AIR-CONDITIONING AND MICROBIOLOGICAL ENVIRONMENT IN THE LECTURE ROOM

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The influence of air-conditioning system and equipment on the indoor environmental conditions inside the lecture room of the Czech University of Life Sciences Prague is described. The microbial contamination (filamentous fungi and bacteria) and also the main microclimatic parameters of the air, temperature and relative humidity of air, were measured and evaluated in relation to different performance conditions of the lecture room. Predominant bacteria detected were Gram-positive cocci, *Micrococcus spp.*, *Staphylococcus spp.,* and endospore-forming bacilli (*Bacillus spp.*, *Bacillus mycoides*). The intensity of air-conditioning influenced the quality of indoor environment in the room. 15 min after starting the air-conditioning the number of airborne bacteria in the lecture room decreased to 35–40 and later on to 20. Conidia of microscopic filamentous fungi were passing through the filters for outdoor air filtering (non-woven polyester fabrics Firon Special G 460 for coarse dust, class filtration G 3). Allergenic *Cladosporium* was dominant and after starting the air conditioning its occurrence in the room increased, no matter if the filters were clean or dusty. The highest values of 555 CFU.m^{-3} were measured in summer after 5 h of air conditioning. Contamination by allergenic *Aspergillus* increased sixteen times after 15 min of air-conditioning performance with dusty air filters.

indoor air quality; airborne yeast; filamentous fungi; bacteria

INTRODUCTION

The influence of indoor climate on human health has been emphasized in numerous scientific and professional papers (Fisk et al., 2007; Mudarri, Fisk, 2007). Some studies dealing with the importance of a suitable microclimate in offices (Seppanen et al., 2006) and at schools (Karwowska, 2003; Grisoli et al., 2012) pointed to the impact of the diseases incidence on the regularity of school or office attendance. Inappropriate microclimate may contribute to a higher sickness rate, and thereby may increase the number of days spent out of office or school (Mudarri, Fisk, 2007). In the study on indoor air microbiological contamination in various rooms of university buildings, multiple growth of bacteria and significant increase of mould spores was observed in the afternoons (Stryjakowska-Sekulska et al., 2007).

Awat et al. (2013) mentioned that the outdoor fungal concentration depends on the locality and season. According to their results fungal concentrations were significantly higher in the rural than in the urban environment. The average indoor and outdoor total fungal concentrations were 608 and 675 CFU.m^{-3} in the urban environment and 1932 and 1872 CFU.m^{-3} in the rural environment, respectively. The greatest concentrations were found in the autumn and spring season. Indoor and outdoor concentrations were significantly correlated ($P < 0.001$).

Recent years results (not yet published) of microbial air quality measurements in the University campus and its surroundings have shown that the outdoor air concentration of bacteria was in the range of 110–1010 CFU.m^{-3} and of yeast and filamentous fungi 170–3890 CFU.m^{-3}, positively depending on air temperature, wind gusts, and amount of necrotic aboveground parts of plants at the place of the measurement, and negatively depending on relative humidity of air.

The objective of the present paper was to show the true microbiological situation in the lecture room equipped with the air-conditioning (AC) system. Microbiological contamination was monitored at different operating conditions inside the room. As the inlet of the described AC system is situated in a quiet place, without traffic and any special source of pollution, no special attention was paid to the problems of outdoor air pollution.

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MATERIAL AND METHODS

The present research was carried out in the lecture room M II of the Faculty of Engineering, Czech University of Life Sciences Prague. The lecture room has the capacity of 121 student places arranged in 11 rows of desks. The room is stepped and has the following dimensions: length 12.2 m; width from 7.7 m in the lowest point (at the presenter' place at the blackboard) to 9.1 m at the uppermost situated entrance place; height of the ceiling in the highest point (by the entrance) 3.2 m and in the lowest (by the blackboard) 4.1 m. The volume of the room is ca.374 m^3. The actual layout of the room is shown in Fig. 1.

The air-condition (AC) equipment is modular and consists of inlet section and outlet section. The inlet part consists of the silencers, filter, valve chamber, heat exchangers (heater and cooler), and a fan connected to the silencer. The direct cooler is connected to the condensation cooling unit, installed on the roof of the auditorium. For humidification, a steam humidifier connected to the drinking water pipeline is used. The outlet part consists of the silencer, filter, valve, and fan. A photo of a special chamber where main part of the AC equipment is situated is in Fig. 2.

The AC system was designed for a fresh air inflow of 4520 $m^3.h^{-1}$ and air outflow of 4083 $m^3.h^{-1}$ (small overpressure). The ventilation rate for one person is ca. 37 $m^3.h^{-1}$ when the room is fully occupied, the intensity of room ventilation being ca. 12 h^{-1}. Between the inlet and the outlet section a rotary regenerative heat exchanger for heat recovery from the exhaust air is installed. The regenerator is an industrial product in which the matrix (heat transfer surface) is in a disk form and the air flows axially.

Fresh and exhaust air is filtered through the pleated filters made of non-woven polyester fabrics Firon Special G 460 (KS Klima-Service a.s., Dobříš, Czech Republic). This material corresponds to the requirements given in the International classification of air filters (CSN EN 779), filtration class G 3 for coarse dust. According to previous information and old international standards, this material should be used for elimination of particles sizing over 10 μm. Basic data on properties of these filters meeting the new international standard are summarized in Table 1. A polluted filter of outside air filtration is shown in Fig. 3.

The complete AC equipment is installed on the supporting frame. Air supply to the auditorium comes via pipes with silencers and inlet ceiling diffusers, built uniformly in the room. Air outlet is ended by two wall grilles situated at the corner of the room above the blackboard (see Fig. 1). Commonly the AC works automatically in dependence on the internal temperature, but during the measurement it was operated manually so as to suit the intended experiments.

Measuring points were situated on the central axis of the lecture room, at three different levels (2^{nd}, 6^{th}, 11^{th} row of school desks), between the air inlets. Measurements included determination of microbial contamination of indoor air in the auditorium during different operating modes of the AC system.

The air samples for microbiological analyses were taken using the microbial air sampler Merck Mas-100 Eco (Merck Group KGaA., Darmstadt, Germany). Air volume of 0.200 m^3 was captured at the height

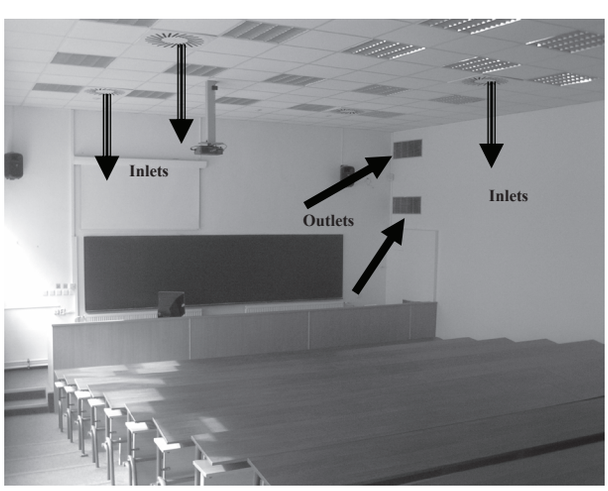

Fig. 1. The lecture room M II with inlets and outlets

Fig. 2. Chamber with air-conditioning equipment

Table 1. Level of separation of filter classes G 3 for filtration of coarse dust

Filter type	EN 779	Average Arrestance (%) (Ashrae Dust)	Final pressure drop (Pa)
Coarse filter	G3	80 ≤ Am < 90	250

Table 2. Measurement in the empty lecture room, air condition (AC) with polluted filters

Time of measurement	Chronological order (h)	AC regime
30 min before	0	OFF
AC START	0.5	ON = START
15 min after	0.75	ON
1 h after	1.5	ON
5 h after	5.5	ON

of 0.8 m above the floor, and cultivated using potato-dextrose agar OXOID CM 139 (PDA; 100%) and nutrient agar OXOID CM 003 (NA; 100%). Colonies of microorganisms were evaluated after 5 days of incubation. Yeast and filamentous fungi were cultivated at 22°C, bacteria at 29°C. Captured microorganisms, which formed colonies on the culture media, were counted and expressed as colony forming units per m^3 ($CFU.m^{-3}$). Dominant genera were determined using the microscope.

Simultaneously with the collection of air samples, also temperature and humidity were measured in the monitored space using a sensor FHA 646 (Ahlborn Mess- und Regelungstechnik GmbH., Holzkirchen, Germany) and a data logger ALMEMO 2690-8 (Ahlborn Mess- und Regelungstechnik GmbH., Holzkirchen, Germany).

The measurements were carried out during the summer holidays (no students inside), and at the beginning of the autumn semester (room full of students) under normal operating conditions with running air-cooling so as to create suitable thermal conditions, in both cases with the AC switched on and off.

Fig. 3. Polluted filter of outside air filtration

The effect of the filter purity on microbial contamination of indoor air was also checked. The measurement scheme description of the research procedure and conditions inside the auditorium during the measurements are summarized in Tables 2 and 3.

Statistical analyses were computed by STATGRAPHICS Centurion XV software (Version 15, 2009), using One-Way Analysis of Variance (Multiple range tests); Scheffé's test ($P \leq 0.01$).

RESULTS

Principal results of microclimate measurement and microbiological evaluation for different AC levels are summarized in Tables 4–9. Tables 4, 5 contain the results of microbiological contamination and air temperature and relative humidity measurements in the lecture room without students. The filters in the AC device were polluted, having been in use for several months prior to the experiment.

Results of the measurements (microbiological contamination, air temperature, relative humidity) in the lecture room with students are given in Tables 6, 7. New filters in the AC device were completely clean.

The occurrence of yeast and filamentous fungi in the lecture room for different AC levels are summarized in Tables 8, 9.

Table 8 contains the results on yeast and filamentous fungi concentration in the empty lecture room (summer). The filters of the AC device were polluted, having been in use for several months prior to the experiment.

The occurrence of the yeast and filamentous fungi in the lecture room full of students (early autumn) is summarized in Table 9. Clean filters in the AC device were completely new.

DISCUSSION

The average concentration of bacteria in the empty room was 207 $CFU.m^{-3}$. 15 min after switching the AC on, the number of airborne bacteria in the lecture room decreased to 75 $CFU.m^{-3}$. But later on, after 5 h of AC running, it attained to 188 $CFU.m^{-3}$. It was probably due to the gradual release of bacteria from the contaminated filters.

Indoor air quality in the classroom full of students with AC out of operation was bad, leading to growing of temperature (from 24.3 to 26.3°C) and relative

Table 3. Measurement in the occupied lecture room, air condition (AC) with clean filters

Time of measurement	Chronological order (h)	AC regime	Number of persons
105 min before	0	OFF	35
15 min before	1.5	OFF	56
AC START	1.75	ON = START	56
15 min after	2.0	ON	56
5 h after	5.5	ON	18

Table 4. Average number, median, and range of bacteria, average temperature (T) and average relative humidity of air (Rh) in the empty lecture room, air condition (AC) with polluted filters

Time of measurement	Average (CFU.m^{-3})	Median (CFU.m^{-3})	Range (CFU.m^{-3})	T (°C)	Rh (%)
30 min before AC	207 ± 28^{a}	225	165–230	23.3	39.4
15 min after AC	75 ± 17^{a}	75	50–100	22.2	43.0
1 h after AC	100 ± 13^{a}	95	85–120	21.8	42.8
5 h after AC	188 ± 38^{a}	200	130–235	21.8	38.8

$^{a–c}$highly significant difference (ANOVA; Scheffé's test; $P \leq 0.01$)

Table 5. Average number, median, and range of yeast and filamentous fungi, average temperature (T) and average relative humidity of air (Rh) in the empty lecture room, air condition (AC) with polluted filters

Time of measurement	Average (CFU.m^{-3})	Median (CFU.m^{-3})	Range (CFU.m^{-3})	T (°C)	Rh (%)
30 min before AC	408 ± 19^{a,b}	410	380–435	23.3	39.4
15 min after AC	372 ± 54^{a,b}	405	290–420	22.2	43.0
1 h after AC	328 ± 42^{a}	345	265–375	21.8	42.8
5 h after AC	633 ± 84^{b}	585	555–760	21.8	38.8

$^{a–c}$highly significant difference (ANOVA; Scheffé's test; $P \leq 0.01$)

humidity (from 46.2 to 54.5%), as metabolic products (energy, vapour etc.) were not ventilated out. Also contamination of indoor air by bacteria, when the AC in the lecture room was out of order, was rising with the number of the present students, from 695 to 1330 CFU.m^{-3}. The highest concentration of bacteria was detected in the 11th row; the source of the bacterial pollution were the students themselves, a majority of them were sitting in the upper part of the lecture room. There was a significant correlation between temperature and concentration of bacteria ($R^2 = 0.727$), but the changes of indoor air parameters were due to the changing number of students and air-conditioning intensity. Correlation between relative humidity and bacterial concentration was not significant. Gorny, Dutkiewicz (2002) reported the normal bacterial concentration range of 88–4297 CFU.m^{-3}. Our results are in accordance with these authors also as concerns the representation of the bacterial species. Gram-positive cocci, *Micrococcus spp*., *Staphylococcus spp*. and endospore-forming bacilli (*Bacillus spp*., *Bacillus mycoides*) were dominant. 15 min after switching on the AC the number of airborne bacteria in the lecture room decreased to 35–40% and later on even to 20%.

When the room was empty and the AC out of operation, the average concentration of yeast was about 55 CFU.m^{-3}, 15 min after switching on the AC the number of airborne yeast decreased to 18–26%, and later on to 5–7%. The highest concentrations of yeast were detected before turning on the AC in the classroom with students. Not all the present genera could be identified, but *Debaryomyces*, *Candida*, *Saccharomyces*, *Rhodotorula*, *Trichosporon*, *Sporidiobolus,* and *Cryptococcus* were the most common. AC reduced their concentration. It is obvious that people are the main source of yeast pollution. Correlations between indoor parameters and yeast and filamentous fungi were not significant.

The average concentration of conidia of filamentous fungi was the same before as well as after air conditioning, but in the genera *Aspergillus* and *Cladosporium* the growth occurred only after turning the AC on. We assume that the increase in the genus *Cladosporium* was due to the number of conidia sucked in from the outdoor air through the filters. Concentration of other filamentous fungi (e.g. *Aspergillus*, *Acremonium*, *Penicillium*) was on the same level or was slightly increasing. In the case of the genus *Aspergillus* the growth was probably caused by dirty filters; due to AC the air get purified and the concentration of conidia gradually lowered.

Conidia of microscopic filamentous fungi (*Cladosporium*, *Aspergillus*, *Penicillium*, *Acremonium*, *Alternaria*, *Epicoccum*, *Fusarium*, *Curvularia*,

Table 6. Average number, median, and range of bacteria, average temperature (T) and average relative humidity of air (Rh) in the occupied lecture room, air condition (AC) with clean filters

Time of measurement	Average (CFU.m^{-3})	Median (CFU.m^{-3})	Range (CFU.m^{-3})	T (°C)	Rh (%)
105 min before AC	830 ± 37^{b}	855	775–860	24.3	46.2
15 min before AC	938 ± 261^{b}	790	695–1330	26.3	54.5
15 min after AC	405 ± 40^{a,b}	405	345–465	24.2	37.9
5 h after AC	210 ± 77^{a}	190	115–325	21.9	31.0

$^{a–c}$highly significant difference (ANOVA; Scheffé's test; $P \leq 0.01$)

Table 7. Average number, median, and range of yeast and filamentous fungi, average temperature (T) and average relative humidity of air (Rh) in the occupied lecture room, air condition (AC) with clean filters

Time of measurement	Average (CFU.m^{-3})	Median (CFU.m^{-3})	Range (CFU.m^{-3})	T (°C)	Rh (%)
105 min before AC	448 ± 22^{a,b}	460	415–470	24.3	46.2
15 min before AC	263 ± 9^{a}	265	250–275	26.3	54.5
15 min after AC	328 ± 6^{a}	330	320–335	24.2	37.9
5 h after AC	320 ± 63^{a}	295	250–415	21.9	31.0

$^{a–c}$highly significant difference (ANOVA; Scheffé's test; $P \leq 0.01$)

Table 8. Occurrence of yeast and filamentous fungi in empty lecture room, air condition (AC) with polluted filters

Time of measurement	Average occurrence (CFU.m^{-3})			
	yeast	filamentous fungi		
		Cladosporium	*Aspergillus*	*Acremonium, Penicillium* and others
30 min before AC	55^{a,b}	293^{b}	3^{a}	60^{a}
15 min after AC	10^{a}	287^{b}	47^{a}	75^{a}
1 h after AC	3^{a}	227^{a,b}	30^{a}	98^{a}
5 h after AC	8^{a}	555^{c}	3^{a}	70^{a}

$^{a–c}$highly significant difference (ANOVA; Scheffé's test; $P \leq 0.01$)

Table 9. Occurrence of yeast and filamentous fungi in the occupied lecture room, air condition (AC) with clean filters

Time of measurement	Average occurrence (CFU.m^{-3})			
	yeast	filamentous fungi		
		Cladosporium	*Aspergillus*	*Acremonium, Penicillium* and others
105 min before AC	355^{c}	38^{a}	8^{a}	55^{a}
15 min before AC	147^{b}	57^{a}	2^{a}	58^{a}
15 min after AC	38^{a}	92^{a,b}	3^{a}	198^{a}
5 h after AC	25^{a}	115^{a,b}	2^{a}	180^{a}

$^{a–c}$highly significant difference (ANOVA; Scheffé's test; $P \leq 0.01$)

Drechslera, *Ulocladium,* etc.) are passing through the filters (G3, G4) for filtering coarse dust and their source can be the air taken in from the outside environment (outdoor air).

Allergenic *Cladosporium* was dominant (its share among all captured filamentous fungi and yeasts made 8–88%). This dominance increased after switching on the AC, no matter if the filters were clean or polluted. The highest average value of 555 CFU.m^{-3} was measured in the empty room after 5 h of AC, and 115 CFU.m^{-3} in the room with students after 3 h and 45 min of AC. Basilico et al. (2007) confirmed the 58.9% dominance of this genus.

Allergenic *Aspergillus* exhibited minor occurrence (0.8–1.8%), which is also in accordance with the conclusions of Basilico et al. (2007) (1.14%). The only exception appeared during the AC operation with dusty air filters, when its occurrence after 15 min increased sixteen times (to 12.6%).

CONCLUSION

The concentrations of bacteria and yeast were the highest prior to turning on the AC in the lecture room, both with or without students, and they were reduced

by air conditioning. The main source of bacterial and yeast pollution were obviously the people.

Based on the analysis of measurement results given in the above discussion we may conclude that the AC significantly contributes to the creation and maintenance of internal thermal comfort, particularly as concerns temperature and humidity in summer. Also a well serviced and maintained AC device can reduce the concentration of microorganisms. The presence of students inside the lecture room, if AC is out of operation, dramatically increases the incidence of bacteria and yeast.

Air conditioning reduced the concentration of a majority of microorganisms, but in the case of the allergenic genus *Aspergillus* and polluted filters the concentration went up sixteen times (to 12.6%). The most likely source were the conidia trapped in the dust-polluted filters. After 5 h of air-conditioning the concentration dropped below 0.8%.

Usage of better filters capable of capturing finer particles, particularly allergenic *Cladosporium,* would greatly contribute to the improvement of the internal environment in the lecture room. It is also important to ensure necessary AC maintenance, especially well-timed changing of polluted filters.

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