A COMPARATIVE AEROMYCOLOGICAL STUDY OF THE INCIDENCE OF ALLERGENIC SPORES IN OUTDOOR ENVIRONMENT

Nicoleta IANOVICI*, Mălina DUMBRAVĂ-DODOACĂ*, Mărioara Nicoleta FILIMON*, Adrian SINITEAN*

* *West University of Timişoara, Faculty of Chemistry-Biology-Geography, Department of Biology, Romania*
Corresponding author: Nicoleta Ianovici, West University of Timişoara, Faculty of Chemistry-Biology-Geography, Department of Biology, Pestalozzi Street no. 16, Timişoara, Romania, phone: +400256592646; e-mail: nicole_ianovici@yahoo.com

Abstract. The aim of our study was to determine the concentrations of *Cladosporium, Alternaria* and *Stemphylium* conidia in four urban areas in Romania: Braşov, Bucharest, Craiova and Timişoara. The sites differed in habitat characteristics, such as vegetation and microclimate. Airborne spores sampling was carried out in these cities by employing volumetric sampling. The samplings with the Lanzoni sampler were conducted in 2005. Spores were identified at the genus level only. *Alternaria* and *Cladosporium* have been identified as the most abundant and frequent aeroallergens in our cities. The totals of airborne spores recorded in this study show a considerable variation. The higher fungal spore concentrations occur in Bucharest and Craiova. The recorded levels of airborne spores in Bucharest and Craiova were close the concentrations that are accepted as threshold levels for provocation of clinical responses. The diurnal periodicity of aerial conidia showed a peak around midday and low counts in the dark. The present study will contribute to our knowledge of airborne spores in Romania.

Keywords: *Cladosporium, Alternaria, Stemphylium,* airborne fungal spores, aeroallergens, outdoor environment

INTRODUCTION

Fungi exist as saprophytes or as parasites of animals and plants. Fungal spores are important components of bioaerosol in outdoor and indoor environments. Airborne fungal spores originate from soil, plants, and vegetal and animal remains. Therefore the fungal composition of a given area is dependent on its geographical location, meteorological factors, vegetation and human activity. In different studies temperature and humidity have been shown to be factors conditioning their presence in the air. The conidia shape and size which, together with meteorological factors, are the most determining factors for the speed of dispersion and deposition [79]. In wheat harvesting liberated immense numbers of fungal spores into the air many of which are carried long distances in wind currents. By convection, they can reach high altitudes in the troposphere and even up to the mesosphere [40]. Aeromycology, or the study of the biology of airborne fungal propagules, seeks to determine the dispersion, ecology and deposition patterns of phytopathogenic and non-phytopathogenic fungal spores. The capability of producing enormous numbers of spores is characteristic of most moulds [76].

Airspora of several species of fungi are known to induce numerous human diseases such as chronic bronchitis, asthma, rhinitis, allergic bronchopulmonary mycoses, and hypersensitivity pneumonitis [61]. These diseases can result from exposure to spores, vegetative cells, or metabolites. The fungal spores are capable of penetrating the lower airways of the lung and mediate allergic reactions. The conidia and fungal spores associated with immediate type of hypersensitivity are usually larger than 5 μm [15]. The site of deposition of spores also depends on whether spores enter the respiratory tract as individual propagules or as aggregates. More than 80 genera of the major fungal groups have been associated with symptoms of respiratory tract allergy. The most important fungal allergen sources are found among the fungi from the class of the *Deuteromycetes* [38, 41, 42, 44, 45, 51].

Respiratory allergy to fungal spores is estimated at 20 to 30% among atopic individuals and upto 6% in the general population [31, 92]. Fewer people are allergic to fungal spores than to pollen, but as mould spores have been implicated as a major risk factor for fatal asthma attacks in USA [90]. Many authors have been many investigations in various parts of the world to determine the presence and sources of allergenic fungal species and to evaluate their seasonal variations [8, 9, 23, 34, 36, 50, 52, 73, 82, 87]. Most studies on aeroallergens employ volumetric samplers, and the results are mainly collated from microscopic examination of pollen, fungal spores, and hyphae to assess the prevalent aeroallergens in any given environment [2, 20, 55].

Cladosporium Link 1816 is an important component of the airborne population of spores, and is known to be allergenic [62]. Many surveys of the occurrence of *Cladosporium* spores in different regions of the world clearly show their dominance in comparison with other spores. This genus comprises more than 30 species. In recent years, the genus *Cladosporium* has been studied extensively on a morphological and molecular basis [43, 46, 68, 80]. *Cladosporium* spp. are causative agents of skin lesions, keratitis, onychomycosis, sinusitis and pulmonary infections [81]. *Cladosporium sp.* is the common cause of allergenic health effects such as Type I allergies and Type III hypersensitivity pneumonitis [76, 91]. About 60 antigens from *Cladosporium herbarum* have been identified by crossed immunoelectrophoresis and about 36 of them have been shown to be allergenic by crossed radioimmunoelectrophoresis. Three major *Cladosporium herbarum* allergens have been purified and characterized [7, 89]. In Denmark, the weekly clinical symptoms and medication scores were positively correlated with airborne spore counts of *Cladosporium* [65].

Kim *et al.* (1996) consider *Alternaria* as an important fungal antigen [53]. The *Alternaria* genus contains 44 species of which most are plant parasites, but a few species are ubiquitous and are also frequently soil-borne [43, 44, 46]. For over 70 years it has been known that spores of the genus *Alternaria* cause asthma [30, 37, 64, 71]. Currently, the study of *Alternaria* is very interesting both from an aerobiological and from a clinical point of view, due to the infecting and allergenic power of their conidia. It is known to be allergenic and can cause Type I allergies and Type III hypersensitivity pneumonitis. It may also cause nasal and subcutaneous infections in immunocompromised individuals [11, 17, 76]. The prevalence of allergic sensitization to *Alternaria* appears to vary with climate. Nearly 50% of children in Arizona, USA [33], are sensitized to the fungus compared with less than 1% of children in Austria [78]. A relation has been demonstrated between high levels of *Alternaria* spores in ambient air and changes in level of airway hyperresponsiveness in *Alternaria*-sensitized children [25]. Andersson *et al*. (2003) conclude that exposure to *Alternaria* spores may be an important cause of allergic rhinoconjunctivitis [4]. Caretta (1992) describes it as being responsible for most respiratory allergies to fungi [16]. Cosentino *et al.* (1995) point to *Alternaria* as the fungus which produces the highest number of positive skin tests, followed by *Cladosporium*, *Aspergillus* and *Candida* [19]. De Blay *et al.* (1996) show a positive correlation between sensitization to this genus and the symptoms of asthma, eczema, and rhinitis [24]. Licorish *et al.* (1985) suggested that with vertical presentation the spore had a diameter small enough to allow it to easily enter the lungs [64].

In Europe, the interest in the spores of these taxa, as allergenic agent, has been very variable in different countries. In the North of the continent their behaviour is well described, because of the high number of allergy symptoms they produce and their socio-economical consequences [36]. In the Mediterranean region, studies of the seasonal variations of *Alternaria* and *Cladosporium* have increased in the last few years [29, 70]. Most authors consider *Cladosporium* and *Alternaria* to be the most important fungal aeroallergens [6, 13, 16, 17, 22, 49, 77, 85]. Only studies have shown that clinical sensitivity to *Cladosporium* and *Alternaria* can exacerbate asthma [71, 90]. The effect of climate change and associated arable production should not be ignored [56].

Stemphylium species are pathogenic to plants, humans and animals and are distributed throughout the world. More than 20 species are known and four more were recently described [45]. *Stemphylium sp.* aerial behaviour was frequently investigated in urban areas, because of their allergenic effects on humans [38, 45]. *Stemphylium* may cause phaeohyphomycosis in humans. It is responsible for producing Type I allergies [76].

Since knowing the local inhalant allergens facilitates diagnosis and treatment of these pathologies, our objective is to contribute with information of existing mycobiota in atmosphere. In this study the concentrations of airborne spores (*Cladosporium, Alternaria* and *Stemphylium*) from four urban areas in Romania (Braşov, Bucharest, Craiova and Timişoara) were investigated. This is the first comparative study conducted in Romania by volumetric method.

MATERIALS AND METHODS

For this study volumetric method has been employed using VPPS Lanzoni 7 day recording spore traps with a flow rate of 10 L/min. Samplers were located on buildings approximately 15 m above ground level. These traps have an autonomy of 7 days and collect spores continuously with a given absorption flux, enabling daily and even hourly concentration data to be obtained. All slides identified in this study are kept at West University, Department of Biology in Timişoara (Romania). All spore counts were obtained daily at our institution during 15 May to 15 July 2005. The traps worked temporarily for trial "Double-blind, randomized, placebo-controlled, phase III study comparing the efficacy and safety of bilastine 20 mg once daily and cetirizine 10 mg for the treatment of seasonal allergic rhinitis" (MDS Pharma Services funded in Romania). The counting of spores was included in the counting procedure of pollen grains. The trapping surface was removed weekly and dissected for microscopic examination. Slides were covered with glycerine jelly mixed with basic fuchsin. The slides were examined under the light microscope. The specific fungal spores were counted with ×400 magnification. Four equidistant transects across the long axis of the tape were scanned at 4 mm intervals and conidia observed in each 2 h transect were corrected for the proportion of the tape examined and the volume of air sampled: it was then expressed as trapped spores per m^3 of air over 2 h. Daily mean concentrations reflect the approximate numbers of spores found during the day [41]. To calculate trends, a linear regression has been performed: the spore count of records for each taxa was correlated with the increase of time. The intra-diurnal variations recorded during the monitored period have been studied.The value for each hour is calculated by dividing the sum of the values of each hour by the number of days in which airspores were present [3].

The identification and counting of spores were limited to genus levels. Smith (1990) was used as reference book for the identification and description of the fungal spore types [83]. Spores of the genus *Cladosporium* are from 2-12 to 3-21/µm, ovoid to cylindrical or irregular olivaceous-brown to hyaline and non septate or with one to three septa spores. They are smooth or verrucate. They may be collected as simple spores or in pairs but are more frequently seen in groups of four or five spores [43, 46, 68, 80]. *Alternaria* spore type shows great morphological variability. Generally, conidia are muriform, several celled with traverse and longitudinal septa, straight, dark, solitary or in chains of two or few conidia, with smooth to minutely verrucose surface [43, 44, 46].

Stemphylium Wallr. 1833 is the anamorph of *Pleospora* Rabenh. ex Ces. & De Not. 1863. Conidia (12-20 x 15-30 µm) are solitary, light brown to black in color, and rough- or smooth-walled. They are oblong or subspherical and rounded at the tips. These conidia have transverse and vertical septations (= muriform conidia) and there is a typical constriction at the central septum. Some species have one pointed conical apex and the other showing lateral conical protrusions. The walls are smooth, verrucose or echinulate and conidia are often constricted at one or more septa and cicatrized at the base [46, 76].

RESULTS

The aim of the present investigation was to monitor the *Cladosporium, Alternaria* and *Stemphylium* airborne spores and to determine their intra-annual variations in four sites of Romania: Braşov, Bucharest, Craiova and Timişoara. Data provided by the temporary stations were collected in 2005.

Cladosporium type spores were trapped most frequently (except five days in Bucharest and one day in Timişoara).The spores count obtained during the period studied (62 days) was 91,958.8 spores, the lowest value being recorded in Timişoara with 54,055 spores. The highest level of conidia emission was recorded in Bucharest with 148,276. Figure 1 shows the variation of conidial counts corresponding to the curve for each sampling site. The highest concentration of *Cladosporium* spores, equal to 8,153.2/m^3/24 h was noted in Bucharest for 18 June. In Craiova the maximum value of the total *Cladosporium* conidia number was recorded on July 5th – 6,063.9 spores /m^3. The daily mean concentrations of *Cladosporium* spores fluctuated between 871.85 – 2,391.54 spores/m^3. Intradiurnal concentrations of *Cladosporium* spores were lowest between 24:00–10:00 and highest between 12.00-20.00. Intradiurnal concentrations of *Cladosporium* spores were highest in Bucharest (280.81 spores/m^3 - 16:00–18:00) and Craiova (219.76 spores/m^3 - 18:00–20:00) (Fig. 2).

The highest concentration of *Alternaria* spores, equal to 674.9 spores /m^3/24 h was noted in Craiova for 9 July. Daily concentrations were very low in Braşov and Timişoara, only exceeding 20 spores/m^3/24h on a few occasions (Fig. 3). The highest level of conidia emission was recorded in Craiova with 5,700.6. The mean spores count obtained during the period studied was 2,925.4 spores /m^3, the lowest value being recorded in Timişoara with 642.6 spores /m^3. The daily mean concentrations of *Alternaria* spores fluctuated between 10.36-91.95 spores/m^3/2h. Increasing trends are observed at Bucharest, Craiova and Timişoara stations. Daily distribution of airborne *Alternaria* conidia presents a minimum of conidia at 24.00–12.00 hours and a maximum at 14.00–16.00 hours (Braşov, Timişoara) and 18.00-22.00 hours (Craiova, Bucharest) (Fig. 4).

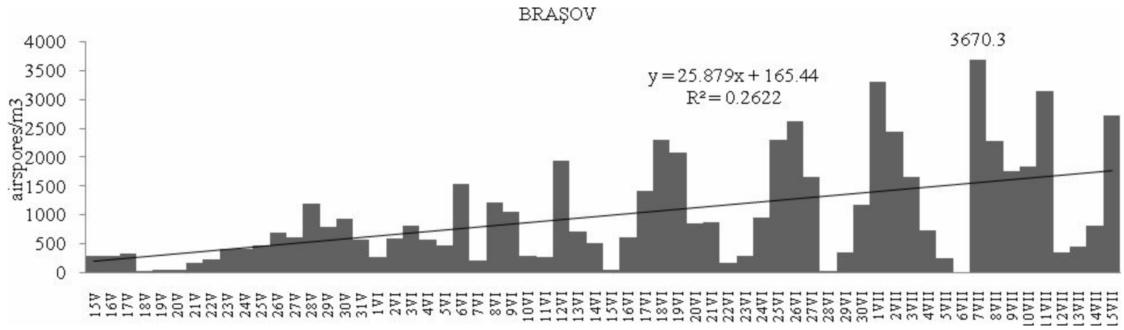
The highest level of *Stemphylium* conidia emission was recorded in Craiova with 763.3, the lowest value being recorded in Braşov with 334.9 spores. The daily mean concentrations of spores fluctuated between 5.4 – 12.31 spores/m^3. The highest concentration of *Stemphylium* spores, equal to 95.2 spores /m^3/24 h was noted in Bucharest for 19 June. Differences between towns concerned total spore counts and numbers of peaks (Fig. 5). Increasing trend is observed at Bucharest station. Decreasing trends are observed at Braşov, Craiova and Timişoara stations. Daily distribution of airborne *Stemphylium* conidia presents a maximum at 14.00–16.00 hours (Braşov, Timişoara) and 18.00-22.00 hours (Craiova, Bucharest) (Fig. 6).

DISCUSSIONS

The occurrence of aeromycoflora in outdoor environment as well as the fluctuations in their numbers and composition is receiving increasingly attention within the framework of potential health hazards to both flora and fauna, including humans. The spores of *Cladosporium, Alternaria* and *Stemphylium* are some of the most abundant allergens world-wide and the prevalence of allergical reactions to them shows great interregional variations [28, 41-47]. The value of this study is to help in the identification of the various components of the outdoor environment. Only a few studies have been carried out in Romania on airborne fungi. Ianovici and Faur (2003) used a volumetric method for the first time in Romania during their investigation of airborne fungi in the city of Timişoara [43]. The monitoring of fungal spores in Timişoara revealed the summer as the most favourable season for *Cladosporium* occurrence. The results in the present study are in agreement with other studies in Timişoara [28, 41, 43, 46, 47]. However, the concentrations never exceeded 3000 spores in previous years.

The threshold concentrations of airborne allergens, for the induction of clinical symptoms, are practically unknown. The rule of the thumb is that a concentration of 3000 spores of *Cladosporium* per m^3 of air are reasonable estimate for the concentrations which may induce clinical symptoms by those taxa. Only few days (9 days in Craiova and 22 days in Bucharest) the concentration of *Cladosporium* exceed the 3000 spores/m^3 of air established by Gravesen as the critical value for symptomatology. Daily concentrations were low in Braşov and Timişoara, only exceeding 3000 spores/m^3 on a few occasions (3 days). The weather in May-July 2005 was dry. The May, June and July were characterised by deficient annual amounts compared to multi-annual averages. The maximal daily concentrations of *Cladosporium* spores which were recorded in the present study, were lower in Braşov and Timişoara, than the suggested critical concentrations. However, the clinical responses of the patients do not depend only on the concentrations of *Cladosporium*. Half of all episodes of increased pulmonary complaints by asthmatic patients coincided with a total fungal spores peak of more than 500 spores/m^3 of air [10]. In all sites the concentration of

Cladosporium spores was high. In our study *Cladosporium* airspores were found to be present regularly. Low humidity explain the higher concentrations in 2005 than in previous years for Timişoara [28, 41, 43, 46, 47].



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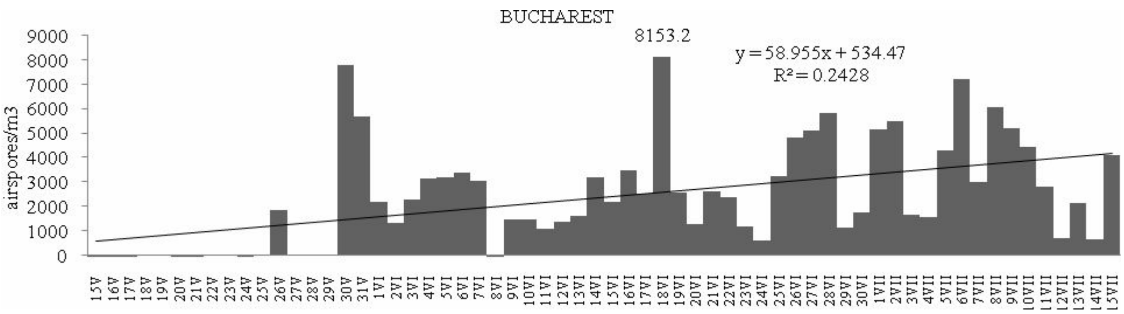

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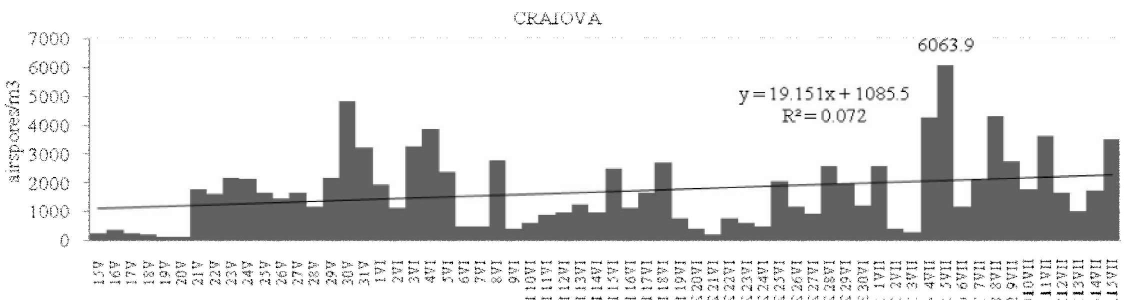

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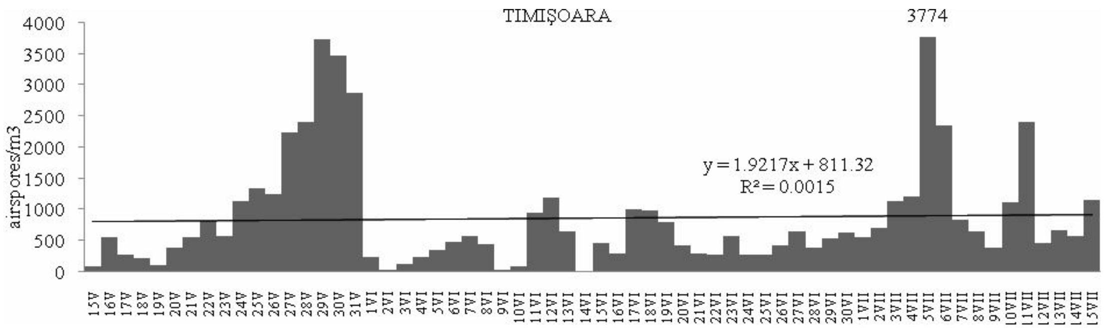


1d

Figure 1 (1a,1b,1c,1d). Daily variations in atmospheric *Cladosporium* spores in the atmosphere of four cities from România, 15V-15VII..

The totals of *Alternaria* spores in the air recorded in this study show a considerable variation. The literature has few references to the number of spores that are needed to provoke an allergic reaction in susceptible individuals [86]. There is considerable variation between those numbers that have been presented. Only few days (18 days in Craiova and 14 days in Bucharest) the concentration exceed the 100 spores/m^3/24h of air established by Gravesen as the critical value for symptomatology [31]. Licorish *et al.* (1985) suggest that of the order of 104–107 spores need to be inhaled over a 24-h period [64]. Hasnain *et al.* (1998) suggest that a mean daily concentration of 50 *Alternaria* spores/m^3/24h are needed to cause sensitisation, after which smaller concentrations can cause symptoms [35]. Frankland and Davies (1965) suggested that when the *Alternaria* count reaches 50 spores/m^3 or above, patients sensitive develop symptoms [30]. The mean number of *Alternaria* conidia isolated in our study was quite similar to that shown by Li and Kendrick in Ontario (61.6 conidia/m^{3}2h) and by Munuera and Carrion in Murcia (31 conidia/m^3/2h) [63, 70]. The intra-diurnal *Alternaria* distribution, with only one daily peak, coincides with that Hjelmroos (1993) detected for Stockholm [36]. The same periodicity was found by Srivastava and Wadhwani (1992) and Fernandez *et al.* (1998) [29, 85].

The totals of *Stemphylium* spores in the air recorded in this study show a considerable variation. Daily concentrations were very low, only exceeding 30 spores/m^3 on a few occasions. Bugiani et al. (2004) introduced the concept of spore peak as a day with more than 30 conidia caught, and considered a peak as an indicator of the potential occurrence of the disease [14]. The behaviour of *Stemphylium* was irregular, with spores concentrations fluctuating considerably. Aerial concentration of conidia showed a series of waves, with periods of spore abundance alternating with periods of spore scarcity [45].

The intra-diurnal variations recorded during the monitored period have been studied. The diurnal periodicity of aerial conidia showed a peak around midday and low counts in the dark [45]. These differences may be due to a different composition of local flora and the influence of weather. The results suggest that temperature is involved in the release of conidia and in the colonisation processes.

Conidial concentrations are known to show a considerable daily variability. Cities have a specific microclimate which can be characterized by, among others, higher daily temperatures and intradiurnal temperature differences and a specific wind direction [52]. The southern cities studied (Bucharest and Craiova) exhibit very similar variation patterns. In contrast, Braşov exhibit the lowest concentrations. Rainfall in Bucharest is 600 mm. Bucharest has a continental moderate climate with hot dry summers, cold winters and an average annual temperature of 10-11ºC while the hottest month of the year is July (23.92°C). Annual mean temperature in Craiova is about 10.8°C while the hottest month of the year is July (22.57°C). Mean annual precipitation is 569.9 mm. The average annual temperature for Timişoara is 10.6°C while the hottest month of the year is July (22.42°C). The annual rainfall is 592 mm. Braşov has a temperate continental climate with cold and relatively wet weather in the mountains. The average temperature of the warmest month of the year, July, is 20.11 in Braşov. Annual rainfall is 1300 mm in the mountains and 600 mm in lower areas. These differences could be due to not only meteorological factors, but also local factors which seem to have a great influence on the distribution of spores in the air. Meteorological conditions may act in several ways on the dynamics of dispersion of particles of biological origin. As well as directly affecting the dispersion itself, they may have an indirect effect by previously influencing the status of the sources. It is probably true that our changing agricultural practices have changed the outdoor air spora over time. Fungal aerosols above forests and grasslands are likely to be considerably different than those above fields of corn,wheat and soybeans [72]. Zureik *et al.* (2002) suggested fungal spores may be inhaled as fragments and other amorphous bio aerosols [39], and the fragmentation of *Alternaria* observed by Corden and Millington (2001) is likely to be increased during combining or mowing, making entry into the lung easy [18]. As *Alternaria* is a saprophyte living on most vegetation, the volumes of dust generated by combine harvesters will include spores which will be dispersed over a wide area. Hyde and Williams were investigating *Alternaria* levels in Cardiff and they postulated that asthma attacks were more likely in the late summer especially near wheat fields or wheat threshing. In the USA the greatest number of *Alter-*

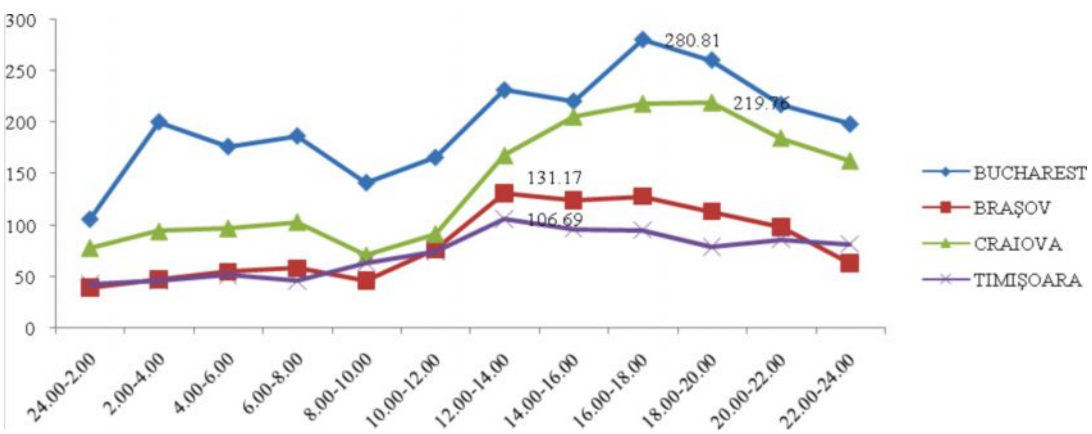
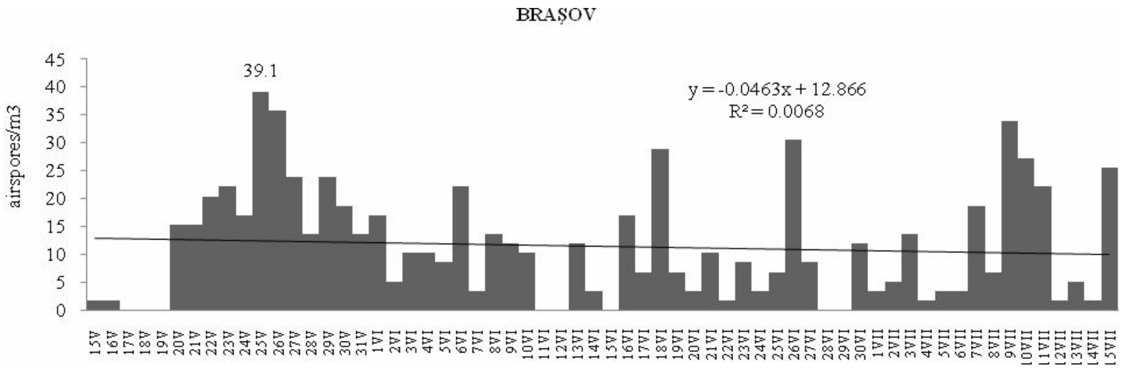


Figure 2. *Cladosporium* intra-diurnal patterns (2-h running mean).

naria spores are found in grain growing areas such as the Midwest [71] and children in Australia had greater *Alternaria* sensitization when they lived in the drier inland regions [74]. These results confirm our data on the city of Braşov. Aerospores concentrations are lower here than in other three cities with different topographic layout, and around which intensive agriculture is practiced. The spores concentrations in the air are increased as a result of agricultural practises such as harvesting of cereal crops.



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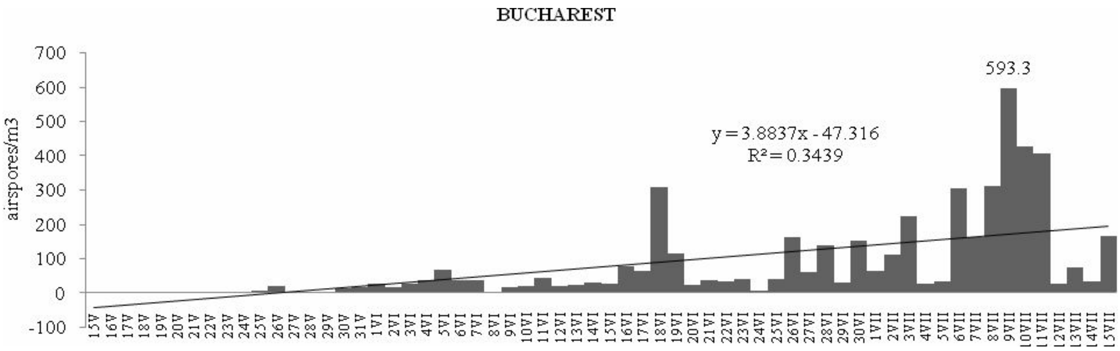

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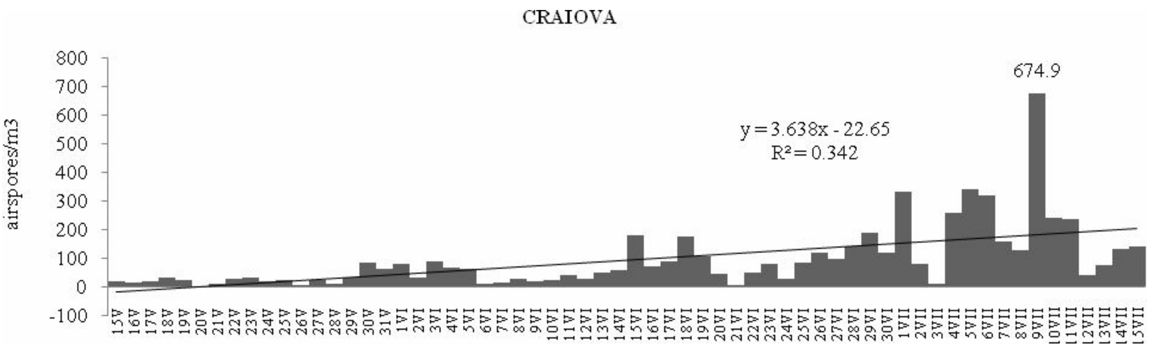

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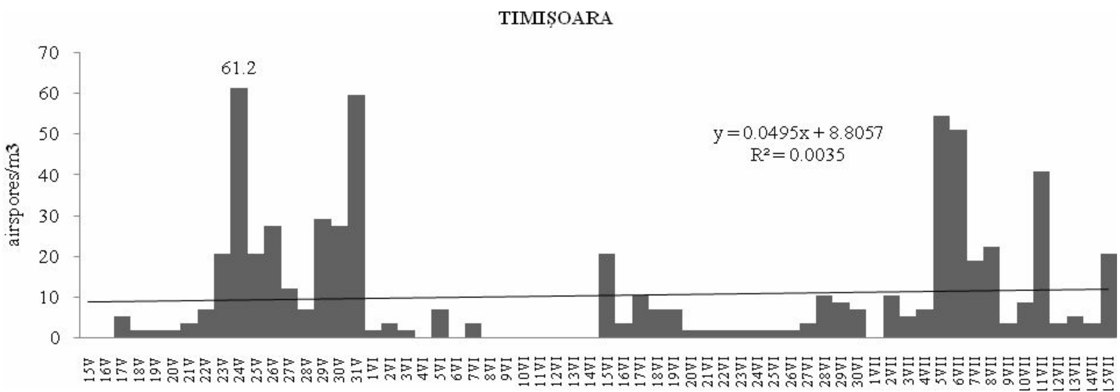

3d

Figure 3(3a, 3b, 3c, 3d). Daily variations in atmospheric *Alternaria* spores in the atmosphere of four cities from Romania, 15V-15VII

Cladosporium is cosmopolitan and colonises a variety of substrates. *Cladosporium* species live as saprophytes or as parasites on many kinds of plants. It is also capable of growing on refrigerated foods. *Cladosporium* species are widespread fungi, including endophytic, fungicolous, phytopathogenic and saprobic species [21, 84]. Saprobic species occur on various senescing and dead leaves and stems of herbaceous and woody plants, are secondary invaders of necrotic leaf spots, and are frequently isolated from air, soil, foodstuffs, paint, textiles and other organic matter [12, 26, 27]. *Cladosporium* belongs to the psychrophilic, xerophilic and xerotolerant group of microorganisms which require low temperature, sometimes below 0°C, with an upper limit about 20°C and relative humidity about 70% for optimal growth. The fungus *Cladosporium* sp. may be regarded as an equivalent to a bio-control fungus, which acts as pathogen to growing leaves of *Parthenium* and provokes their distortion and reduction of photosynthetic area and later control the buds growth and seed development [59].

Alternaria Nees 1817 is a common pathogen of cereals, vegetables and weeds [52]. Species of the *Alternaria* genus are examples of opportunistic plant pathogens that may contaminate a wide variety of crops in the field and cause post-harvest decay of various fruits, grains, and vegetables [32]. They may be responsible for spoilage of these commodities during their refrigerated transport and storage, which not only results in economic losses to growers and commercial marketers, but may also threaten consumers' health [58]. The fungi of this species are able to synthesize and excrete various primary and secondary metabolites, e.g., phytotoxins and mycotoxins [1]. *Alternaria* is largely a soil-borne fungus that lives on rotting organic matter including wood, leaf debris, and grasses[5].

Stemphylium is a genus of plant pathogens and saprobes in the Pleosporaceae [48]. *Stemphylium* causes extended necrotic areas on leaves, shoots and fruits, which are unmarketable. Few works were aimed at studying dynamics of *Stemphylium* conidia in relation to plant disease development, with particular reference to garlic, onion, leeks, lentil, tomato, pepper and pear [54, 57, 60, 67, 75, 88]. In plants, *Stemphylium* spp. have a wide range of hosts including leguminous and non-leguminous crops. *Stemphylium* spp. are also pathogenic on many horticultural cash crops and cause losses up to 100% yield loss in cotton in Brazil [66]. The fungus survives on many different hosts in the eastern and midwestern USA and in Canada. *Stemphylium* is widely distributed on decaying vegetation and in the soil. Most fungi belonging to the genus *Stemphylium* are saprophytes growing on dead plants and cellulose materials. *Stemphylium* species are able to grow as endophytes in the living leaves of various plants [45].

These data show that these fungi are spread in all environments and causes damage not only to human health. This information explains the nature of which high concentrations of conidia in the air. The amount of these fungi is related to the amount of dead vegetation available for it to consume, so its seasonal pattern reflects that of the local vegetation [5]. These fungi have a wide spread in both urban and rural areas. *Cladosporium, Alternaria* and *Stemphylium* fungi grow on plants and therefore the source of exposure is outside, although significant exposure may also occur inside peoples' homes [69].

In this study, *Cladosporium* and *Alternaria* type spores were trapped most frequently and were included as major components in the air. Minor components included *Stemphylium* airborne spores (less frequent and sporadic type) [41]. *Cladosporium* is the leading allergenic mold observed in our climate and is a major source of inhalant allergens. *Alternaria* is one of the most common atmospheric mould spores found in the Romania, the greatest numbers being found in Craiova and Bucharest areas. *Stemphylium* type is common airborne conidia, the greatest concentrations being found in Craiova and Bucharest areas, too. This comparative study shows that for Romania (especially in Bucharest and Craiova) these airborne spores are a potential danger to public health. Testing of allergic patients will have to include these types of fungal spores, especially in summer.

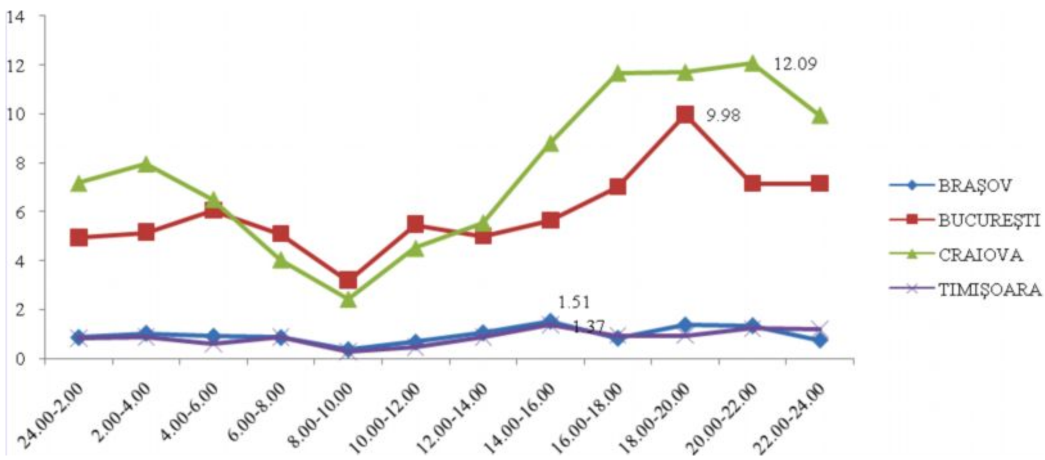

Figure 4. *Alternaria* intra-diurnal patterns (2-h running mean).

The investigated airborne fungal spores are allergenic to fewer people. Therefore, monitoring of this aeroallergens may be useful for patients, clinicians and the general public in the Romania. This study was based on 62 days investigations; additional studies over a longer period are needed to provide a more profound insight into the relationship between spores content in the outdoor environment and allergic manifestations affecting patients. Our present study made an important contribution for determination of levels and types of airborne fungi in Romania. In addition, further studies are needed to be carried out because of urbanization, increasing air pollution and industrialization.

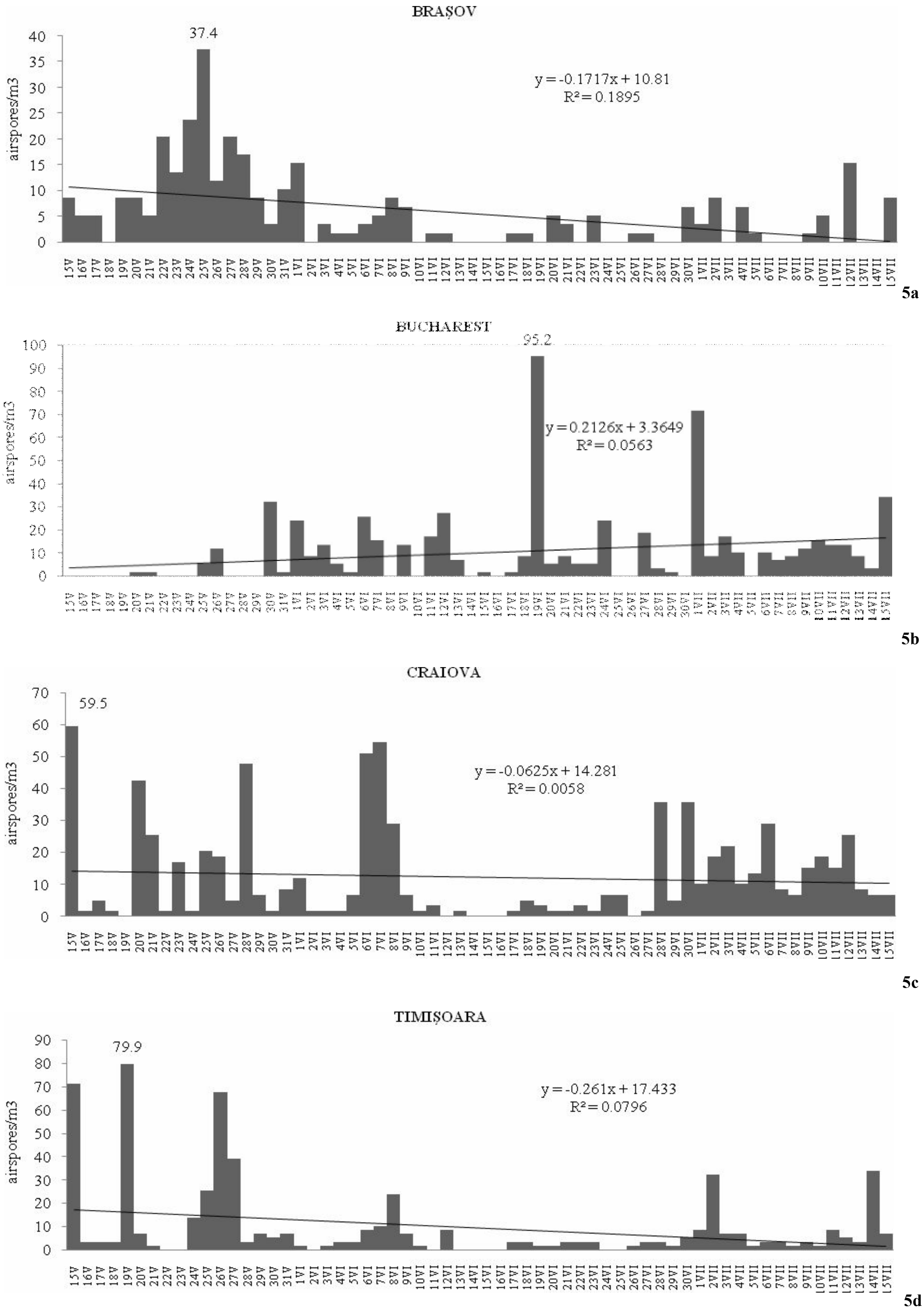

Figure 5 (5a, 5b, 5c, 5d). Daily variations in atmospheric *Stemphylium* spores in the atmosphere of four cities from Romania, 15V-15VII

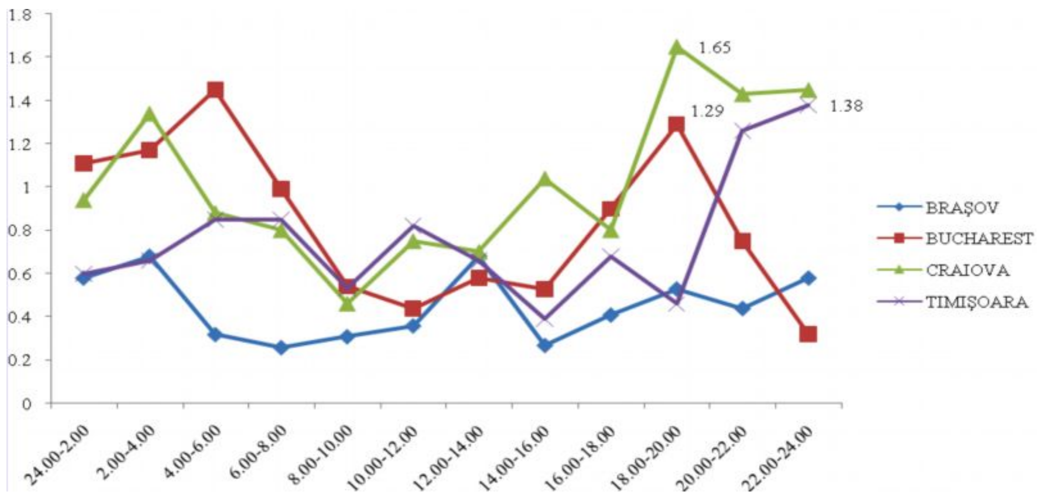

Figure 6. *Stemphylium* intra-diurnal patterns (2-h running mean)

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