A year-round investigation of indoor airborne fungi in Croatia

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This study assessed the composition of aeromycota at a grain mill and four dwellings (two apartments and two basements) as well as in outdoor air during one year in Zagreb, Croatia. The incidence of Aspergilli from sections Flavi, Nigri, and Versicolores was also assessed. Airborne fungi were collected using an air-sampler and DG-18 agar plates. The average concentrations of airborne fungi in the grain mill ranged from 14,310 to 40,000 cfu m\(^{-3}\), which was above the hazardous level (\(10^4\) cfu m\(^{-3}\)), whereas the values statistically estimated using Feller’s correction were up to six times higher. Concentrations in the apartment (163-1244 cfu m\(^{-3}\)) were lower than in outdoor air (286-2090 cfu m\(^{-3}\)) and lower than in the basement (697-1203 cfu m\(^{-3}\)), except in the warmer period of the year when they were similar. The most abundant species throughout the year were Cladosporium spp. (90-100 %), Penicillium spp. (40-100 %), and Alternaria spp. (10-100 %), which are common for temperate climates. Aspergilli from the Flavi (50-100 %) and Nigri (15-40 %) sections as well as A. ochraceus (15-60 %) and Eurotium spp. (85-100 %) were the most abundant at the grain mill and were rarely found in outdoor air. In the basement, Aspergilli (Versicolores) were more abundant than in the apartment. The excess of aeromycroparticles in the grain mill throughout the year may have represented a serious health risk to mill workers. This is the first Croatian one-year study of indoor airborne fungi in a grain mill and dwellings; however, monitoring should continue over a longer period.

KEY WORDS: Flavi; grain mill; Nigri; residential areas; Versicolores

Many studies have recently investigated the levels of airborne fungi in indoor environments and their role in the initiation and/or progression of chronic respiratory diseases like asthma and chronic rhinosinusitis (1-6). In addition, the European Environment Agency (EEA) has published a report on air quality in Europe in 2012 (7), where the most severe health effects were attributed to air pollution by particulate matter (PM). The highest mortality caused by PM clearly stems from particle fractions 2.5 µm in diameter, which represents 40-80 % of PM mass concentration in ambient air in Europe. Fungal spores are regularly found in outdoor and indoor air as part of PM. Their size ranges usually from 1 to 10 µm with variations even among the same species. Besides fungal spores, fragments of mycelium may also be released in the air with sizes even smaller than fungal spores (8-10). Inhalation of these particles represents a threat to human health due to possible deposition in the lower respiratory system and interactions with lung tissue. No uniformness in the suggested guidelines for acceptable levels of fungi in indoor environments has yet been achieved, and there is no dose-response relationship between the concentration of airborne fungi and adverse health effects. Moreover, the immunocompetence of an
exposed individual plays a crucial role in both the type
and intensity of possible effects.

Airborne fungi are of special concern in industrial
environments such as grain mills or agricultural
warehouses, where their concentration may be up to
a hundred thousand times higher than in outdoor air.
It has been suggested that occupational concentrations
of airborne fungi above 10^4 cfu m^(-3) should be
considered a health hazard in non-sensitized subjects,
although this does not mean that such a working
environment will necessarily lead to any respiratory
symptoms (9-13). Considering the limited number of
studies addressed at the fungal burden in occupational
environments and urban homes in our country, the aim
of the present study was to investigate the composition
and concentration of aeromycota in industrial and
residential environments in Zagreb, Croatia, for which
studies addressed at the fungal burden in occupational
period (2012) in two-month intervals at a grain mill
and dwellings over a one-year period in Croatia.

**MATERIALS AND METHODS**

**Sampling and determination of airborne fungi**

Airborne fungi were collected over a one-year
period (2012) in two-month intervals at a grain mill
(GM) situated near Zagreb, Croatia, and in residential
locations which included two apartments (AP) and
two basements (BS). The differences between the
chosen apartments and basements were considered
irrelevant for this study. Samples of outdoor air (ODA)
were also collected. The floor area of the APs was
approximately 70 m^2 each, both inhabited and without
visible mould growth. The BS’s were situated in the
same dwellings as the APs. These were unoccupied
and served as a repository for the inhabitants. Visible
mould growth as well as typical mouldy odour was
observed in both basements. The GM was situated in
an industrial area 40 km outside Zagreb and served
for processing cereals, mainly corn and wheat, as well
for short-term flour storage. It was organised as a
building consisting of a big reception and depository
at the ground floor, milling site located at the first floor,
and sieving site located at the second floor. The air
was quite saturated with grain particles, but no mouldy
odour was observed. Samplings were done in the
middle of the working week in the morning during the
most intensive and constant milling operations at 20
locations, including site of grain/flour exchange, site
of flour storage, site of sieving, and site of milling.
Twenty samples were taken from the APs at locations:
kitchen, dining room, living room, bedroom, and
bathroom. Samples were also taken from 10 locations
in the BS, two at each site. The total number of samples
was 420. Air temperature (°C) and relative humidity (%)
were measured by thermo-hygrometer (Boneco,
Widnau, Switzerland) at every location during each
sampling. Fungi were sampled 1 m above ground
using a Mas 100 Eco air sampler (Merck, Darmstadt,
Germany) with 400 holes (hole to agar impactor) and
dichlorane 18 % glycerol agar (DG-18) plates (25).
The impaction velocity of the sampler was approximately
10.8 m s^(-1) and airflow rate 100 L min^(-1). Because of the
high contamination level, a volume of 10 L (sampling
time 6 s) was chosen for sampling in the GM, while
50 L min^(-1) (sampling time 30 s) was applied at the
other sampling locations. After field sampling, the
plates were incubated for 5 days at 25±2 °C, after
which the developed fungal colonies were counted.
The concentrations of airborne fungi were expressed
as colony forming units (cfu) per volume of air
sampled (expressed in cubic meter), i.e. cfu m^(-3).
Feller’s correction (probable statistical total) was
applied to all of the samples (24). The airborne fungi
were identified on the basis of their macro- and
microscopic characteristics after subculturing on
Czapek, Malt Extract and Potato Dextrose agar (Fluka,
Sigma-Aldrich, Steinheim, Germany), according to the manuals (25, 26). In order to separate the aspergilli of interest, they were isolated on Czapek Yeast Agar [CYA, (26)] and Malt Extract Agar (MEA; Becton Dickinson, MD, USA) and incubated at 25 °C in the dark for seven days (Aspergillus section Flavi and section Nigri) and ten to fourteen days for Aspergillus section Versicolor. The identification of Aspergillus species was conducted according to Pitt and Hocking (25) and de Hoog et al. (26).

Statistics

Concentrations of airborne fungi at each location are represented as mean±SD of cfu m⁻³. For each plate, Feller’s correction (probable statistical total) was applied - as the number of viable particles impinged on a plate increases, the probability of the next particle going into an “empty hole” decreases. The probable number of viable particles calculated from Feller’s formula \(\Pr=N 1/N + 1/N-1 + 1/N-2 + 1/N-r+1\), given by the manufacturer (Merck KgaA, Darmstadt, Germany; \(\Pr=\) probable statistical total; \(r=\) Number of cfu counted; \(N=\) total number of holes in the sampling head).

The Kolmogorov-Smirnov test was used to verify whether the variables were normally distributed. Concentrations of airborne fungi were logarithmically transformed to normalise distribution. When normality was achieved, ANOVA and Tukey multiple comparison tests were used. Otherwise, Kruskal-Wallis test was applied, followed by Dunn’s multiple comparison test. Spearman’s correlation coefficients between average fungal concentrations at each location and ambient parameters (temperature and relative humidity) were also calculated.

RESULTS AND DISCUSSION

Data related to average concentrations of airborne fungi (cfu m⁻³) and average temperature and relative humidity specific for each sampling site and each period of year are presented in Table 1. Levels of aeromycota in the GM (up to 40,000 cfu m⁻³) were significantly higher during the whole year than at the other sampling sites \((p<0.0001)\). Statistically corrected concentrations at the GM were up to 6 times higher than the measured values. Compared to observations from studies on similar occupational environments such as another grain mill [max. concentration 1.7x10⁴ cfu m⁻³ (27)], rice mill [mean 13.71x10⁴-48.42x10⁴ cfu m⁻³; (28)], outdoor agricultural area [mean 72-1796 cfu m⁻³; (29)], agricultural non-point source during wheat harvesting season [mean 10⁵-10⁶ cfu m⁻³; (30)], or sawmills (mean 1700-7300 cfu m⁻³; (13)] our analysis showed an overload with airborne fungi in the GM throughout the entire year. Taking into account statistically estimated values (up to 2.6x10⁶ cfu m⁻³), levels of airborne fungi in the GM were 20 times higher than the occupational concentration of airborne fungi considered hazard to human health (>10,000 cfu m⁻³). Variations of airborne fungi levels in the GM showed no correlation to both temperature and relative humidity (Table 2). This was not surprising because the measured levels were the result of fungal contamination of grains that developed during storage, before delivery to the GM.

Seasonal variations of airborne fungi in the AP and BS did not exhibit a similar pattern. In the BS, airborne fungi exceeded 1000 cfu m⁻³ from May to November while the levels in January, March, September, and November were significantly higher as compared to the AP \((p<0.001)\). Variations of airborne fungi levels in the BS also did not show a correlation to temperature and relative humidity (Table 2). Taken together, these observations suggest a more pronounced fungal contamination in the BS probably due to poor maintenance and ventilation.

Seasonal variations of airborne fungi levels in the AP and ODA showed a similar pattern and positively correlated to temperature changes \((R=0.6661, R=0.5409)\) but not to relative humidity. However, the levels of aeromycota in the AP obtained in January, March, July, and September were significantly lower than in ODA \((p<0.001)\).

The observed concentrations of fungi in ODA (240-2090 cfu m⁻³) were twice as high as the concentrations observed in similar investigation conducted in 2002/03 in Austria [100-1000 cfu m⁻³; (31)]. In that same investigation, apartments without visible mould growth had a maximum of fungal spore concentration of 300 cfu m⁻³. The highest observed concentration in the AP from our investigation was 13 times higher (4000 cfu m⁻³) and more than 16 times higher (5060 cfu m⁻³) in the BS. Our previous year-round investigation, which was also performed in 2002/03, at outdoor air locations in Zagreb (32) showed that airborne fungi peaked in August and September (up to 400 cfu m⁻³), which was five times lower than the levels measured in that same period in this study. Since Zagreb and the Austrian region...
Table 1 Quantitative composition of airborne fungi throughout one year at locations as follows: grain mill (GM), apartments (AP), basements (BS), outdoor air (ODA)

<table>
<thead>
<tr>
<th>Sampling period</th>
<th>GM</th>
<th>Concentration of airborne fungi (cfu m$^{-3}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Min-max*</td>
<td>Absolute values</td>
</tr>
<tr>
<td>January</td>
<td>25630±16440*</td>
<td>4000-40000</td>
</tr>
<tr>
<td>March</td>
<td>38250±7826*</td>
<td>5000-40000</td>
</tr>
<tr>
<td>May</td>
<td>26745±16719*</td>
<td>4100-40000</td>
</tr>
<tr>
<td>July</td>
<td>40000±0*</td>
<td>40000-40000</td>
</tr>
<tr>
<td>September</td>
<td>14310±11222*</td>
<td>7400-40000</td>
</tr>
<tr>
<td>November</td>
<td>6270±2661*</td>
<td>1700-12600</td>
</tr>
<tr>
<td>AP</td>
<td>163±102.7</td>
<td>20-460</td>
</tr>
<tr>
<td>March</td>
<td>182±123.3</td>
<td>40-520</td>
</tr>
<tr>
<td>May</td>
<td>1244±1014</td>
<td>320-40000</td>
</tr>
<tr>
<td>July</td>
<td>997±448.5</td>
<td>140-1660</td>
</tr>
<tr>
<td>September</td>
<td>462±176.9</td>
<td>200-960</td>
</tr>
<tr>
<td>November</td>
<td>223±107.1</td>
<td>40-420</td>
</tr>
<tr>
<td>BS</td>
<td>697±931.4 ab</td>
<td>40-3600</td>
</tr>
<tr>
<td>May</td>
<td>1123±853.6</td>
<td>240-3320</td>
</tr>
<tr>
<td>July</td>
<td>1034±464.4</td>
<td>480-2140</td>
</tr>
<tr>
<td>September</td>
<td>1041±751 ab</td>
<td>240-3800</td>
</tr>
<tr>
<td>November</td>
<td>1203±1444 ab c</td>
<td>180-3060</td>
</tr>
<tr>
<td>ODA</td>
<td>286±66.2 ab</td>
<td>204-440</td>
</tr>
<tr>
<td>March</td>
<td>884±1711 ab</td>
<td>180-5880</td>
</tr>
<tr>
<td>May</td>
<td>1350±550.6 ab</td>
<td>800-2600</td>
</tr>
<tr>
<td>July</td>
<td>2090±235.6 ab</td>
<td>1800-2500</td>
</tr>
<tr>
<td>September</td>
<td>860±250.5 ab</td>
<td>640-1520</td>
</tr>
<tr>
<td>November</td>
<td>240±62.5</td>
<td>120-340</td>
</tr>
</tbody>
</table>

*p<0.0001 GM compared to all other sampling sites; *p<0.0001 BS vs. AP; **p<0.0008 BS vs. AP; ***p<0.0001 ODA vs. AP; **p<0.0008 ODA vs. AP; ***p<0.0001 ODA vs. BS; ***p=0.0465 ODA vs. BS; ***p<0.0001 BS vs. ODA
investigated in the previously mentioned study are in the same climate zone, we can speculate that the higher levels of airborne fungi in ODA and in the AP and BS in this study could be related to current climate changes. However, the sample size and number of locations prevent us from reaching a firm conclusion.

The qualitative composition of airborne fungi at each site and in each sampling period is presented in Table 3. The qualitative composition of airborne fungi in the GM differed from the other sampling locations. The most dominant species in the GM belonged to *Aspergillus* spp. and *Eurotium* spp. (85-100 %). The most prevalent fungal genera in the residential areas were similar to those in ODA: *Cladosporium* spp (45-100 % in AP and 55-95 % in BS), *Penicillium* spp. (85-100 % in AP and 70-100 % in BS), and *Aspergillus* spp. (20-80 % in AP and 50-95 % in BS).

High frequencies of *Aspergillus* spp., *Eurotium* spp., *Cladosporium* spp., *Penicillium* spp. and *Alternaria* spp. at places where people live and work raises concern because all of them have proven to be causative agents for fungal allergies and other respiratory disorders (27, 33). Aspergilli of interest (sections *Flavi*, *Nigri*, and *Versicoloraes*) were frequent only at certain locations. We did not specify any species from these sections because the applied methods were sufficient only for sectional classifications. Due to morphological similarities among species from each section, specific DNA sequence data is required (16, 34-38).

Species belonging to section *Flavi* were very frequent in the GM (50-100 %) and present throughout the year. The sampled plates after incubation sometimes revealed overgrowth by these species, indicating a very high concentration of their viable conidia in the air. *Eurotium* spp. was also dominantly present in the GM (90-100 % samples) throughout the year and their mycelia usually covered the entire plate. Black aspergilli were isolated from GM samples and the peak was observed in May (55 %). As for the other mould genera, *Cladosporium* spp. was frequent in GM samples (60-95 %) followed by *Wallemia* spp. (40-90 %), *Ulocladium* spp. (5-80 %), and *Alternaria* spp. (5-10 %). Our failure to isolate the aforementioned species from samples taken in certain periods may have been due to overgrowth by section *Flavi* and/or *Eurotium* species.

We can also speculate on the observed frequencies of section *Versicoloraes* isolates. These species are known for their very small conidia (around 3 μm) and even if sampling is successful, their growth is too slow which causes them to become overwhelmed by other species. Despite this, we occasionally succeeded in isolating these species from GM samples (5-40 %). *Versicoloraes* species reached their peak in November, which was probably due to the fact that other airborne fungi were at their lowest concentrations. Considering the very high concentration of airborne fungi in the GM, exposed workers are under risk. Moreover, due to the enormous amount of fungi belonging to section *Flavi*, there is the possibility of toxigenic strains and consequently aflatoxin B1 (AFB1). Recently it has been proven that black aspergilli are able to produce fumonisins in high quantities (23). Consequently these mycotoxins might be inhaled through grain and flour dust or via conidia of black aspergilli. Well-known producers of fumonisins belong to the genus *Fusarium* and they are present in high amounts at places where maize is processed and stored (26). Because *Fusarium* spp. conidia are not likely to be airborne, we managed to isolate these species only in September (10 %). However, the presence of *Fusarium* spp. is expected throughout the year and so is the presence of fumonisins in maize.

Significant differences have been observed in the composition of aeromycota in residences compared to ODA. Sometimes, plates taken in the BS were overwhelmed in growth by *Aspergillus* section *Versicoloraes*. This indicated drastic fungal contamination with these species at particular sites, since a frequency of *A. versicolor* above 20 % is considered an indicator of indoor fungal contamination

### Table 2 Correlation between concentration of airborne fungi (cfu m⁻³), temperature, and relative humidity

<table>
<thead>
<tr>
<th>Sampling site</th>
<th>Grain mill</th>
<th>Apartments</th>
<th>Basements</th>
<th>Outdoor air</th>
</tr>
</thead>
<tbody>
<tr>
<td>Parameter</td>
<td>T (°)</td>
<td>rh (%)</td>
<td>T (°)</td>
<td>rh (%)</td>
</tr>
<tr>
<td><em>p</em>-value (two-tailed)</td>
<td>0.8998</td>
<td>0.2336</td>
<td>0.0476</td>
<td>0.8479</td>
</tr>
<tr>
<td>R square</td>
<td>0.0045</td>
<td>0.3295</td>
<td>0.6661*</td>
<td>0.01035</td>
</tr>
</tbody>
</table>

*T-temperature
rh-relative humidity
*<p><0.05 significant positive correlation
Table 3 Qualitative composition of airborne fungi at selected locations

<table>
<thead>
<tr>
<th>Airborne fungi</th>
<th>January (%)</th>
<th>March (%)</th>
<th>May (%)</th>
<th>July (%)</th>
<th>September (%)</th>
<th>November (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Absidia spp.</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Alternaria spp.</td>
<td>10</td>
<td>15</td>
<td>25</td>
<td>20</td>
<td>10</td>
<td>0</td>
</tr>
<tr>
<td>Aspergillus ochraceus</td>
<td>40</td>
<td>0</td>
<td>20</td>
<td>0</td>
<td>15</td>
<td>0</td>
</tr>
<tr>
<td>Section Flavi</td>
<td>80</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>50</td>
<td>0</td>
</tr>
<tr>
<td>Section Nigri</td>
<td>40</td>
<td>6.2</td>
<td>5.2</td>
<td>0</td>
<td>15</td>
<td>10</td>
</tr>
<tr>
<td>Section Versicolores</td>
<td>0</td>
<td>45</td>
<td>60</td>
<td>0</td>
<td>5</td>
<td>40</td>
</tr>
<tr>
<td>Aspergillus terreus</td>
<td>100</td>
<td>80</td>
<td>95</td>
<td>90</td>
<td>95</td>
<td>45</td>
</tr>
<tr>
<td>Aureobasidium spp.</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>5</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Botrytis spp.</td>
<td>0</td>
<td>0</td>
<td>5</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Chrysonilia sitophila</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>5</td>
</tr>
<tr>
<td>Cladosporium spp.</td>
<td>60</td>
<td>45</td>
<td>55</td>
<td>100</td>
<td>55</td>
<td>65</td>
</tr>
<tr>
<td>Culvularia spp.</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Epicoccum spp.</td>
<td>10</td>
<td>0</td>
<td>5</td>
<td>0</td>
<td>40</td>
<td>5</td>
</tr>
<tr>
<td>Eurotium spp.</td>
<td>90</td>
<td>60</td>
<td>40</td>
<td>50</td>
<td>100</td>
<td>20</td>
</tr>
<tr>
<td>Fusarium spp.</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Mucor spp.</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Penicillium spp.</td>
<td>100</td>
<td>90</td>
<td>85</td>
<td>90</td>
<td>95</td>
<td>100</td>
</tr>
<tr>
<td>Rhizopus stolonifer</td>
<td>10</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Ulocladium spp.</td>
<td>0</td>
<td>20</td>
<td>50</td>
<td>0</td>
<td>5</td>
<td>0</td>
</tr>
<tr>
<td>Wallemia spp.</td>
<td>0</td>
<td>30</td>
<td>30</td>
<td>10</td>
<td>60</td>
<td>5</td>
</tr>
</tbody>
</table>

1 - grain mill, 2 - apartment, 3 - basement, 4 - outdoor air
(21). One of the mould sources in the air of indoor environments is damp building material. Aspergillus species are often found on substrates such as walls of residences and working interiors, where their survival depends only on water activity (a_w). Aspergilli are primary colonizers and can be customized to a_w values below 0.8, which allows them to succeed in damp indoor environments more likely than other moulds (22, 39). Aspergillus versicolor is the most frequently reported fungal species in section Versicolores from damp indoor environments and its presence might be related to Sick Building Syndrome (17-20). Recent publications based on the molecular identification of Aspergillus species (Versicolores) indicated that Aspergillus versicolor sensu stricto were not common in buildings. However, it has also been reported that Aspergilli from this section isolated from indoor air are highly diverse in temperate regions (36), so we cannot exclude the presence of A. versicolor sensu stricto in our isolates. Considering the variety of recently identified Aspergillus species from this section (36), it is likely that more than one was able to produce sterigmatocystin. Therefore, their presence in high concentrations in indoor air could represent a serious threat to human health. Samples from the AP did not indicate overgrowth by these species, although their presence was persistent. Relative humidity measured at the AP in periods when indoor spaces are heated (January, November) ranged from 41 to 49 % and a high rate of condensation on windows was observed. In that same period, the ambient temperature was 18.3-18.6 °C and these conditions were considered supportive for sporulation and mycotoxin production by A. versicolor (40).

CONCLUSIONS

Considering the observed and analysed data, there is a threat of chronic exposure to secondary metabolites (sterigmatocystin, fumonisins, and aflatoxin). These metabolites can accumulate in spores and mycelium fragments in unknown concentrations. When people are exposed to high concentrations of mycotoxins, especially from contaminated food, one can reasonably easily establish a connection with a certain disease. However, when they are present in very low concentrations, it is very difficult to study and evaluate their effects. Moreover, there are many unknown facts about these effects when inhalation is the primary means of exposure. However, there is a remarkable amount of studies proving toxic, mutagenic, and carcinogenic properties of mould secondary metabolites on respiratory surrogate tissues, cell lines, and animal respiratory system, as well as case-reports addressing adverse health effects in humans (4, 41-46). Therefore, monitoring of airborne fungi in occupational as well as living environments is of great importance in the prevention of unfavourable effects on human health.

Acknowledgements

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Conflict of interest

The authors declare that they have no conflict of interest.

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Sažetak

Jednogodišnje istraživanje razine plijesni u zraku unutarnjih prostora

Cilj rada bio je ispitati varijacije učestalosti (%) i koncentracija (cfu m⁻³) plijesni u zraku mlina žitarica i četiri stambeni prostora (dva stana i dva podruma) u odnosu na vanjski zrak tijekom jedne godine u Zagrebu (Hrvatska). Učestalost *Aspergillus* vrsta iz sekcija *Flavi*, *Nigri* i *Versicolores* također je ispitan. Plijesni iz zraka su uzorkovane pomoću uređaja Air-sampler MAS 100 Eco u kojem su postavljene DG-18-agarske ploče. Prosječna koncentracija aerogenih plijesni u mlinu bila je između 14.310 i 40.000 cfu m⁻³, što je veće od koncentracije (10⁴ cfu m⁻³) koja se smatra opasnom za zdravlje. Procijenjene vrijednosti koncentracija plijesni u zraku mlina, dobivene Felerovom korekcijom, čak su šest puta veće od izmjerenih koncentracija. U stanovima (163-1244 cfu m⁻³) koncentracije aerogenih plijesni bile su manje nego u vanjskom zraku (286-2090 cfu m⁻³) i podrumima (697-1203 cfu m⁻³), izuzev u toplijim mjesecima kada su izmjerene vrijednosti bile slične. Tijekom godine dominirale su vrste iz rodova *Cladosporium* spp. (90-100 %), *Penicillium* spp. (40-100 %), i *Alternaria* spp. (10-100 %), koje su uobičajene u područjima s umjerenom klimom. Aspergile iz sekcija *Flavi* (50-100 %) i *Nigri* (15-40 %), *A. ochraceus* (15-60 %) i *Eurotium* spp. (85-100 %) dominirali su u zraku mlina, a u uzorcima vanjskog zraka rijetko su detektirane. Vrste *Aspergillus* iz sekcije *Versicolores* s većom učestalošću nađene u podrumima nego u stanovima. Veliko opterećenje mlina aerogenim pljesnima tijekom cijele godine može biti opasno za zdravlje radnika. Ujedno, ovo je prvo jednogodišnje ispitivanje sezonske varijabilnosti u sastavu plijesni u zraku mlina žitarica i stambenih prostora u Hrvatskoj. Međutim, monitoring treba nastaviti tijekom dužeg razdoblja.

KLJUČNE RIJEČI: Flavi; mlin; Nigri; stambeni objekti; Versicolores

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