, including shape, statistical grey-level and specific pore/colpus features are extracted from the images by pattern recognition software tools (Boucher et al. 2002; Chen et al. 2006). These methods report various levels of success in identification of specific pollen types: 77% in samples from airborne pollen (Boucher et al. 2002) or 97.2% in samples containing three allergenic pollen taxa (Chen et al. 2006).

2. A fully integrated pollen sampling system that automatically collects, prepares and records by making use of a conventional light microscope (Ronneberger 2007). The method developed for the recognition of the pollen employs digitised images, using the grey-value of each pixel (Ronneberger et al. 2002). This system reached a recognition rate in "real world" samples of 84.3% (Ronneberger 2007). Up until now, it was not developed beyond a stage of a prototype and it did not reach the stage of becoming commercially available.
3. Other systems do not make use of digitised images of pollen, but are based on the technology of particle counters by laser light.
   - In the system described by Kawashima et al. (2007), pollen is characterised by the sideways and forward scattering of laser light. Air, containing the airborne particles, is passed through the optical system and irradiated by a laser beam. The scattering of light signals caused by the pollen grains is recorded in real time and processed by a computer. During a sampling period in late summer, pollen from nettle (*Urticaceae)*, ragweed and grass (*Poaceae)* could be separated well by different scattering patterns. For other European pollen taxa, the system has not been tested yet.
   - In Japan, another real-time airborne pollen counter was developed by the company Kowa. The technology is based on a laser particle counter and on the characteristic distribution of pollen on the scattered diagram according to the grain size versus the fluorescent hue. In Japan, this counter is used by the Tokyo pollen information network systems (Suzuki et al. 2008).
   - Recently, a new methodology was presented on the 9th International Congress of aerobiology: the WIBS 4 (Wide-Issue Bioaerosol Spectrometer). This instrument combined information from laser light scattering with 2D-spectroscopic measurements. The instrument was successfully used in an area with a low diversity of pollen (Sodeau et al. 2010).
4. Another method is based on the Coulter counting principle (Zhang et al. 2005). Pollen was suspended in a KCl aqueous suspension and passed through a microchannel. The changes in conductance, due to the passing of the pollen, were recorded and analysed. In this system juniper (*Juniperus)* and grass pollen could be discriminated.

Airborne Allergen Monitoring Instruments

Allergologists have become increasingly interested in questions concerning the allergenic potency of pollen, which cannot be answered by the conventional pollen counts. For the purpose of allergen measurement in pollen new types of pollen traps needed to be developed, which now allow the application of immunological analysis methods like ELISA or immuno-fluorescence. Spieksma et al. (1990, 1999) used a high volume sampler that operated at 1,130 l/min equipped with a five-stage cascade impactor (HSV), sampling onto glass fibre impaction sheets to fractionate particles by size class. The HVS had a high capture efficiency (96–99 % for particles > 0.3 μm diameter) but sampling was not isokinetic or unidirectional. Also, Rantio-Lehtimaki et al. (1994) used a static, size selective bio-aerosol sampler (virtual impactor) with a flow rate of 18.5 l/min that collected samples onto three filters. Alternatively, Emberlin and Baboonian (1995) collected particles over a wide range of sizes in Eppendorf tubes for immunological analysis using a cyclone sampler operating at 16.6 l/min (Mullins and Emberlin 1997).

More recent approaches include the Coriolis® Delta of Bertin that works at a flow rate of 300 l/min and transfers pollen into a liquid collection media. It has an efficiency of 90–100% for particles with a diameter of 3 μm upwards (Bertin Technologies 2007).

Other samplers have been designed that are able to separate particulate matter from ambient air according to its size. The Andersen sampler aspirates 28 l/min of air (Andersen 1958). This sampler may contain up to six impaction stages, capturing particles with an aerodynamic size of PM > 8.2 μm, 10.4 μm > PM > 5.0 μm, 6.0 μm > PM > 3.0 μm, 3.5 μm > PM > 2.0 μm, 2.0 μm > PM > 1.0 μm and < 1 μm, enabling the study of the distribution of the allergenic particles according to their sizes (De Linares et al. 2007, 2010). The Chemvol® high-volume cascade impactor represents a more recent development (Demokritou et al. 2002). Ambient air is aspirated at 800 l/min and split into three identical airstreams, each impacting on a porous polyurethane impacting substrate. A cascade of stages with cut-offs at 10, 2.5, 1, 0.12 μm and an absolute stage can be mounted. Pollen and allergen are mostly detected in the stage PM > 10 μm with an additional 10–15% of allergen in the stage10 μm > PM > 2.5 μm. The smallest stage (particles with a diameter of 2.5 μm > PM > 0.12 μm) is seldom used when working with pollen or allergen in ambient air as no allergen was found in this stage. The authors postulate that this could be due to concomitant collection of diesel particles at this stage, absorbing the possibly available allergen (Buters et al. 2010). This phenomenon could be shared by all samplers.

Personal Samplers

The Burkard Personal Volumetric Air Sampler (Burkard Manufacturing Co., Rickmansworth, UK) is a portable battery-powered device similar in operation to the Hirst trap. Air is drawn through a vertically orientated slot-shaped intake and impacted directly onto an adhesive covered microscope slide. The sample may thus be examined under the light microscope with little additional effort (Aizenberg et al. 2000). Whilst the term "personal sampler" is often applied to such portable devices, true personal samplers that sample from the breathing zone are designed to be worn by one person. For instance, the CIP 10, available from Arelco, was developed by

INERIS (Industrial Environment and Risks National Institute) to measure the exposure of workers to dusts in coal mines (Arelco 2004). The sampler can be set up to select either the respirable alveolar, thoracic or inhalable particle size fraction, collected in a foam filter. The Button Aerosol Sampler (SKC, Eighty Four, PA, USA) is a filter device that targets the inhalable particle fraction. Air is aspirated at a rate of 4 l/min through a curved, porous inlet designed to minimise wind sensitivity and promote uniform particle distribution. Sample analysis is by microscopy, immunoassay or PCR (SKC 2010). The Nasal Air Sampler is a passive impaction device worn inside the nasal cavity, thus truly measuring personal exposure rather than particle concentration in the breathing zone. Inhaled air is drawn past a specially designed adhesive strip onto which particles with sufficient inertia are impacted. Samples may be analysed using ELISA, or be mounted for microscopic examination (Graham et al. 2000).

Quality Standard

In order to produce comparable aerobiological data a Quality Control working group has been established within the European Aerobiology Society (EAS) in 2008, which intends to create an internationally recognized standard. As a first step towards such a quality standard a preliminary list of "Minimum requirements" for all monitoring stations involved in the European Aeroallergen Network (EAN) has been compiled. More details of the Quality Control working group discussions have been published in the IAA Newsletters (beginning with number 67: http://www.isac.cnr.it/aerobio/iaa/IAABULL.html).

4.2.2.3 Applications of Aerobiological Monitoring

Mandrioli and Ariatti (2001) stated that aerobiology must be considered as a discipline by itself as well as a tool for other disciplines. Because pollen grains of a number of plant species induce allergic reactions, aerobiology and in particular airborne pollen research developed as a discipline in close relation to medical research. In addition to that, allergenic airborne pollen is the only object of aerobiological research for which routine monitoring on a daily basis is widely accepted and often implemented in country, regional or continental networks, producing long time series of data available from numerous regions worldwide. Bryant (1989) identified pollen as "fingerprints of plants", which are closely related with flowering, reproduction and distribution of the vegetation. Such relationships provide the potential for airborne pollen observations to be used in wide spectra of studies dealing with anemophilous plant species.

Frenguelli (1998) reviewed the potential contribution of aerobiological observations to agriculture. There it can for instance be applied to yield forecasting. Two approaches have been followed: (1) the pollen index indicates the number of developed flowers, which correlates with the number of fruits in monoecious plants; (2) the pollen index indicates the amount of pollen available for fertilisation in anemophilous plants (a higher number of successful fertilisations is linked with a higher fruit productivity). These approaches were successfully implemented in forecasting the production of olives (*Olea Europeae*, Moriondo et al. 2001; Galán et al. 2004, 2008; García-Mozo et al. 2007a), grapes (*Vitis vinifera*, Cristofolini and Gottardini 2000) and even forest species, such us oaks (*Quercus*, García-Mozo et al. 2007b) or birch (Litschauer 2003).

On the other hand, with the development of Genetically Modified (GM) crops, there emerged the need for monitoring the potential gene flow (Stokstad 2002). In that context, aerobiological observations could offer data on pollen dispersal patterns primarily originating from wind pollinated plants, such as oak (Schueler et al. 2005), which show an intermediate to high potential of gene flow (Govindaraju 1988). In the case of oilseed rape (*Brassica*), although it is primarily insect-pollinated, aerobiological studies prove transport of airborne pollen (Fiorina et al. 2003). These findings address the potential risk of gene flow in GM oilseed rape threatening the surrounding crops. Therefore, aerobiology should support the development of methods to predict the concentration of viable pollen as a function of distance from the source. Such predictions should be a guideline for policy makers when defining distances needed between crop fields in order to prevent gene flow.

Aerobiological observations enable indirect analysis of plant responses (in particular linked to male reproductive systems) to environmental factors. For example, stress situations caused by frost could lead to male sterility, which would result in a lower pollen index, as it was observed in the case of the Mediterranean cork-oak (*Quercus suber*, García-Mozo et al. 2001). Also, climate change induced differences in the timing and duration of the flowering phenophase of anemophilous plants that could be observed by analysis of the duration, start and end dates of the airborne pollen season (Tedeschini et al. 2006; Frei and Gassner 2008). Such changes were also observed by Fotiou et al. (2011) who modelled the flowering process of north- and south-facing populations of spreading pellitory (*Parietaria judaica)* and compared the duration of the flowering and pollen seasons locally.

Pollen analysis has a great potential for providing a continuous record of pollen production going back thousands of years, due to the fact that pollen is produced in large quantities, dispersed widely and remains well preserved in wet anaerobic environments. In order to be able to reconstruct past climate, Autio and Hicks (2004) suggested deducing an empirical relation between pollen production and meteorological conditions. Applying sedimentational Tauber traps, these authors analysed the annual variation in pollen production in the studied area in relation to meteorological parameters. The network of stations for pollen monitoring using Hirst type volumetric traps is well distributed all over Europe. Furthermore significant correlations exist between data obtained by volumetric Burkard and sedimentation Tauber traps (Levetin et al. 2000). This allows the description of the influence of climate variability on pollen production and deposition, which supports the quantitative reconstruction of past climate.

4.2.2.4 The European Pollen Information System

At present, the Europe-wide pollen information system consists of two coherent units:

- the European aeroallergen database EAN
- the public web portal www.polleninfo.org

EAN Database

Both units form the basis of the aerobiological unit. Access to the database is restricted to a defined user group: those who contribute data have read access to all available data for internal use. By agreement, the use of data for publication or for commercial purposes without the consent of the data owners is prohibited. Founded in 1988, the database has got a new structure in 1999 and 2009.

The majority of data sets cover the last decade, but some time series extend back as far as to 1974. In total, over 700 monitoring stations from 38 countries are incorporated in the database, collecting pollen and spore counts of over 200 different taxa. This results in about 1 million annual reports (pollen types per station and year). The main goal concerning pollen information services is to assist in forecasting and to help in developing and testing forecast models. Another frequently used feature is providing data for multicentre studies in allergology, forestry, and climatology.

The Public Web Portal (www.polleninfo.org)

Since 1997, a European platform for pollen information has been provided for the public (www.cat.at/pollen/). A hierarchic structure allows navigation from general overviews down to highly specific local information contents. The up-to date information county by country are available both in English and in the country language(s). The main goal is to provide links to pollen information services on a common European pollen information portal – in particular for travellers and for vacation planning. A new platform www.polleninfo.org has been launched in mid April 2003 with financial support from epi Ltd. replacing the old cat.at/pollen/site.

4.3 Modelling and Forecasting of the Pollen Season

Although very different in the way of being observed and measured, phenological events and pollen counts can be traced back to the same phenomenon, the flowering of plants. Similarly, both kinds of data can in many respects be modelled with a similar set of observation-based models. Simple regression models can predict entry dates of phenological phases and likewise the start, peak and end of the pollen season or, given a greater number of independent variables, the day to day variability of the pollen counts. Phenological models will equally well predict the entry dates of phenological phases as well as the start, peak and end of the pollen season.

Phenological models are sometimes grouped into the class of process-based models, because they are built on assumptions rooted in experimental results on plant physiological responses to various environmental variables (Chuine et al. 2000). The other modelling approaches presented in this section are summarised under the term observation-based models, because no a-priori assumptions are involved. This includes regression models, time-series modelling and a survey about applications of artificial intelligence methods to pollen data. A discussion of special problems related to numerical pollen forecast completes the literature survey.

4.3.1 Observation-Based Models

Observation-orientated models relate pollen records (dependent variable) to one or more variables (independent variables) that can be measured or predicted, and are constructed without knowledge of the sources, emission or calculations of diffusion (Norris-Hill 1995). On the contrary, everything starts from the pollen counts being recorded at the pollen traps (the receptors). Pollen data usually produce mean daily values for the studied area and in some cases hourly or 2-h values. These data can be used for producing forecasts of day-to-day variations in pollen concentrations, or for predicting characteristics of the pollen season, such as start dates and severity.

All methods use certain mathematical tools in order to describe and imitate the behaviour of pollen count (its temporal and in some cases spatial variations); they may be applied for better understanding, description and knowledge concerning pollen season problems. The most rudimentary method for pollen forecasting is the pollen calendar. Recording seasonal variations in the timing and abundance of different pollen types is the first task undertaken by operators of pollen-monitoring sites. When sufficient data has been recorded, it is possible to relate temporal variations in pollen records (diurnal variations, daily average values or seasonal characteristics) to meteorological data, such as temperature and rainfall. This is achieved using a variety of statistical techniques that include correlation analysis, parametric Pearson correlations and non-parametric Spearman Rank correlation analysis (e.g. Stach et al. 2008; Smith and Emberlin 2006; Galán et al. 2000; Rodriguez Rajo et al. 2005), Factor Analysis (Makra et al. 2004) and hierarchical multiple regression analysis (Emberlin et al. 2007). The results from these analyses can be used to improve qualitative predictions or provide the theoretical rationale on which quantitative forecast models are built.

4.3.1.1 Regression Models

Regression analysis is an empirical technique that is often used in aerobiological studies. It is used to predict a score on one variable from a score on the other and as a result is often referred to as a causal method of statistical modelling. Causal models predict the future by modelling the past relationships between a dependent variable and one or more other variables called either independent or predictor variables. The goal of regression analysis is to arrive at the set of B values for independent variables, called regression coefficients, that bring predicted Y values from the equation as close as possible to the Y values obtained by measurement. The ability to find potentially causal relationships that not only predict but also explain the dependent variable makes regression analysis a very powerful technique (DeLurgio 1998; Tabachnick and Fidell 2001).

In simple bivariate (two-variable) linear regression, a straight line between the two variables is found. The best fitting straight line goes through the means of X and Y and minimises the sum of the squared distances between the data points and the line (Kinnear and Gray 1999; Tabachnick and Fidell 2001). This technique is useful when the dataset is not large enough for multiple regression analysis (Stach et al. 2008) as studies based on relatively small datasets are inappropriate for the multiple regression process because they result in inflated regression coefficients of determination (R^2 values) and inaccurately estimated coefficients. In such cases, it may be more suitable to use simple linear regression (Ong et al. 1997).

In multiple regression analysis, the values of the dependent variable are estimated from those of two or more independent variables (Kinnear and Gray 1999). Multiple regression analysis makes a number of assumptions about the data (such as normality, linearity and homoscedasticity) and is not forgiving if they are violated (Kinnear and Gray 1999; Pallant 2001). Many of the problems associated with these factors can be addressed by transforming the data prior to analysis. Different methods of data transformation have been used by a variety of authors, mainly when predicting daily variations in pollen counts; these include square root (Smith and Emberlin 2005), lognormal (Alcazar et al. 2004) and log10 (Stach et al. 2008). Toro et al. (1998) investigated the use of different methods of data transformation. The authors transformed daily mean grass pollen data (x_t) expressed as the number of pollen grains/m^3 into different scales before attempting to construct regression models: $\log(x_t+1)$, $\sqrt{x_t}$, and $\ln((x_t{*}1{,}000,\ \ x)+1)$. The latter method is the transformation proposed by Moseholm et al. (1987). Toro et al. (1998) found that a regression equation obtained from data transformed by square root usually resulted in a better prediction because substantial errors can be introduced in de-transforming the data to the usual scale (pollen grains/m^3), although its R^2 value was lower than equations obtained with other transformations. Furthermore, it was suggested that the transformation proposed by Moseholm et al. (1987) should not be used to construct short-term predictive models because the margin of error is further increased by the inclusion of an additional factor $\sum x$ (total of pollen grains for the whole season), which has to be predicted before it can be transformed (Toro et al. 1998). In addition to data transformations, datasets also have to be cleaned prior to analysis in order to remove or alter univariate and multivariate outliers that regression models are sensitive to, as they can distort statistics (DeLurgio 1998; Tabachnick and Fidell 2001).

There are several methods of multiple regression analysis commonly used in observation based modelling. Step-wise multiple regression analysis has often been used in aerobiological studies (Galán et al. 1995; Goldberg et al. 1988; Bringfelt et al. 1982; Moseholm et al. 1987). An alternative method used in aerobiological research is standard multiple regression analysis (Stach et al. 2008; Smith and Emberlin 2005; Makra et al. 2004), where all the independent variables are entered into the equation simultaneously and each independent variable is evaluated in terms of its predictive power compared to that of the other independent variables (Kinnear and Gray 1999; Pallant 2001). However, non-linear statistics should be used, if the data do not show a normal distribution (Toro et al. 1998), such as polynomic regression (Antepara et al. 1995) or semi-parametric Poisson regression models, where the variance of the data is proportional to its mean (Stark et al. 1997; Erbas et al. 2007).

Regression analysis has been used for predicting daily values (Stach et al. 2008; Smith and Emberlin 2005), as well as the start (Emberlin et al. 1993a; Laaidi 2001a, b; Galán et al. 1998, 2001a; Frenguelli et al. 1989; Davies and Smith 1973), peak (Orlandi et al. 2006), duration (Laaidi et al. 2003) and severity (Emberlin et al. 1993b; Galán et al. 2001b) of pollen seasons and the beginning of flowering (Crepinsek et al. 2006). A number of different independent variables were used in these analyses. Variables that affect the timing of pollen release from allergenic plants are used to predict the start of pollen seasons and the beginning of flowering, such as monthly (Stach et al. 2008; Emberlin et al. 1993a; Galán et al. 1998, 2001a; Frenguelli et al. 1989; Davies and Smith 1973; Crepinsek et al. 2006) or 10-day periods (sometimes referred to as decades-of-days; Stach et al. 2008; Smith and Emberlin 2005; Spieksma and Nikkels 1998) of meteorological data, as well as winter averages of the North Atlantic Oscillation (Stach et al. 2008; Spieksma and Nikkels 1998). Similar variables are also used when attempting to predict the severity of seasons (Emberlin et al. 1999; Laaidi 2001b; Galán et al. 1998). Observed pollen season starting dates, which occur before the start dates of the pollen season to be modelled, can also be considered as only or additional independent regression parameter (Norris-Hill 1998).

A variety of different independent variables have also been used to predict daily average pollen counts, and include minimum (Toro et al. 1998), maximum (Iglesias et al. 2007; Rodriguez Rajo et al. 2004, 2005; Mendez et al. 2005; Toro et al. 1998) and mean (Toro et al. 1998; Rodriguez Rajo et al. 2004; Goldberg et al. 1988) daily temperatures, rainfall (Stark et al. 1997; Rodriguez Rajo et al. 2004; Toro et al. 1998), relative humidity (Stach et al. 2008; Smith and Emberlin 2005; Toro et al. 1998), sunshine hours (Stach et al. 2008; Toro et al. 1998), wind speed (Bringfelt et al. 1982) and also direction and persistence (Damialis et al. 2005), and the amount of pollen recorded in the previous days (Stach et al. 2008; Smith and Emberlin 2005; Iglesias et al. 2007; Sánchez-Mesa et al. 2002; Rodriguez Rajo et al. 2004, 2005; Mendez et al. 2005). The division of grass pollen seasons prior to analysis has become an accepted methodology when attempting to predict daily pollen counts, because the relationship between pollen counts and environmental data tends to change during the pre-peak and post-peak periods (Sánchez-Mesa et al. 2003; Galán et al. 1995; Toro et al. 1998). Examples include splitting the grass pollen season into two and defined the pre-peak period as dating from the beginning of the main pollen season to the peak day itself (Toro et al. 1998), or dividing the grass pollen seasons into three (pre-peak, peak and post-peak periods of pollen release) because pollen counts around the peak day behave as one population, whereas pollen counts from the pre-peak and post-peak periods can be treated separately (Smith and Emberlin 2005).

When selecting the independent variables to enter into regression analysis, it is important to avoid multicollinearity, which exists when the independent variables are highly correlated. For instance, highly correlated variables (such as maximum, minimum and mean daily air temperatures) should not be included in the same multiple regression. It is also important not to attempt to assess prediction accuracy with the same data used to construct the model (Stark et al. 1997).

4.3.1.2 Time Series Modelling

Modelling and forecasting of pollen counts based on regression equations is simple and straightforward and can be carried out with any statistical package providing multivariate regression procedures. However, it suffers from a number of disadvantages. The most important one is that usually time is kept fixed and different time periods are handled as different variables, which prevents any exploration of the role of timescales. In order to do this, it is usually preferable to use time-series approaches.

The classical time-series method for the analysis and forecasting of pollen levels is the Box-Jenkins approach (Box et al. 1994). This is based on the successive refinement of the model by fitting different deterministic and stochastic components of variability. First, the average value is found and then subtracted from the series. Then, a trend is fitted to these centred residuals and the values of this trend-line are subtracted to get the detrended values. Next, successive years are stacked on a daily, weekly or monthly basis and we find the average over all years for each day, week or month. For example, if we use a monthly basis, the model $S(m)$ for $m =$ June is the average of the detrended values for June averaged over all years. Finally, after this cyclic model has been subtracted from the data, the autocorrelation structure is fitted using ARIMA techniques (Box et al. 1994). Ideally, at the end of this stage, the residuals should be free of correlation. This is a widely-used approach, which can be regarded as standard, and features in several aerobiological studies (Moseholm et al. 1987; Stephen et al. 1990; Rodriguez-Rajo et al. 2006; Aznarte et al. 2007). The model may thus be described as:

$$X(t) = M + Tr(t) + S(m_t) + \varepsilon(t) \tag{4.2}$$

Here, $Tr(t)$ is the trend component at time t, a steadily rising or falling background that is sometimes observed in pollen records (Damialis et al. 2007), and M is the mean value of detrended series. The seasonal component is $S(m_t)$, where m_t is the month (or week or day) of the year. Finally $\varepsilon(t)$ is the residual noise, whose structure can be described by an ARIMA model. It is often desirable to transform data, for example using the logarithm or square-root, before carrying out a Box-Jenkins analysis.

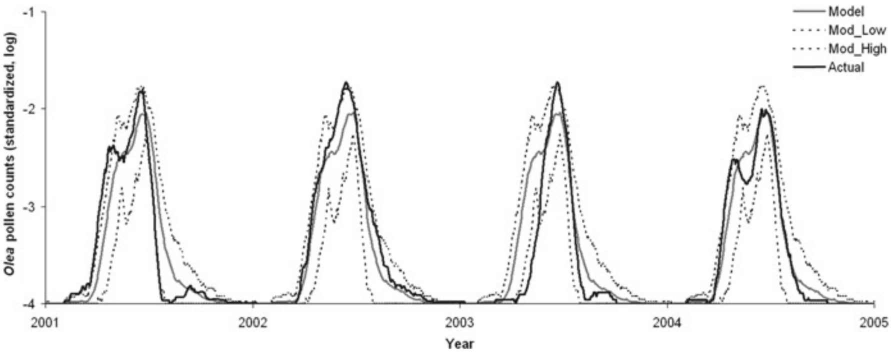

Fig. 4.4 Application of the Box-Jenkins method to forecast olive pollen levels in Thessaloniki for the years 2001–2005 based on a model parameterized for the years 1995–2000. The data and predictions are resolved on a weekly basis per m^3 of air. The units in the range have been standardised and logarithmically transformed. We plot $X_t = \log(P_t + 1)$, where P_t is the pollen level, against time t

This is especially true in the case of pollen data, where variability is strongly skewed. Covariates such as temperature and humidity can also be built into this forecasting approach (Moseholm et al. 1987).

Figure 4.4 shows an example of this approach applied to a pollen series (olive) from Thessaloniki, for the years 2001–2005. It is clear that this model can be used to fit pollen data and it should be possible to make predictions of future pollen concentrations, at least for certain groups.

However, the basic Box-Jenkins approach can also be quite limited. Figure 4.5 illustrates some major difficulties of forecasting in the context of pollen dynamics. Noting that the scale is logarithmic, where the model fails to predict, the divergence can be very large (e.g. end of 2001, first cycle). Seasons can be irregular, so in 2003 (third cycle), the season begins later and is shorter than expected. Variations in the seasonality of pollen concentrations are primarily driven by ecological factors that may in turn be driven by climate or ecological interactions leading to shifts of the peak (Ocana-Peinado et al. 2008). The statistical behaviour of the counts does not conform to the usual patterns. For example, though data are extremely right skewed, a log-transformation does not remove it (Aznarte et al. 2007). The background "noise" generates variability on all scales. It cannot be removed by smoothing. For example, in Fig. 4.4, it is clear that the cleft in the peak moves about on a scale of weeks. This multi-scale behaviour (Halley and Inchausti 2004) has major consequences for the design of forecast procedures. In Fig. 4.4, the data have been normalised by year, that is, each year's data have been scaled so as to lead to the same total for each year. Figure 4.5 shows weekly data, without this pre-processing, for the family Poaceae, in Thessaloniki, over the years 1996–2004. Yearly counts vary considerably, so this introduces an extra twist that illustrates the direction we need to take in pollen season forecasting.

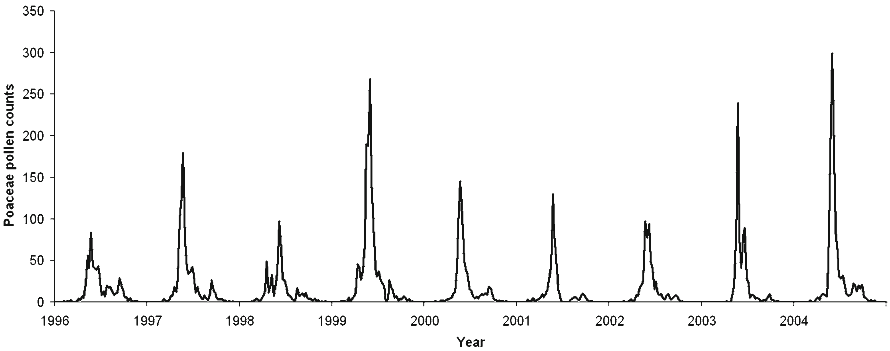

Fig. 4.5 Weekly counts of pollen of the Poaceae family in Thessaloniki, for the years 1996–2004 inclusive. The data are resolved on a weekly basis per m^3 of air. Because of the large between-year variability, no model curve is drawn. The time axis begins on January 1, 1996 and ends December 31, 2004. There is no transformation of pollen levels P_t

There is clearly a year to year variation of pollen counts. Thus, we have a more suitable model for pollen prediction:

$$X(t) = M + Tr(t) + A(y_t)\cdot S(m_t) + \varepsilon_t \tag{4.3}$$

Here, the seasonal component is the product of a random annual component A for the year y_t, and the cyclic seasonal component that depends only on the month, m, as before. This modified Box-Jenkins model addresses some of the multiple-scale variability that needs to be included in any attempt to forecast pollen levels.

Other time-series based approaches include neural networks (Arizmendi et al. 1993) or neuro-fuzzy approaches (Aznarte et al. 2007) and functional regression (Ocana-Peinado et al. 2008). To assess the relative success of forecasting, various comparisons have been made, but as yet have been mainly confined to specific places, taxa and timescales. Thus, there is a need for considerable work on the multiple-scale nature of pollen variability, which can be addressed within a suitably modified Box-Jenkins framework or using other time-series based methods. It is too early to say, which of the methods is the best. Investigations into appropriate measures of deviation of models from data are needed to quantify the relative merit of different models.

4.3.1.3 Computational Intelligence

Environmental data are very complex to model due to underlying interrelations between numerous variables of different type. However, as standard statistical techniques may possibly fail to adequately model complex, non-linear phenomena and chemical procedures, the application of Computational Intelligence (CI) methods for forecasting of a wide range of air pollutants and their concentrations at various time scales, perform usually well in atmospheric sciences.

Computational Intelligence methods, such as Neural Networks, Classifications and Regression Trees, Self Organising Maps, Support Vector Machines, etc. are advanced tools for knowledge discovery and forecasting parameters of interest. CI methods can be used for multiple tasks, such as classification, numerical prediction, clustering etc., while the main advantage of these methods is the accuracy combined with computational efficiency. CI techniques such as Artificial Neural Networks (ANNs), Classification and Regression Trees (CART) and Support Vector Machines have already been applied for analysing and forecasting air pollution parameters (Slini et al. 2006; Karatzas and Kaltsatos 2007). The performance of CI methods is similar or in some cases better compared with that of deterministic models, when applied to the atmospheric environment (Kukkonen et al. 2003), thus CI methods are appropriate tools to be applied for the development of operational forecasting, among others.

The application of CI methods for analysing and modelling pollen concentration data has increased in the recent years, since it was identified that methods, such as Artificial Neural Networks and Neuro-Fuzzy models, clearly outperform traditional linear methods in forecasting tasks (Sánchez-Mesa et al. 2002; Ranzi et al. 2003; Aznarte et al. 2007). Most of these applications have taken into account daily average pollen concentrations and meteorological parameters, aiming at forecasting pollen concentration of certain species 1 to 5 days ahead. CI methods have also been applied in order to investigate the relationships between pollen and air pollution with very promising results (Voukantsis et al. 2010), while papers published concerning the use of CI methods for analysing and forecasting pollen data are appearing more and more frequently (Degaudenzi and Arizmendi 1998; Aznarte et al. 2007; Voukantsis et al. 2011).

The rest of this chapter presents a short description of some of the most popular CI methods applied in atmospheric sciences are included, based on Tzima et al. (2007).

Decision trees usually assume that the function f(x) to be learned, is constant in intervals defined by splits on the individual attribute axes. Internal nodes of the tree implement split decisions based on impurity measures (defined in terms of the class distribution of records before and after splitting), while leaf nodes define "neighborhoods" of records, each of which is assigned a specific class attribute value (class label).

In neural networks the target function f(x) is implemented as a composition of other functions $g_i(x)$:

$$f(x) = K\left[\sum_i w_i g_i(x)\right], \tag{4.4}$$

where K is some predefined transfer function, such as a member of the sigmoid family (typical for multi-layer perceptron networks) or a radial basis function (as in RBF Networks). Given a specific task to solve, and a class of functions F, the set of observations is used in order to find the optimal target function that minimizes a predefined cost function. For CI applications, where the solution is dependent on the training data, the cost must necessarily be a function of the observations, such as the mean-squared error between the network's output, f(x), and the target class value y over all the example pairs.

Neural networks can be effectively applied to classification problems, even in the presence of large datasets. However, the resulting model's robustness depends heavily on the appropriate choice of the model (network size and topology), the cost function and the learning algorithm. Inappropriate implementations, combined with bad choice of a training data set, typically impair the classifier's generalisation ability or lead to model overfitting.

The self-organising map (SOM) also referred to as Kohonen Network, is a subtype of artificial neural networks. SOM is based on competitive learning, which runs in an unsupervised manner, aiming at selecting the so called winning neuron that best matches a vector of the input space. In this way, "a continuous input space of activation patterns is mapped onto a discrete output space of neurons by a process of competition of the neurons in the network" (Haykin 1999). This makes SOM one of best methods for modelling a knowledge domain with the aim to reveal topological interrelations and hidden knowledge, via the visualisation of the network's neurons.

SOM is capable of learning from complex, multi-dimensional data without specification of the output, thus making it very appropriate to be applied in pollen and atmospheric quality data. The resulting nonlinear classification consists of clusters that can be interpreted via visual inspection. The method's unsupervised learning algorithm involves a self-organising process to identify the weight factors in the network, reflecting the main features of the input data as a whole. In that process, the input data is mapped onto a lower dimensional (usually two-dimensional) map of output nodes with little or no knowledge of the data structure being required (a characteristic of the method that renders it appropriate for data compression). The output nodes (neurons) represent groups of entities with similar properties, revealing possible clusters in the input data. It should be noted that, although the method is unsupervised in learning, the number of the output nodes and configuration of the output map (number of nodes included, etc.), need to be specified before the learning process.

Rule-based algorithms apply "if…then…" rules of the form (Condition)->y, where Condition is a conjunction of observable attributes and y is the class label, where the values are put. The collection of rules may contain rules that are:

- mutually exclusive (each record is covered by at most one rule) or not (the rule set is ordered or employs a voting scheme);
- exhaustive (each record is covered by at least one rule) or not (a record may not trigger any rules and be assigned to a default class).

Among others, advantages of rule-based algorithms include the fact that they are easy to interpret and highly expressive. Moreover, they are fast to generate and can classify new instances rapidly, with a performance comparable to that of decision trees.

Bayesian classifiers compute conditional probability distributions of future observables given already observed data. More specifically, the analysis usually begins with a full probability model – a joint probability distribution for all attributes including the class – and then uses Bayes' theorem to compute the posterior probability distribution of the class attribute. The classifier's prediction is the value of the class attribute that maximizes this posterior probability. Naïve Bayes classifiers additionally assume independence among all attributes, given the class, when computing posterior probabilities.

Despite the fact that the independence assumptions made by Naïve Bayes classifiers are often inaccurate, the latter have several interesting properties that may prove useful in practice. They are robust to isolated noise points and irrelevant attributes and can handle missing values by ignoring the instance during probability estimate calculations. Moreover, their independence assumption allows for each distribution to be estimated independently as a one dimensional distribution, thus alleviating problems such as the "curse of dimensionality". Finally, another advantage of all Bayesian classifiers is their conceptual and interpretation simplicity, rendering them appropriate for use by non-domain experts.

Support vector machines (SVMs) were introduced by Vapnik in 1963. The original algorithm defines a method for finding the optimal hyperplane that separates, with the maximum margin, a set of positive examples from a set of negative examples. Thus, it is a linear classifier. A later extension of the algorithm, though, proposes the use of the "kernel trick" to maximum-margin hyperplanes: every dot product in the original algorithm is replaced by a non-linear kernel function, allowing the transformation of the feature set to a high-dimensional space, whose hyperplanes are no longer linear in the original input space.

4.3.2 Process-Based Phenological Models

Phenological models determine the entry dates of phenological phases as function of environmental factors. First efforts date back to the idea of Reaumur (1735), who explained spatial and temporal differences in phenological entry dates as a result of different heat sums during plant development. During the past years a number of review articles about phenological modelling have been published so that we will keep this sub-chapter as concise as possible. In addition, we will evaluate the current phenological modelling scene and touch a number of problems, which have been discussed only marginally in the literature. Special sections will deal with the application of phenological models to seasonal pollen modelling. More extensive reviews about phenological models are to be found in Hänninen (1995), Menzel (1997), Chuine et al. (1998, 2003), Chuine (2000), in Schwartz (2003), Schaber and Badeck (2003), Chuine and Belmonte (2004) and Linkosalo et al. (2008). The phenological models introduced here refer to plants of medium to high latitudes, whereas low latitude phenological modelling requires different approaches (Hudson et al. 2010).

Generally two different kinds of phenological models exist. The purely statistical-empirical approach relates the entry dates usually with mean temperatures over certain time periods preceding the phenological occurrence date. No mechanistic details of the relationship between plant physiology and environment are considered. The second kind of phenological models, also called process based models, is based on experimental studies about possible mechanisms, which are supposed to govern the relationship between plant physiology and the environment. During the vegetation period the buds for the next season are created, but remain in an inactive state called dormancy. During autumn and early winter dormancy is overcome through chilling. After dormancy has been released, the development of leaf, flower and shoot buds begins in the following spring. The timing of these events is the crucial point for the plants. If it occurs too early, frost might damage the plant organs, if it happens too late, the plant suffers from a loss of photosynthetic potential (Linkosalo et al. 2006). Although the high-temperature requirements of bud burst and flowering are well established, there is great uncertainty about the mechanisms enabling bud development. Among the basic factors governing the seasonal plant development are:

1. chilling temperature
2. forcing temperature
3. photoperiod
4. water availability

Phenological models may consider at least one or any combination of these four factors. There follows a short description of phenological models, which use individual factors or combinations of them, as found in literature.

4.3.2.1 Models Considering Thermal Forcing Only

The Thermal Time or Spring Warming model or Growing Degree Day models ignore the chilling requirement and consider only the temperature forcing in spring (Linkosalo et al. 2008). They describe the start of bud development in spring, omitting the dormancy phase altogether. Thermal Time models implicitly assume that environmental conditions required to release dormancy have been met before the starting date of temperature sum accumulation. The start date of temperature summation can be fixed or determined via an inverse procedure separately for each phase and station. The entry date t_2 is a function of the starting date of temperature accumulation t_1, and the temperature sum F between t_1 and t_2 above the temperature threshold T_b: $t_2 = f(t_1, T_b, F)$. The state of forcing F (forcing units usually in degree days) is represented by

$$F = \int_{t_1}^{t_2} R_f(T)dt \qquad (4.5)$$

where t_1 is the starting date of temperature accumulation, t_2 the entry date of the phase and R_f the rates of forcing, which are defined as

$$R_f = \begin{cases} 0 \;\; if\; T < T_b \\ TT_b \;\; if\; T \geq T_b \end{cases} \qquad (4.6)$$

where T is the daily mean temperature and T_b the temperature threshold.

A number of variants and simplified versions of Thermal Time model have been applied to the pollen period and compared with each other (García-Mozo et al. 2000). For instance the threshold temperature may be omitted and daily maximum temperatures may be added up from the end of the chilling period to the entry date. Most works prefer the daily mean temperature as input for the calculation of temperature accumulation, whereas some use alternatively the maximum daily temperature. Weighting of the temperature before summation has also been considered by some authors (Clot 1998; 2001).

An interesting alternative formulation of the thermal accumulation method is described in Aono and Kazui (2008), who consider the thermal energy accumulated during the developmental period of the plants. The daily DTS (number of days transformed to standard temperature) value is a ratio expressing the amount of growth that occurs in one day at the actual daily mean temperature with respect to that which occurs at the temperature set as a standard. The authors claim their method to produce a generally 0.4 days lower RMSE in comparison to the conventional degree day models.

4.3.2.2 Models Considering Chilling Only

For some species it turned out that there exists a useful relationship between the date, when the required chilling hours have been accumulated and the onset date of bud burst. Orlandi et al. (2004) tested two different chilling models and their ability to predict the onset date of bud burst in olive trees. Similarly, it appears that the chilling requirements exert a greater influence than the heat requirements for the start of black alder pollen release in the Mediterranean region (Jato et al. 2000)

4.3.2.3 Models Considering Thermal Forcing and Chilling

The following models include a description of the dormancy and the thermal forcing factor. Chilling requirement must be met before ontogenetic development can commence. Here follows a short description of each of the thermal forcing and chilling models with their underlying speculative assumptions:

- Sequential models are based on the assumption that chilling units must have been accumulated completely before accumulation of heat units can commence (Linkosalo et al. 2006).
- Parallel models consider chilling and forcing factors too, but assume that both processes may proceed in parallel.
- Alternating models assume a negative exponential relationship between the sum of forcing units required for completion of quiescence and the sum of chilling units received (Chuine et al. 2003)
- The deepening-rest model stipulates that the state of chilling must increase, before it can loosen its block on assimilation of heat units again.

- The Four Phases model assumes three phases of dormancy (pre-rest, true-rest and post-rest) before the phase of quiescence. This is formalised by an increasing temperature threshold for forcing during pre-rest and a decreasing temperature threshold for forcing during post-rest, and buds cannot respond to forcing temperature at all during true rest (Chuine et al. 2003).

4.3.2.4 Models Considering Thermal Forcing, Chilling and Photoperiod

The evidence showing that the dormancy is released solely by the chilling requirement is far from solid (Linkosalo 2000). There is in fact evidence that increasing day length has to do with the onset of ontogenetic development.

4.3.2.5 Models for Herbaceous Species Considering Temperature, Photoperiod and Soil Water Availability

Among the commonly recognised environmental factors governing the beginning of flowering of grasses, such as temperature and photoperiod, water availability plays a dominant role, especially in Mediterranean areas (Clary et al. 2004). Although various authors have developed models for predicting daily grass-pollen concentrations (Moseholm et al. 1987; Emberlin et al. 1999; Sánchez-Mesa et al. 2002), few papers have addressed the development of models to forecast the main pollen-season dates, i.e. start date and peak date (Clot 1998; Chuine and Belmonte 2004; Laaidi 2001a; Stach et al. 2008; García-Mozo et al. 2009). The main difficulty in developing forecasting models for this taxon is that grass pollen counts are an amalgam of pollen from many species, and pollen release dynamics prompt a large number of peaks (Férnández-González et al. 1999; Emberlin et al. 1999). García-Mozo et al. (2009) developed process-based models to predict the start- and the peak-date of the grass pollen season. The models take into account the effects of temperature, photoperiod and water availability on the timing of grass flowering in Spain. Apart from predicting the pollen-season start and peak dates, process-based models provide information on (i) the Poaceae response to weather-related factors, (ii) the period during which these factors affect grass growth, and (iii) the relationship between photoperiod, temperature and water availability for the flowering of grasses.

4.3.2.6 Generalised Phenological Models

There are two even more generalised models, which can be summarised in a separate group (Linkosalo et al. 2008). They are based on the idea that a model with a flexible structure will conform to the essential features of phenological control when fitted to a dataset.

- The Unified model was developed by Chuine (2000), where various weighting functions regulate the relationship between temperature and the development of chilling and forcing. Weighting is summarised by two generalised functions with all together nine parameters, which must be determined by a numerical optimisation procedure.

- The Promotor-Inhibitor model by Schaber and Badeck (2003) is based on the idea that a hypothesised balance or ratio between promotory and inhibitory agents determines the physiological state of development of the plant and its reaction to external driving forces.

4.3.2.7 Thermal Time Models Incorporating Real Time Phenological and Pollen Data

A number of authors experimented successfully with the idea to hinge the Thermal Time model forecast on the observation of entry dates of previously flowering species (Driessen and Moelands 1985; Driessen et al. 1989; Frenguelli and Bricchi 1998; Norris-Hill 1998). This approach requires real time phenological observations or pollen counts of the preceding species, which might not be readily available everywhere.

4.3.2.8 Optimisation of Model Parameters

From the schematic representation of for example the Thermal Time model $t_2 = f(t_1, T_b, F)$ as explained above, it becomes clear that the three parameters t_1, T_b and F have to be determined such that the phenological model yields best results with lowest error values.

In many cases it may be sufficient to work with a-priori fixed parameter values. From experimental evidence, for instance it appears that for a great number of species in the temperate zone 5°C represents an optimum threshold T_b (Frenguelli and Bricchi 1998; Jato et al. 2000). For colder regions 0°C has been suggested (Gerad-Peeters 1998; Clot 1998) and in warmer climates some authors have proposed 12.5°C (Alcalá and Barranco 1992; Galán et al. 2001a).

It is difficult to generalise model parameter values across a number of species or over large areas, as it turned out that the threshold temperature of certain species and phases depends on environmental factors, like the bio-climatic region and altitude. In the case of olive, the optimum threshold temperature was 10°C in Malaga (5 m.a.s.) but 12.5 in Córdoba (123 m.a.s.) within the same bio-climatic belt (thermo-Mediterranean). Different plants in the same locality have different temperature requirements: i.e. in Córdoba province, it has been defined at 11°C for oaks (early spring flowering), and 12.5°C for olive (late spring flowering, Galán et al. 2001a, 2005; García-Mozo et al. 2002. Ribeiro et al. (2006) suggest around 9°C for the olive in Portugal, but Orlandi et al. (2005) use temperatures between 7 and 15°C for the olive, in Spain and Italy, respectively.

For the start date of temperature accumulation t_1, a number of suggestions can be found in the literature. In Europe, 1 January has been suggested for early flowering species (Frenguelli and Bricchi 1998; Ribeiro et al. 2006; Orlandi et al. 2005), whereas 1 March for late flowering ones (Alba and Díaz de la Guardia 1998; Clot 1998).

Today's numerical techniques make it comparatively easy, to determine the optimum values of model parameters. A number of authors rely on LUT (Look Up Table) methods to optimise model parameter values. The phenological model is applied through a range of parameter values with a set increment. With the help of a cost function, which can be the squared error, standard deviation or root mean square error (RMSE), the optimum values are determined (for instance Van Vliet et al. 2002; Crepinsek et al. 2006; Migliavacca et al. 2008). If the dimensionality of the phenological model is not too high (not more than two or three model parameters), the LUT results can be plotted and viewed (Fig. 4.6). In case of phenological models, it turned out that the RSME values form a valley with any number of very differing optimum parameter values along the valley floor with similarly low RSME values. One may conclude that there is no unique solution to the problem and any one from an infinite number of parameter value sets is equally well describing the phenological behaviour. The objective selection of the most adequate set of parameter values constitutes a problem, which has not been solved yet. One way to arbitrarily select the most appropriate set is to calculate a mean from a certain fraction of the best parameter values.

Apart from the graphical visualisation of the minimisation problem, another advantage of the LUT method consists in its robustness. But the computational effort increases quickly with each additional dimension respectively model parameter to be optimised.

The other group of works prefer numerical methods to find the optimum parameter values of the phenological model. Kramer (1994, 1995) applies various numerical procedures (FITNONLINEAR from the GENSTAT package, NAG subroutine E04FCF, Downhill Simplex from Press et al. (1992) or a Newton approximation), whereas Linkosalo et al. (2009) rely on the direct search algorithm of Hooke and Jeeves (http://www.netlib.org./opt/hooke.c). The Simulated Annealing Algorithm provided in Press et al. (1992) has gained some popularity among phenological modellers (for instance Chuine et al. 1998 or Schaber and Badeck 2003). In comparison with the LUT methods, the numerical algorithms are computationally much more efficient, especially, if the phenological model needs considerably more than two or three parameters to be optimised. On the other hand, the procedure may converge or not to the global minimum. The Simulated Annealing Algorithm tries to overcome that problem by introducing a random fluctuation, which helps the procedure to step over local minima. In some cases, it might be an advantage to apply more than one method, especially, if the results of a numerical procedure are doubtful.

To assess the model quality, Galán et al. (2005) provide Root Mean Square Errors (RMSE) for a temperature sum model for olive in Spain. The RMSE range between 6.2 and 7.8. The mean absolute difference between the modelled and the actual date was 4.8 days using independent data. In a similar but older study (Galán et al. 2001a), this number was 4.7 for olive (though not tested on independent data).

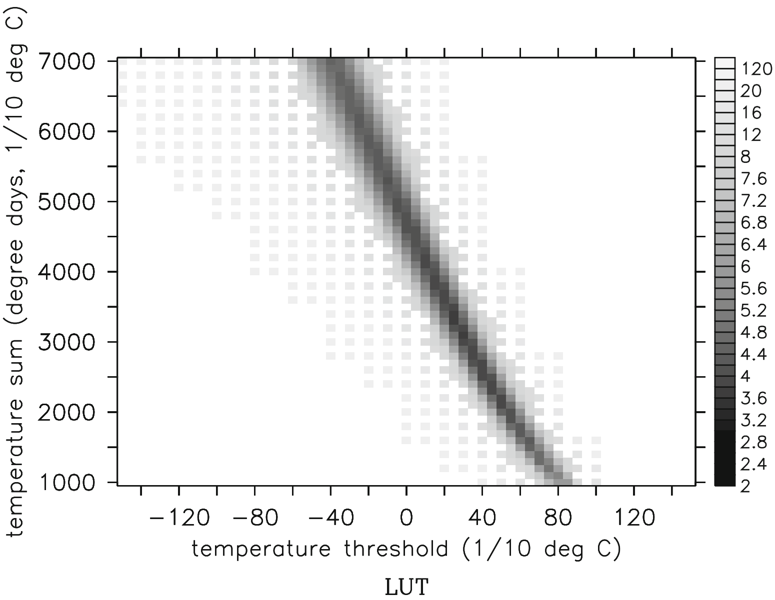

Fig. 4.6 Sample plot of a LUT (Look Up Table): the phenological phase is lilac beginning of flowering at the Austrian station of Kremsmuenster (1951–2004), the minimisation function is the RMSE (Root Mean Square Error) depicted in various shades of grey at starting date (t_1) yearday 71 (2 March). A three step approximation procedure selects only the relevant area and leaves the rest white

4.3.2.9 Application of Process Based Phenological Models to Pollen Season Modelling

The year-to-year variability of the beginning of the flowering season is strongly linked with the year-to-year variability of the atmospheric conditions prior flowering inception. The modelling and forecasting of the start of the flowering season as function of the atmospheric conditions is therefore very useful for the pollen forecast procedure. Once flowering has started, the subsequent temporal development of the pollen concentration follows a certain pattern, which can also be modelled (Linkosalo et al. 2010).

The application of phenological models to predict the start, peak and end of the pollen season of various anemophilous species appears well established and generally yields reliable results (Thibaudon and Lachasse 2005). Nevertheless, the application of phenological models to the pollen season modelling is not straightforward and therefore the underlying ideas should be critically reviewed. The following two assumptions are tacitly expected to be true:

- Assumption 1. If the start of the local flowering season has been observed, local pollen shedding has also started.

- Assumption 2. If pollen of a certain species has been recorded by the local pollen sampler, local pollen shedding has started.

If Assumption 1 holds, one could apply a phenological model to predict the entry of the flowering phase and thus have an indication of the beginning of the local pollen season. This has actually rarely been done because of a number of reasons. Generally, the species range of pollen data is much wider than that of phenological networks. Phenological and pollen networks have been created independently, are not coordinated and are usually run by different organisations, so that pollen and phenological species overlap only to a minor extent in most networks. Only in a few cases the density of phenological data are sufficient to calibrate a phenological model to support the forecast of the beginning of the pollen season.

Assumption 2 implies that pollen is not transported over larger distances and that the local pollen record faithfully reflects the local pollen shedding after flowering of the local plants has commenced. But Estrella et al. (2006) did show that in the case of birch, in Germany, Assumption 2 needs not be true. They found major temporal discrepancies between local phenology and local pollen concentration. Pollen can be transported over large distances from areas with currently flowering plants to the recording site, where plants have either not yet commenced flowering or have stopped flowering already. Therefore, the locally observed entry date of the phenological phase of flowering, the locally recorded beginning of the pollen shedding and the beginning of the pollen season may not be identical.

Usually, Assumption 2 is applied and a phenological model is fitted for a date relevant for the pollen season, like start and end, according to a selection criterion, or peak of the pollen season. In order to convert pollen concentration values to one or more of such dates, a range of definitions has been suggested in literature. Although transport processes influence the local pollen concentration, pollen season start dates (regardless of how they are defined) can be modelled as function of temperature sums.

4.3.3 Special Problems in Pollen and Phenological Modelling

4.3.3.1 Application to Large Areas

When designing a pollen forecast procedure, one has to choose an area large enough to accommodate for a possible long range transport of pollen. This, in turn, requires the modelling of the start and progress of the pollen season over a larger area, beyond national boundaries and with a spatial resolution, much higher than that of existing networks. In complex terrain, for instance, it makes much sense to calculate the phenological entry dates on a grid with a high spatial resolution, either few km or even <1 km, because deviations in elevation between the DEM and real topography can cause a substantial shift of entry dates modelled on a grid (entry dates may vary between 20 and 40 days/1,000 m elevation, Scheifinger et al. 2002). Typically, the density of the phenological, pollen or meteorological networks is not high enough to enable such a procedure for the required spatial resolution.

The spatial robustness of phenological models has been tested with a set of European phenological and pollen data (Chuine 2000; Chuine and Belmonte 2004). In some cases, models fitted with data from one station set predicted pollen season entry dates well at a set of neighbouring stations or even at stations more than 900 km apart. If the model gave reliable results in the area, where it was fitted, the probability was high that it also worked well at distant stations. The geographical range of the applicability of the model parameters appeared to be to some extent also species dependent.

García-Mozo et al. (2008) grouped Spanish pollen stations according to phenological model parameter values. Phenological models fitted with local pollen and meteorological data yielded the best results (75–95% explained variance); phenological models fitted with regionally deduced parameter values resulted in lower explained variances at individual stations (55–85%), whereas phenological models fitted with parameter values deduced on the basis of all stations gave the lowest explained variance (51%). One might conclude from that study that in order to achieve the best fit, only locally deduced model parameters should be applied or a method needs to be found to interpolate the model parameter values to each individual climate station or grid point.

White et al. (1997) developed a phenological model for the onset of greenness of deciduous broad leafed forests and grasslands of the temperate zone based on the Thermal Time model approach including radiation. They found that the temperature sums combined with radiation sums at onset of greenness are a function of average annual temperature and radiation. Applying this relationship they were able to model the onset of greenness over the contiguous US with a high degree of accuracy (mean absolute errors ranged from 5.3 to 7.1 days).

4.3.3.2 Real Time Modelling

Numerical pollen forecast procedures require real-time operational phenological models and the spatial interpolation of phenological entry dates (Helbig et al. 2004; Sofiev et al. 2006). The temporal development of flowering in space with the subsequent pollen emission is the essential input for atmospheric transport models. Phenological real-time observation is still in its infancy and cannot be used for the purpose of pollen forecast. Therefore, phenological models have to simulate the developmental stages of the plants in real time. If the entry of the flowering phase has been calculated in an area, another model must assess the quantity of pollen emitted into the atmosphere, which is then input for the dispersion model. All process-based phenological models are basically suitable for such a real-time application. As real-time phenological observation systems (remote sensing via satellites and digital cameras) and real-time pollen measuring devices are being developed, the question of assimilation of such data into the operation models becomes relevant in the near future.

In some cases, the pollen forecast procedure is designed so that an interpolation of observed or modelled phenological entry dates is not necessary. If the prognostic atmospheric model provides a temperature field with the required spatial resolution, the phenological model can directly be applied to the temperature data on the same grid (Puppi and Zanotti 1992; Kawashima and Takahashi 1995; Hidalgo et al. 2002). Possible model temperature biases have to be taken into account.

4.3.3.3 High Resolution Spatial Representation of Phenological Entry Dates

In other cases it might be desirable to interpolate phenological entry dates observed at network stations or modelled on a comparatively coarse grid to a Digital Elevation Model (DEM) with a higher spatial resolution (for a more detailed overview see Jeanneret and Rutishauser 2010). For pollen modelling purposes, for instance, the spatial distribution of phenological entry dates must be available on a DEM, which resolves the main topographical features of the area. A great mismatch between the spatial resolution of the phenological information and the real topography inevitably leads to an equivalent mismatch between the modelled entry dates on the DEM and on the real topography.

A number of methods have been developed to produce phenological maps. Beginning with Ihne's (1885) work, a good historical overview and a description of the theoretical background of spatial interpolation of phenological observations is given in Puppi and Zanotti (1989). If the terrain is largely flat or the grid is rather coarse, one might just interpolate the entry dates straightforward with Inverse Distance Weighting (IDW, Ahas et al. 2002; Scheifinger et al. 2002). If the area is large (from a few hundred up to a few thousand kilometer in diameter) and it turns out that the relationship between phenological entry dates and space is strict (for instance >70%), a multiple regression model can be applied, where the phenological entry dates are modelled as a function of station longitude, latitude and elevation (Rötzer and Chmielewski 2001). Small scale topographical features are considered by height reduced methods, like reduced detrended Kriging (Badeck et al. 2004) or via a radiation model, which is applied to a high resolution DEM to subsequently provide the spatial weights for the phenological entry dates (Chytry and Tichy 1998). A more complex approach is presented by Puppi and Zanotti (1989), where the phenological entry dates are calculated as a function of a number of independent environmental variables, like altitude, slope, incident solar radiation, tree layer cover, urbanisation or geomorphologic features (sides and bottom of the valley) via a set of regression equations. Geostatistical software can facilitate interpolation and visualisation of interpolated fields (García-Mozo et al. 2006).

4.4 Discussion and Summary

This section is thought to present a few ideas, which pop up when one begins to reflect about the compiled snap shot of the current state of the art about monitoring and modelling of pollen counts, pollen season and phenology.

Each of the highly diverse scientific disciplines contributing to aeropalynology has its own history, where tools and technologies have been developed and data sets collected. These factors could be called science intrinsic factors. Similarly, external factors have evolved related, for instance, with the public health issue of pollinosis. Driven by external factors, the accumulated expertise of each discipline is being summarised by a small scientific community within the field of aeropalynology, which has just begun to exploit their heritage in a most fruitful manner. Pollen transport models, which are being developed by a number of European weather services, can be cited as an example for such a combined interdisciplinary effort, where biologists, meteorologists and physicians work together.

Plant physiology, atmospheric dispersion and the human immune system are rather complex research objects, so that progress in many aspects of aeropalynology is counterbalanced by an increasing number of unresolved questions. Thus, this field appears both challenging and fascinating.

4.4.1 Monitoring

The chapter about monitoring reviews the current situation of three data sources, which are directly related with aeropalynology: phenological observations, pollen counts and remote sensing of the vegetation activity. Up to now, all three data sets more or less co-exist, without much exchange or fusion. Any assimilation of two or all three of them into one is in its infancy, if existing at all. A number of assimilation techniques have been developed in earth sciences, from which suitable ones could be chosen and adapted. Phenological observations, pollen counts and remote sensing information on the state of the vegetation could then be assimilated into pollen emission models and finally into the numerical pollen forecast. Pollen modelling would benefit a great deal from such a data fusion.

Over the last decades, consistent monitoring efforts of various national networks have created a wealth of pollen concentration time series. These constitute a nearly untouched treasure, which is still to be exploited to investigate questions concerning pollen emission, transport and deposition.

New monitoring methods emerge, which allow measuring the allergen content in pollen. This adds a new dimension to the problem of pollen related allergies. Results from research on the allergen content in pollen, like the HIALINE project (http://www.hialine.com/en/klinikum-rechts-der-isar-der-technischen-universitaet-muenchen.php) are expected to make the operational pollen forecasts more specific, which in turn helps the sufferers to improve their avoidance strategies.

4.4.2 Modelling

Although process-based phenological models have been around for a couple of decades, a number of problems remain to be solved, when applying them to statistical and numerical pollen forecast models:

- Model quality
  - Model quality is restricted by the noise inherent in the data. Phenological entry dates are observed subjectively and pollen counts are influenced by plant distribution and atmospheric factors. Models can not be more accurate than the observations they are based on.
  - The Thermal Time Model appears to exhaust the noisy information contained in commonly available observational data sets. Attempts have been made to explicitly incorporate plant physiological concepts into the process-based phenological models, but model quality could not substantially be improved beyond that of the simple Thermal Time models (Chuine 2000; Schaber and Badeck 2003; Linkosalo et al. 2008).
- Operational statistical or numerical pollen forecast ideally requires the model results over a large area on a grid. Up to now, only a few studies have proposed methods to model phenological entry dates over a larger area and a practical solution is still absent.
- As already mentioned in the monitoring section, the assimilation of phenological observations, pollen counts and remotely sensed information about vegetation will emerge as one of the central topics in the field of numerical pollen forecast.

Regression models, where the pollen count is modelled as function of a number of environmental variables, are well established and widely used to reliably improve the operational day to day pollen forecast. More elaborate statistical techniques, like computational intelligence methods, have still to become established for the operational pollen forecast. Some statistical packages offer such methods, like neural networks in the SPSS package, which can be run on personal computers.

The question, which of the models, regression or process-based, is superior, cannot yet be answered. Reviewing the wide range of models for forecasting the start of the pollen season, no superior model can be identified. Laaidi et al. (2003) employed a temperature sum model and a regression model using a number of predictors (air temperature, rainfall, relative humidity, sunshine duration and soil temperature) to forecast the start of the pollen season of *Ambrosia*. The regression model performed better during calibration, but the temperature sum model showed better results when tested on independent data. Chuine et al. (1999) conclude from a model comparison study that there is no single model that accurately predicts the dates of flowering of every species. Depending on the species, different models may perform best. Even among a single species there is not one model performing best, because relationships between the species and the environment may differ according to the climatic region. This emphasises the importance of careful evaluation and testing of various models for predicting the start of the pollen season.

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