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Seasonality in quantity of atmospheric fungal aerosol in Tartu (Estonia)

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Abstract: The quantity of fungal spores collected in routine aerobiological monitoring of atmosphere in Tartu 1989–1994 was studied with respect to two aspects: daily mean concentration (measured as the number of spores in cubic metre of air in 24 hours), and spore totals of a period (measured as the number of spores in cubic metre of air in a period). The aerobiological calendar method was used to indicate seasonal character of the quantity of spores. During the 6-year period the main spore season occurred from March to November, the total annual amount of fungal spores exceeded 1×10^6 spores in cubic metre per year. The seasonal dynamics was expressed clearly in both daily mean concentrations of spores and seasonal spore totals.

Kokkuvõte: Atmosfääris esinevate seeneeoste hulga sesoonsus Tartus.

Tartus aastatel 1989–1994 aerobioloogilise seirega kogutud seeneeoste hulka uuriti nii ööpäevase keskmise kontsentratsiooni kui aastaajati esinenud hulkade seisukohast, kasutades aerobioloogilise kalendri meetodit. Kuue aasta keskmisena leiti, et seeneeoste põhihooaeg kestab märtsist novembrini, aastane eosehulk kuupmeetris õhus ületab miljoni piiri, ning et sesoonsus on tugevasti väljendunud nii ööpäevases eosekontsentratsioonis kui aastaajalistes eosehulkades.

INTRODUCTON

Organisms utilise the atmosphere as a medium to move from one terrestrial habitat to another (Isard & Gage, 2001). A great amount of fungi – it was estimated by Kendrick (1990) that there are over 100000 fungal species whose spores become airborne – accomplish this by drifting passively and allowing atmospheric motion systems to transport their spores. That is why fungal spores are present in the air abundantly and ubiquitously. The composition and concentration of atmospheric spores as well as factors that impact them have been studied in great detail worldwide; reviews of earliest work are given by Stepanov (1935, 1962), Ingold (1965), Gregory (1973), and Edmonds (1979); one of the latest studies is made by Zoppas et al. (2006).

Atmospheric fungal spores have frequently been studied on aims to understand their impact on health, environment, or on agricultural and forest production. In these cases, only certain taxonomic or morphologic groups of spores are considered, and the population of airborne spores has been treated only partially. They could also be treated totally – all spores together as the fungal part of the atmospheric aerosol, which is known also as the "coarse aerosol" that mainly has the aerodynamic equivalent diameter in the size range of 2.5–10 μm (Bauer et al., 2002). In such cases it is expected that the data on fungal aerosol represent large temporal and spatial units, such as decades of years and a geographical area which circumscribes the distribution of a major assemblage of fungi and plants. To get such spore data, the monitoring of spores must fulfil four demands. (1) Aerosol particles are collected using volumetric method. (2) Spores are counted directly using microscope. (3) Collection of particles is carried out at the height of 10–25 m above ground level. (4) Monitoring continues longer than two decades. There are few studies in which spore monitoring fulfilled these demands (Millington & Corden, 2005; Corden et al., 2003; Corden & Millington, 2001; Hollins et al., 2004). All of those studies concern airborne spores of certain genera, not the whole population of atmospheric fungal spores. For all spores, a 10-year series is the longest which has been studied (Hjelmroos, 1993). Stockholm, Sweden (Hjelmroos, 1993) and Turku, Finland (Rantio-Lehtimäki et al., 1985) are the sites nearest to Estonia, where the whole population of atmospheric fungal spores are studied.

For many users of spore data it is essential that the data on the quantity of atmospheric fungal aerosol represent not only decades and landscape regions but annual dynamics as well.

In mid- and high latitudes, due to great differences in distribution of solar radiation during the year, high seasonality occurs in climate, weather conditions, atmospheric aerosol, plants, fungi and in other components of biogeosphere. Phenological calendars (PC) are used for describing the seasonality of places, years, or individual species (Ahas & Aasa, 2003). For atmospheric fungal aerosol, the PC method could not be used exactly – developmental phases of those living organisms who are settled could not be involved. This mismatch is caused by the diversity of processes that govern the influx of spores into the air. Spores appear in atmosphere due to 1) emission from local sources, 2) transport by air masses and 3) re-emission of once sedimented spores. The emission from local sources correlates with phyto- and myco-phenological stages of the site very well but that does not always do the transport by air masses. Transport by air masses, routine monitoring and easy identification are the prerequisites of the choice of indicators of seasonal events. The Estonian aerobiological calendar (Saar, 2001) has been used for measurement of time when describing the annual dynamics of atmospheric fungal aerosol (AFA).

The aims of the present paper are 1) to test the aerobiological calendar method for the analysis of seasonality in AFA, 2) to find seasonal regularities in the quantity of AFA on the basis of the data collected in routine aerobiological monitoring in Tartu from 1989 to 1994.

The quantity of spores is described as daily mean concentration of spores (the number of spores in cubic metre of air in 24 hours), and spore total of a period (the number of spores in cubic metre of air in a period).

Concentration dynamics of airborne fungal spores in Tartu was partially studied by Ülle Hanson (1995).

Explanation of terms:

PSST – spore total of pollen season, the sum of average spore concentrations of all days in an aerobiological pollen season; measured as the number of spores in cubic metre of air in pollen season

ESpST – spore total of early spring, the sum of average spore concentrations of all days in an aerobiological early spring; measured as the number of spores in cubic metre of air in early spring

Seasonal spore total – general name for spore totals of early spring (ESpST), mid-spring (MSpST), late spring (LSpST), early summer (ESuST), mid-summer (MSuST), late summer (LSuST), and early autumn (EAuST)

MATERIAL AND METHODS

Monitoring site

Estonia is located in a transition zone between continental and maritime climate in East Europe, in the mixed forests subzone of the temperate forest zone. Tartu is a town in the eastern part of Estonia, on the Ugandi Plateau (N-S distance about 90 km, W-E distance 60 km; variations in elevations in the interval of 30–120 m a.s.l). The main factors, contributing to the landscape character are plains with reddish-brown non-calcareous moraine and primeval valleys that separate the plains (Arold, 2005). A river flowing in a NW-SE valley splits the town into halves. The monitoring site was situated at the SW edge of the town (58°21'23"N and 26°40'50"E), on the highest place (77 m a.s.l) of it and its surroundings.

The average values of climatic characteristics in Tartu during 1966–1998 were as follows: annual temperature 5 °C, monthly temperature with maximum of 16,7 °C in July and minimum of -6,4 °C in January, annual precipitation 605 mm, snow cower duration 106 days (Jaagus, 1999).

The main factors of the land cover of the Ugandi Plateau are forestland (about 50% of the area), arable land (40%), and swamps and bogs (11%) (Arold, 2005). Mixed forests have spruces, birches and pines, and to a lesser extent aspens, lindens and oak trees in the tree layer. Spruce forests and pine forests are also present. Grasslands are characterized by communities of *Festuca rubra – Cynosurus cristatus* and *Agrostis capillaris – Festuca rubra* (Arold, 2005). Alders and willows are widely spread. The vegetation in and around the town is rich and consists mainly of *Betula, Picea, Pinus, Tilia, Quercus,*

Acer, Populus, Fraxinus and *Ulmus* trees, as well as of grasses, among other plants. Drastic changes occurred in land cover in 1992–1993, when many cultivated fields were abandoned. Until 1991 the agricultural industry was both large and varied, with the predominance of grain crop, vegetables, and meadows and pastures. In 1992–1993 many fields were covered with weeds, replaced by grasses little by little.

Tartu is a university town as well as an industrial one (population 100000). In 1989–1994 there were 1–5 storey buildings mainly. The monitoring site was located in the semi-urban area, approximately 4 km of the town centre. In the surroundings as well as in the monitoring place, there were neither obstacles nor barriers to air motion.

Sampling and assay methods

Concentrations of pollen grains and fungal spores were determined using the methodology accepted in the routine aerobiological monitoring by European Aeroallergen Network. This methodology is characterised by the operational principles "impaction onto solid surface" for samplers and "direct counting using microscopy" for assay methods (Mandrioli et al., 1998). Aerosol particles were collected using a continuous volumetric 7-day sampler, a modified Hirst (1952) spore trap (Burkard Manufacturing Co.), with a flow rate 10 l minute^{-1}. It is an impactor with a 2×14 mm intake orifice through which the sampled air is impacted onto a collection surface moving at 2 mm h^{-1}. The sampler located on the roof of a building at the height of 14 m above ground and collected the aerosol all the year round. The tape (polyester Melinex film) was placed on the rotating drum and its collecting surface was coated with a mixture of vaseline and paraffin wax in toluene. The exposed tape was cut in 24-hour sections, each of them corresponding to one day. A section was mounted on a microscopic glass slide in a jelly medium Gelvatol and covered by a cover glass. Aerosol particles were observed directly by the magnification of 600×. By screening slides, random fields method was used (Mäkinen, 1981; Käpyla & Penttinen, 1981) whereas 8 random microscopic fields per 2 hours were studied, all together 96 fields per day. The studied area, i.e. the area from which all spores and pollen grains were counted, made up 1.2% of daily slide surface. Visual identification was based upon morphological features such as colour, size and shape. The morphological identification of spores and pollen grains was based on the reference collection of fungal spores and pollen grains, atlases (Kuprianova & Alyoshina, 1972, 1978; Nilsson et al, 1977; Bobrov et al., 1983; Nilsson, 1983; Wilken-Jensen & Gravesen, 1984;), manuals (Ogden et al., 1974), encyclopedias (Kremp, 1965), identification keys (Fægri & Iversen, 1989) and specific mycological literature (Bondartsev, 1953; Blumer, 1967; Ignatavičiutė, 1975; Minkevičius, 1984). All spores, except spores of Pteridophyta, were identified as fungal spores. Therefore, in the present paper, the term "fungal spores" is used in a large sense. The concentration of fungal spores was expressed as the average number of spores in day in cubic metre of air.

The monitoring of fungal spores was performed continuously from May 5, 1989 to December 31, 1994 extending throughout most of the year (Fig. 1). Spores were counted and their concentration was determined during the pollen seasons, while spore presence was checked in periods from the end of pollen season until the beginning of climatic winter.

Measurement of time

The aerobiological calendar method was used when time in annual cycles was measured. Temporal signposts of those seasons, which occur in the period from the end of the cold season through the warm season until the beginning of the next cold season (excl.), are chosen from among indicators of developmental phases of pollen aerosol (Tables 1 and 2). Temporal signposts of those seasons which occur in the period from the beginning of the cold season until its end (excl.) are chosen from among indicators of physical quality of the air.

RESULTS

Pinpoints of main spore seasons

From 1989 to 1994 the main fungal spore seasons covered 61–76% of annual circles (Table 2 and 3, Fig. 1). These spore seasons began, depending on the year, in different annual stages of the landscape: in climatic winter, in climatic late winter, or in climatic early spring. The main spore seasons of the years 1992 and 1993 started in the period with cold weather,

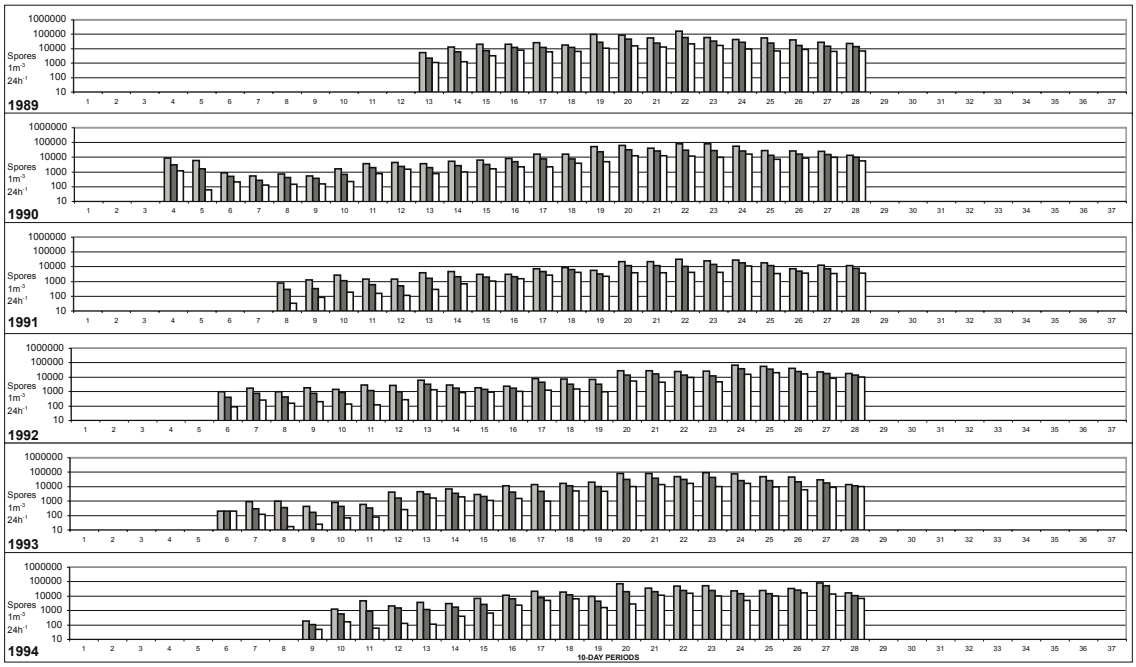

Fig. 1. Average and extreme values of daily mean concentrations in 10-day periods in Tartu 1989–1994. Data are absent for periods with no bars. Legend: dark grey – average value of concentrations in a period of ten days, pale grey – maximal value, white – minimal value.

Table 1. Indicators of the beginning of aerobiological seasons in Estonia

Season	Beginning indicator
Early spring	Appearance of pollen of the group of *Alnus* and *Corylus* into atmosphere
Mid-spring	Appearance of pollen of *Populus* into atmosphere
Late spring	Appearance of pollen of *Ulmus* into atmosphere
Early summer	Appearance of pollen of Poaceae* into atmosphere
Mid-summer	Appearance of pollen of Urticaceae into atmosphere
Late summer	Appearance of pollen of *Artemisia* into atmosphere
Early autumn	Permanent transition of pollen concentration of *Artemisia* through 30 grains 1 m^{-3} 24 h^{-1}
Late autumn	Disappearance of pollen from atmosphere
Winter	Permanent transition of air temperature through 0 °C

* valid for territory with non-calcareous cover (southern and eastern Estonia)

those of the years 1990 and 1994 in the period of snow melting, and that of the year 1991 after the final disappearance of snow cover and before the permanent increase of daily mean air temperature above +5° C. In five of six years, the main spore season ended by the first formation of snow cover and the first cold days when daily maximum temperature remained below zero. Thus, the main spore season was mostly stopped by the start of the period with unstable snow cover when freezing and melting alternates. In the year 1992, in the case of omission of climatic early winter, the main spore season ended after the formation of the period of cold weather.

Comparison between annual spore counts

There were considerable differences in the PSST (Table 3) within the 6 years monitored. The total for 1989 was 2.5 times of that in 1991. These totals were in a range from 1.21×10^6 to 3.1×10^6 spores m^{-3}. There was no correlation between the duration of pollen season and PSST.

Table 2. Beginning date and duration of aerobiological seasons in Tartu 1989–1994

Beginning date

Season	1989	1990	1991	1992	1993	1994	mean and range
Early spring	no data	Feb 11	March 17	Feb 22	Feb 28	March 29	March 5, [Feb 11, March 29]*
Mid-spring	no data	March 26	Apr 12	Apr 16	Apr 17	Apr 16	Apr 11, [March 26, Apr 17]*
Late spring	no data	Apr 14	May 3	May 4	Apr 25	Apr 26	Apr 27, [Apr 14, May 4]*
Early summer	May 16	May 21	June 10	May 22	May 17	May 31	May 25, [May 16, June 10]
Mid-summer	May 26	June 20	June 23	June 13	June 19	June 17	June 15, [May 26, June 23]
Late summer	July 19	July 27	July 26	July 17	July 20	July 18	July 21, [July 17, July 27]
Early autumn	Sept 4	Aug 18	Aug 25	Aug 10	Aug 24	Aug 21	Aug 22, [Aug 11, Sept 4]
Late autumn	Sept 25	Sept 26	Oct 11	omitted	Sept 29	Oct 1	Oct 5, [Sept 25, Oct 24]
Winter	Nov 22	Nov 15	Dec 5	Oct 10	Nov 9	Nov 8	Nov 12, [Oct 10, Dec 5]

Duration, in days

Season	1989	1990	1991	1992	1993	1994	mean and range
Early spring	no data	43	26	54	48	18	38, [18, 54] *
Mid-spring	no data	19	21	18	8	10	15, [8, 21] *
Late spring	no data	37	38	18	22	35	30, [18, 38] *
Early summer	10	30	13	22	33	17	21, [10, 33]
Mid-summer	54	37	33	34	31	31	37, [31, 54]
Late summer	47	22	30	24	35	34	32, [22, 47]
Early autumn	21	39	47	75 *[2]	36	41	43, [21, 75]
Late autumn	58	50	55	omitted	41	38	27, [38, 58] *[3]
Winter	81	122	79	140	140	104	28, [79, 140]

* in 1990–1994; *[2] early autumn finished when pollen season ceased out, in Oct 24; *[3] excluded 1992

Seasonal patterns

Despite the differences in PSST, seasonal pattern was similar in all years: the vernal part was much lower than that from mid-summer to autumn (Table 4). MSpST was the lowest, being in range from 4×10^3 to 29×10^3 spores m^{-3}. The peaks, depending on the year, were LSuST or EAuST; except the year 1990, when the MSuST was the biggest. The ranges of both LSuST and EAuST were from 3×10^5 to 16×10^5 spores m^{-3}. Three from seven seasonal totals – MSuST, LSuST and EAuST – contributed 92% of PSST (averaged over 5 years), while four of them – ESpST, MSpST, LSpST and ESuST – made up only 8% (Table 4).

Increase in proportion of the duration of a season did not always correspond to increase in proportion of a seasonal spore total (early spring 1993, early summer 1993, late spring 1994, mid-summer 1994; Fig. 2). In some cases of normal duration of the season, the proportion of the seasonal spore total was higher than normal (mid-summer 1990) or lower than normal (early autumn 1991, mid-summer 1992). In the case of the year 1991, the seasonal pattern of spore totals and season durations of was the closest to the 5-year mean one.

Daily fluctuations

The daily mean concentrations of spores fluctuated widely, ranging from 17 (in early spring 1993) to 159,260 spores m^{-3} (in late summer 1989). The ranges of the daily mean concentrations in 10-day periods are shown on Fig. 1, and those in seasons are presented on Fig. 3. The ranges show a regular pattern of seasonal variation. The range of late summer and early autumn concentrations differed clearly from those of vernal concentrations (Fig. 3). In the years 1990 and 1993 the late summer and early autumn ranges differed from all three vernal ranges – from the early spring, mid-spring and late spring ranges; while in the years 1991 and 1992 they differed only from the early spring

Table 3. Main seasons of fungal spores, pollen seasons and PSST in Tartu atmosphere in 1989–1994

Year	1989	1990	1991	1992	1993	1994
Duration of main spore season (days)	No data	277	263	245	254	224
Spore concentration data exist						
Start of pollen season	May 5 *[1]	Feb 11	March 17	Feb 22	Feb 28	March 29
End of pollen season	Sept 24	Sept 25	Oct 10	Oct 23	Sept 28	Sept 30
Duration of pollen season (days)	No data	227	208	245	213	186
Spore concentration data are absent						
Start of late autumn	Sept 25	Sept 26	Oct 11		Sept 29	Oct 1
End of late autumn	Nov 21	Nov 14	Dec 4		Nov 8	Nov 7
Duration of late autumn (days)	58	50	55	-14 *[2]	41	38
PSST (10^6 spores m^{-3})	3.10 *[3]	2.40	1.24	2.27	2.73	2.33

*[1] aerobiological monitoring began (somewhere in late spring)
*[2] the last days of pollen season overlapped by climatic winter
*[3] less than really (due to the missing vernal part)

Tabel 4. Seasonal spore totals in Tartu 1989–1994

Absolute values (10^3 spores m^{-3})

	1989	1990	1991	1992	1993	1994	mean
ESpST	no data	27	17	38	14	14	22
MSpST	no data	12	17	29	4	6	14
LSpST	no data	86	69	34	61	63	63
ESuST	55	154	64	40	115	109	96
MSuST	1115	819	286	199	533	262	420
LSuST	1560	545	390	342	1174	825	655
EAuST	369	753	399	1592	826	1055	925
PSST	3099	2396	1242	2275	2729	2334	2195

Relative importance (as ratio to PSST)

	1989	1990	1991	1992	1993	1994	mean
ESpST	no data	0.011	0.014	0.017	0.005	0.006	0.011
MSpST	no data	0.005	0.014	0.013	0.002	0.003	0.007
LSpST	no data	0.036	0.056	0.015	0.023	0.027	0.031
ESuST	0.018	0.064	0.052	0.018	0.042	0.047	0.045
MSuST	0.360	0.342	0.230	0.088	0.195	0.112	0.194
LSuST	0.503	0.228	0.314	0.150	0.430	0.353	0.295
EAuST	0.119	0.314	0.321	0.700	0.303	0.452	0.418
PSST	1.000	1.000	1.000	1.000	1.000	1.000	1.000

range. The vernal concentrations were lower than those of late summer and early autumn. The ranges of early summer and mid-summer concentrations overlapped partially in both vernal ranges, and late summer and autumnal ranges.

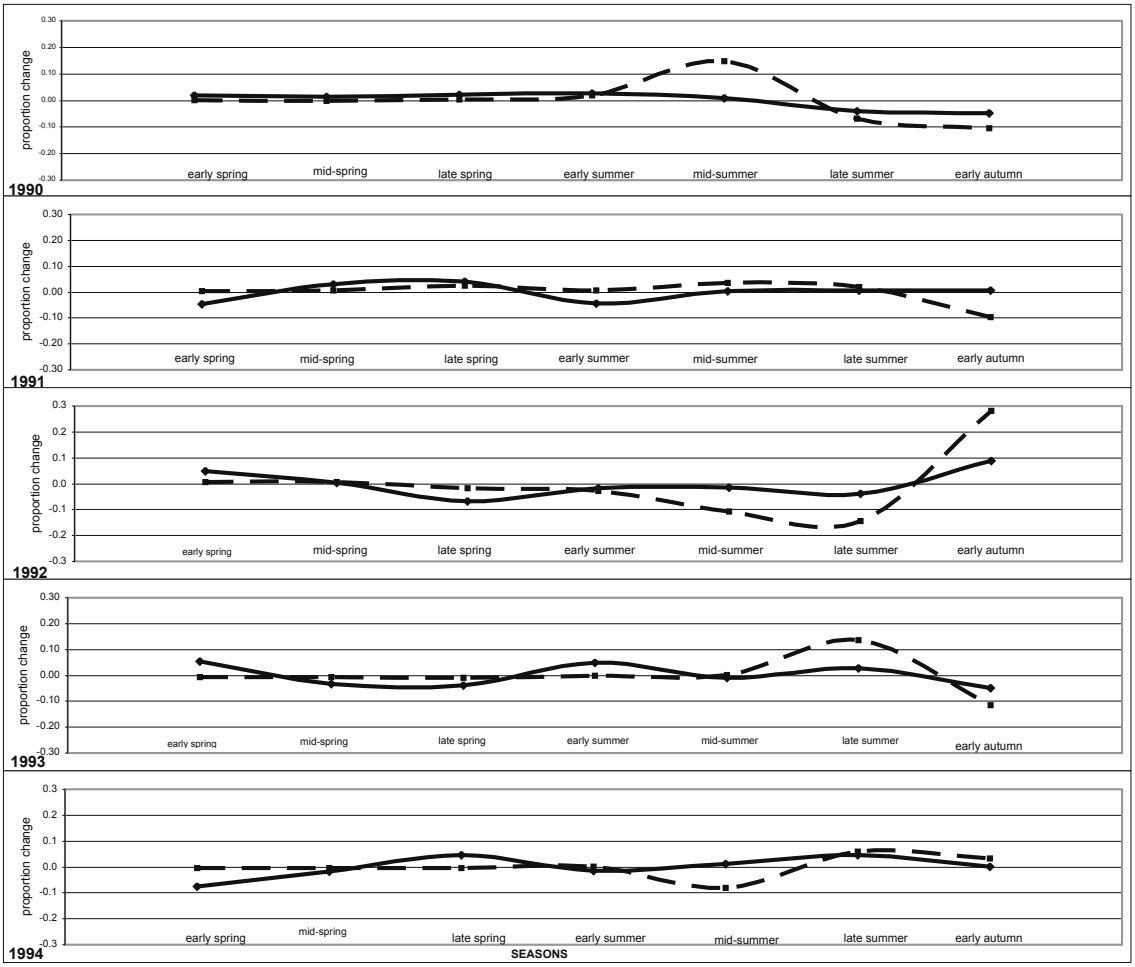

Fig. 2. Annual dynamics of durations and spore totals of seasons in Tartu 1990–1994. Both durations and spore totals are given as their proportions in the pollen season, and presented with respect to the 5-year mean values. Solid line – season duration, dashed line – seasonal spore total.

DISCUSSION

May the results in Tartu 1989–1994 be applied to the last decades of 20th century as a whole? One has to consider two circumstances.

Firstly, in the temperate zone of the Northern hemisphere, spore data collected in one place year by year during three decades on the British Isles (Corden et al., 2003; Millington & Corden, 2005) as well as during one decade on the Scandinavian Peninsula (Hjelmroos, 1993) demonstrate that there is great variation both within the year and between the years in the concentration of airborne spores, and in the spore totals. The extremes of annual spore totals differed 12 times for *Aspergillus / Penicillium* in 1970–2003 in Derby, 10 times for *Alternaria* in 1970–2001 in Derby, 30 times for *Alternaria* in 1970–1996 in Cardiff; in Stockholm 5 times for *Alternaria,* 4 times for *Cladosporium,* and 1.8 times for all spores (1980–1989). Hjelmroos said in 1993: "The investigation period of 10 years is too short a time to demonstrate any long-term fluctuation… That must be largely dependent on long-term climatic fluctuations." Her statement is confirmed by the abovementioned 31-year and 27-year observation series of *Alternaria* – the

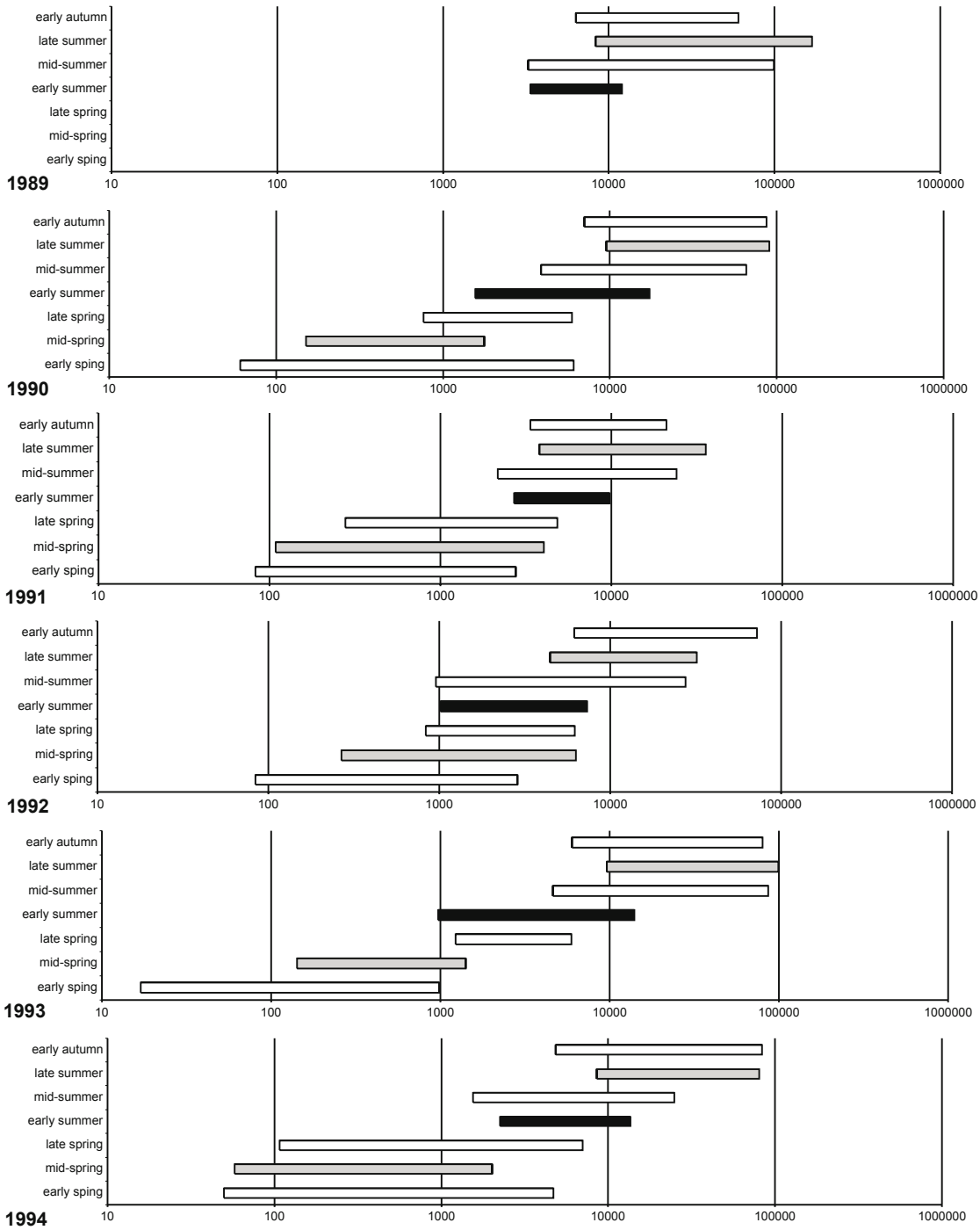

Fig. 3. Seasonal ranges of daily mean spore concentrations in Tartu 1989–1994. Concentration is given as the number of spores in cubic metre of air in 24 hours.

amplitude of variation in annual spore totals in three decades was bigger than that in one decade, respectively 2 and 6 times. Comparing all spores with *Alternaria* and *Cladosporium* spores (observation series in Stockholm), one can see that the annual totals of all spores varied less than the totals of spores of both genera. Such rate seems natural because the composition of all spores formed due to the variety of species with various dependences on temperature, humidity, precipitation and solar radiation; the whole population of atmospheric spores is less dependent on meteorological and other environmental conditions than airborne spores of single genera. Furthermore, the following examples illustrate how spore data could represent or could not represent the fluctuation. In Turku, Finland, the annual total of all spores for 1977 was 1.7 times of that in the previous year (Rantio-Lehtimäki et al., 1985). In Melbourne, in a cool temperate city in the Southern hemisphere, the annual total of all spores for 1992 was nearly 3 times of that in two succeeding years, giving a ratio of 3.2:1.1:1 (Mitakakis et al., 1997). Now, comparing the Tartu results with the British and Scandinavian data described above, we can say: if the extremes of the annual totals varied 2.5 times, then quite a large part of fluctuation that could have occurred in last decades should be overwhelmed by the 6-year data series.

Secondly, discussing the interannual variability, we have to consider the rhythm and length of oscillation in spore data. Spore data collected in one place year by year during three decades (Corden et al., 2003; Millington & Corden, 2005) show irregular oscillation cycles in daily mean concentrations, annual totals as well as in annual peak values. This is natural because in temperate Europe many climatic parameters and phenological phases have most frequently periods of 2.5, 4, 8, 11, 14, 18 and 22 years (Ahas & Aasa, 2003). As seen on Derby and Cardiff data, and on Stockholm data, very many 2-year and 3-year intervals could show only the higher part or only the lower part of the range of spore data. Otherwise, many 6-year intervals include a central part of the range of spore data. It is highly probable when the fluctuation in 6-year spore data is large, as in the case with the data of Tartu.

We can conclude that the years 1989–1994 give spore data that covered a big part of the general interannual variability which occurred in the quantity of the fungal aerosol in Tartu in the last decades of the 20th century. There is no background to estimate, whether the part with lower or higher values of the variation range has been represented.

CONCLUSIONS

Aerobiological calendars with signposts derived from pollen aerosol can be used to describe seasonality of atmospheric fungal aerosol in mid- and high latitudes in regions where winter is long and with snow cover.

In the atmosphere of Tartu, the main spore season lasted 227 days and occurred from March 5 to November 12 as an average of the 6-year period from 1989 to 1994.

The total annual amount of fungal spores occurring in Tartu atmosphere was great in 1989–1994, with PSST (which overwhelmed not the whole but the most part of annual total) varying from 1.21×10^6 to 3.1×10^6 spores m^{-3}.

In the quantities of atmospheric fungal aerosol, the seasonality was expressed clearly in both daily mean concentrations of spores and seasonal-spore-totals. The patterns (as averages of the 6-year period) were as follows in Tartu:

1% of PSST occurred in early spring (lasted 38 days), the concentration range was 10–6000 spores m^{-3} 24 h^{-1},

0.7% – in mid-spring (15 days), 60–6000 spores m^{-3} 24 h^{-1},

3% – in late spring (30 days), 100–7000 spores m^{-3} 24 h^{-1},

4% – in early summer (21 days), 1000–16000 spores m^{-3} 24 h^{-1},

19% – in mid-summer (37 days), 1000–96000 spores m^{-3} 24 h^{-1}.

30% – in late summer (32 days), 4000–160000 spores m^{-3} 24 h^{-1}; the annual peak day of concentration occurred in four of six years during this season;

42% – in early autumn (43 days), 3000–80000 spores m^{-3} 24 h^{-1}; the annual peak of concentration occurred in two of six years.

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