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Monitoring of Respiratory Health Risks Caused by Biomass Storage in Urban-Type Heating Plants

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Abstract: The aim of this work was to carry out long-term monitoring on the concentration and identification of phytopathogens in wood chip storage areas in urban-type heating plants. Three municipal heating plants in the central part of Slovakia were selected. The plants store biomass in large-capacity piles with a volume of 4 to 5000 m^3. Samples were obtained every year in the 2017–2022 period from the surface of the piles and from a depth of 0.5 m. Their moisture content was determined in the laboratory and the microbial analysis was performed by an accredited laboratory. The average number of colonies of phytopathogens did not differ significantly in individual years. The highest number of colony-forming units per gram was achieved by the species of the genera *Penicillium* and *Aspergillus*. In terms of occurrence in individual years and the frequency of occurrence in individual samples, the most frequently recorded species were *Mycelia Sterilia*, *Aspergillus brasiliensis*, *Aspergillus unguis*, and *Yeasts*. Based on the results achieved, in the future it will be necessary to establish legislative frameworks for these risks and, at least at the national level, work procedures for individual work activities, so that the health and life of the workers of the plants, as well as residents in the vicinity of this type of plant, are not endangered.

Keywords: wood chips; storage; health risks; phytopathogens; heating plants; wood biomass; airborne fungi

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1. Introduction

After the pandemic period and the deterioration of the global geopolitical situation, there was extraordinary pressure on the prices of fossil fuels, and subsequently, the demand for renewable energy sources also increased sharply [1–3]. In the past year, the lack of raw-wood materials in the European market placed increased demands on the optimal planning of logistics and biomass storage even for local producers of heat and electricity, which use biomass as the main energy source [4–7]. In most urban-type heating plants, storage piles are built for approximately 2 to 3 months, where, especially during the heating season, there is a continuous supply of new raw materials and also a continuous consumption of the stored material [8–10]. As a result of the course of biological processes in larger volumes of stored wood biomass, heat generation, changes in the moisture content and energy parameters and, last but not least, the formation of spores of phytopathogens and molds occur [11–16].

Wood dust and the production of phytopathogens during the storage of biomass in larger volumes represent a serious respiratory health threat for people and can cause a number of diseases that can result in the development of occupational diseases or even death [17–21]. While the existence of these risks and their identification is scientifically proven, there are still quite a few works that accurately quantify them and establish the limits of health risks as well as suggest proposals of measures for their minimization [22,23]. The risks related to the overheating of stored biomass and the possible occurrence of fire are relatively well documented [24–26].

Under the conditions of Slovakia, work with these risk factors is regulated by legislation in the form of Government Act 356/2006 Coll. and 83/2013 Coll. [27,28]. This legislation characterizes wood dust as a proven carcinogenic harmful factor and a technical guide value is set for it. Adherence to the technical guide values reduces the likelihood of harmful effects on health, but they cannot be completely excluded. They are the basis for preventive and protective measures. The technical reference value for wood dust is at the level of 2 mg per 1 m^3 of air at a temperature of 20 °C and an atmospheric pressure of 101.3 kPa (760 mm of mercury column) [27]. In terms of protection, workers should be equipped with protective clothing and personal protective work equipment for personal respiratory protection, which they must use during the entire duration of extraordinary exposure to carcinogens or mutagens. For phytopathogens, most species are classified in group no. 2: groups that can cause disease in humans and could pose a hazard to workers but are not likely to spread in the population, where effective prophylaxis or treatment is usually available. Most of the species occurring during biomass storage are also indicated for the development of allergic diseases [28]. However, permissible values and levels of exposure to these harmful factors are not specified in the legislation. Of the protective measures in the external environment for industrial processes, only protective clothing for workers is prescribed and the material should be handled in such a way as to minimize the risk of release into the air. Operational practice proves that in the case of industrial operations with biomass storage, even these simple protective measures are mostly not observed. There are also no limits on the distance of the location of such operations from human dwellings, which in the case of urban-type heating plants in inner cities and towns can represent a health risk for their residents.

In addition to energy use, there is another use of biomass as an innovative engineering material bonded with mycelium. Then, the substrate should also not contain any living biological impurities [29,30].

The study aims to identify health risks and propose effective countermeasures and legislative changes related to wood chip storage at urban heating plants for workers and residents.

2. Materials and Methods

2.1. Biomass Samples Collection

For the collection of sample material, three municipal heating plants in the central part of Slovakia were selected, which are separated from each other in cities with a radius of 45 km (storage 1, 2, and 3). These heating plants were chosen mainly for the similarity of operating conditions. All three heating plants are located in the inner city of cities that are similar in area size and population. They are available within a relatively small radius and are owned by the same enterprise. Climatic and meteorological characteristics are very similar in the given locations. The majority of urban-type heating plants in Slovakia work in approximately similar operating conditions. Therefore, there is no reason to believe that the results would be fundamentally different in the case of other urban-type heating plants in Slovakia. All three plants use forest chips mixed from coniferous and non-coniferous tree species in a ratio of 4:1 as the main energy source. They are stored in large-capacity piles with a volume of 4 to 5 thousand m^3. Residential buildings were always located near the piles at a distance from 35 (Figure 1) to 214 m as the crow flies.

Samples were obtained every year in the 2017–2022 period. Each year, in the last week of October, 5 samples were obtained from each storage location from two height levels (2 samples from a depth of 0.5 m and 3 samples from the pile surface). In each year, 15 samples were evaluated (a total of 90 samples). We chose two sampling height levels because the temperature and moisture characteristics of the stored biomass change fundamentally just below the surface and we wanted to verify the effect of these different conditions on the number of phytopathogen spores produced. Since the volume and shape of the pile changed every year due to the continuous supply and removal of biomass, the sampling locations also differed every year. They were chosen so that they were evenly distributed over the entire surface of the pile. The samples were obtained by placing them into plastic bags with a volume of 2 L using a metal spatula (Figure 2).

Figure 1. Storage 1 and biomass storage distance from civil buildings.

Figure 2. Sample collection in plastic bags.

Since all storage sites of heating plants are in the same region at a small distance, they also show similar climatic and meteorological characteristics. The average annual air temperature in the monitoring period at the sampling sites varied from 8.3 to 9.8 °C, and the average annual precipitation varied from 833 mm to 1071 mm. Similar conditions prevailed at all three sampling points throughout the monitored period. Two specimens were always collected from each sampling location, with one specimen used for microbiological analysis and the other for determining the relative moisture content.

2.2. Laboratory Analysis

To determine the moisture content, the samples were dried and weighed [31,32]. The samples were dried at 105 °C ± 2 °C to a constant weight. After weighing the samples on laboratory scales with an accuracy of 0.01 g, the relative moisture content values of wood chips were calculated. The relative moisture content at the individual sampling points was calculated by the ratio of the weight of the water contained in the samples to the weight of the wet samples, expressed as a percentage (Formula 1) [31,32]:

$$\mathrm{w_r} = (\mathrm{m_w} - \mathrm{m_0})/\mathrm{m_w}.100\ (\%), \tag{1}$$

$\mathrm{m_w}$—wet fuel sample weight (kg);

$\mathrm{m_0}$—weight of fuel sample after drying (kg).

Microbiological analysis of the samples was performed in an accredited laboratory of the Regional Office of Public Health in the Slovak Republic in two phases. In 2017–2018, the total number of phytopathogens was identified and individual species were identified. Since 2019, the laboratory has been able to determine the abundance of individual species of phytopathogens. Therefore, it was possible to evaluate the abundance of individual species only from 2019. The exact methodology of laboratory work is defined by standards [33–35] and is described in detail in studies [14,16]. The number of microorganisms was determined as the number of colonies forming a unit per gram ($\mathrm{CFU.g^{-1}}$) and is calculated according to the standard STN ISO 21527-2 [33]. Calculation on the abundance of individual types of phytopathogens was carried out in accordance with STN 56 0100 [35]. Identification of individual genera or species was made on the basis of morphological and cultivation characters according to the keys given in the literature [36,37].

Microsoft Excel (version 2013, Microsoft Corporation, Santa Rosa, CA, USA) and STATISTICA 12.0 (version 12.0, StatSoft Inc., Tulsa, OK, USA) software were used for calculating and visualizing the results.

3. Results

In the 2017–2022 period, the total number of spores of phytopathogens obtained in the samples was evaluated. For the 2019–2022 period, the abundance of individual species of phytopathogens (micromycetes) and the abundance of Yeasts were also evaluated.

3.1. Total Number of Phytopathogens and Relative Moisture Content of Stored Material

Since, in terms of technical possibilities, the sampling places were mostly from the upper layers of the stored pile, where the freshest material is assumed, the relative moisture content of the biomass was also relatively high. The average relative moisture content of the samples was at the level of 62.01%. More than 90% of all samples had a relative moisture content in the range of 60%–70%. Only eight samples out of a total of 90 had a moisture content between 10 and 50%. It is clear from this that the absolute numbers of individual spores of phytopathogens were identified at relative moisture contents of the samples above 60% (Figure 3).

It is also clear from Figure 3 that at storage sites 2 and 3, the number and moisture characteristics of the samples obtained were very similar. At storage no. 1, the relative moisture content of the samples obtained also varied significantly, as well as the number of colonies of phytopathogens, which in one case reached 5.9×10^7 $\mathrm{CFU.g^{-1}}$. This sample came from the surface of the pile and its relative humidity was the second highest of all samples collected (72.08%).

The distribution of the total number of phytopathogens depending on the place of sampling and the given year is evident from Figure 4. This graph also shows a difference between storage 2 and 3 where there were significant differences in the number of colonies depending on whether the samples were obtained from the surface of the pile or half a meter deep.

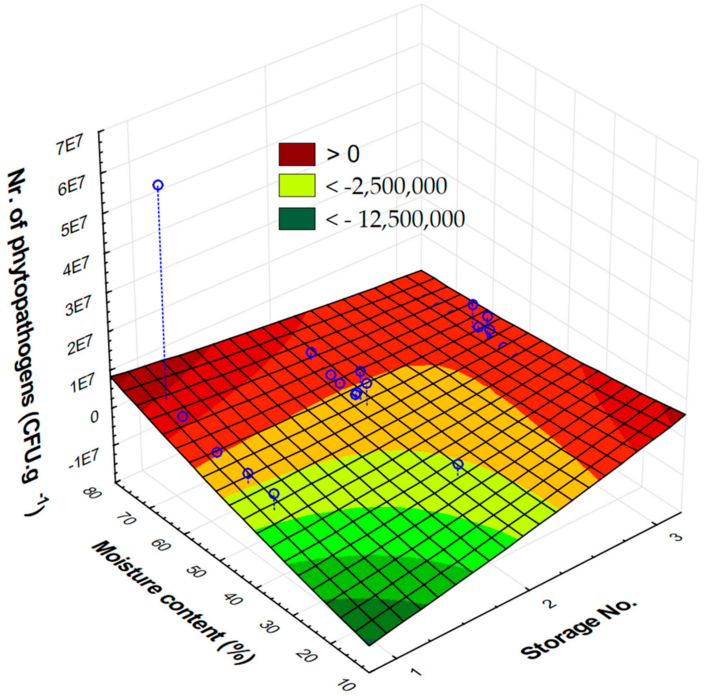

Figure 3. The number of colonies of phytopathogens per 1 g depending on the moisture content of the samples and the place of collection.

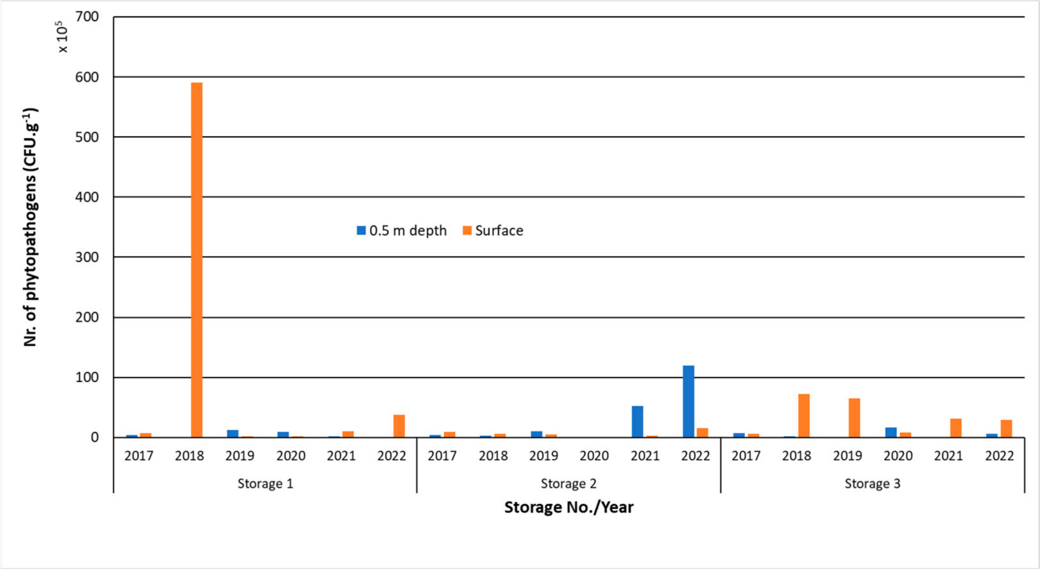

Figure 4. The number of colonies of phytopathogens per 1 g depending on the year and location of sampling.

However, except for one extreme case in 2018, we can state that the average number of phytopathogen colonies did not differ significantly in individual years. There is no logical reason for this significant difference. It may have been sample contamination, which is highly unlikely given the way the samples were collected and analyzed. It was probably a very specific material from a certain production location, where other technologies and methods of transportation and storage could have been used. However, we were unable to obtain or verify this information from the company. After excluding outliers and extreme values, it can be concluded that the average concentration of phytopathogens in wood chip samples did not change significantly in individual years and there are only slight differences between them. Atmospheric conditions were different in individual years and were also affected by the pandemic and the suppression of work activities. However, it did not fundamentally affect the number of colonies of phytopathogens (Figure 5).

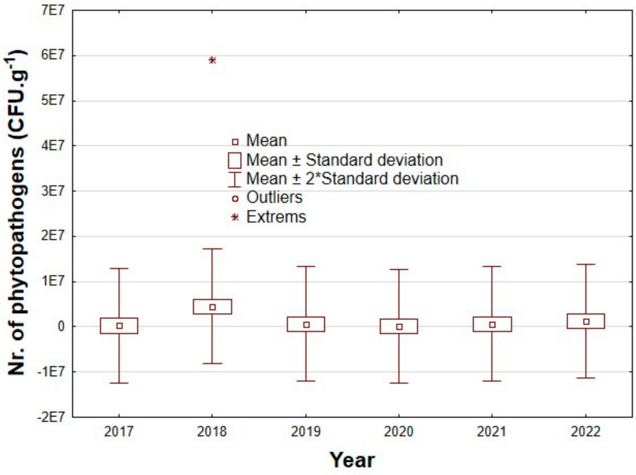

Figure 5. The average number of occurrences of phytopathogens in individual years and at individual sampling sites.

A partial difference in the number of colonies of phytopathogens can be observed depending on the sampling location (Figure 6). While thermal plants 2 and 3 show similar spore concentrations, in thermal plant 1 this concentration was almost double during the entire period. However, this result is mainly affected by one sample from 2018, in which extremely high concentrations of spores were identified.

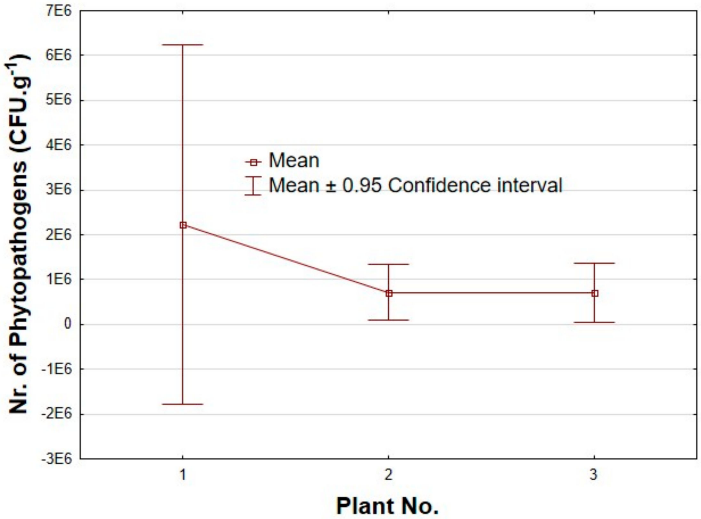

Figure 6. The average number of occurrences of phytopathogens at individual sampling sites.

Based on these results, it can be concluded that despite the regular natural circulation of biomass in heating plants' storage, the number of concentrations of spores of phytopathogens is maintained at the same level and thus represents a permanent risk to human health. Danger to human health is primarily caused by some identified types of phytopathogens and their concentration levels.

3.2. Identified Species of Phytopathogens and Their Number

Figures 7 and 8 show the identified species of phytopathogens and their abundance depending on the location and year of sampling. All species that have been recorded at least in one year at the given storage location are listed. The highest abundance of phytopathogens in samples from the 0.5 m level was at storage no. 2. The species of the genera *Penicillium* and *Aspergillus* reached the highest abundance. Numerous Yeast colonies were also recorded at all three storage areas. In terms of occurrence in individual years and the frequency of occurrence in individual samples, the most frequently recorded species were *Mycelia Sterilia*, *Aspergillus brasiliensis*, *Aspergillus unguis*, and Yeasts.

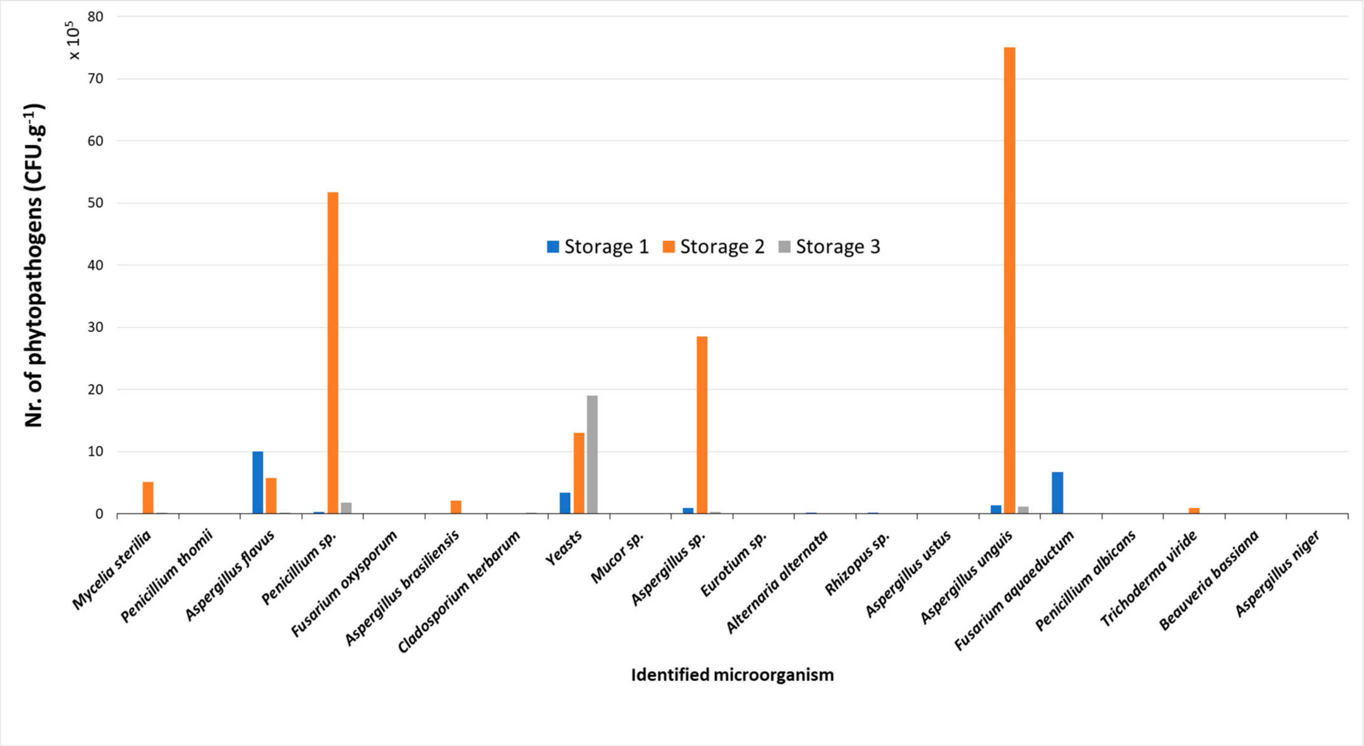

Figure 7. Identified phytopathogens and their number at individual sampling points from samples from a depth of 0.5 m.

Figure 8 shows the frequency of the identified species of phytopathogens from biomass samples obtained from the surface of stored piles in urban-type heating plants. It is obvious that the number of colonies is much higher on the surface of the piles than at a depth of 0.5 m. This is mainly due to better access to oxygen in this surface layer. The highest abundances of phytopathogens in this case were recorded at storage no. 3, where the species of the genus *Aspergillus* reached the highest abundance. As in samples from a depth of 0.5 m and in surface samples, numerous Yeast colonies were recorded at all three storage areas. The *Mycelia Sterilia* species had the highest frequency of occurrence in individual samples, including *Penicillium* sp., *Aspergillus brasiliensis*, *Aspergillus unguis*, *Aspergillus flavus,* and Yeasts. It can therefore be concluded that the height level of sampling was not decisive in terms of frequency of occurrence in individual samples. The sampling depth had an effect only on the abundance of individual colonies of the given species. The health risk for human health can be characterized as the same regardless of whether the sample came from the surface of the stored pile or from a depth of 0.5 m.

The species of the genus *Aspergillus* mainly cause Aspergillosis, which mainly affects the lungs and causes breathing problems, or a serious lung infection. In addition to the lungs, it can also affect the heart and brain. It represents a high risk, possibly life-threatening, for immunocompromised patients [38]. Relatively high concentrations of species of this genus in almost all samples obtained indicate a potential health risk.

The species of the genus *Penicillium* sp. and *Aspergillus* spp. are also known producers of mycotoxins (e.g., Citrinin, Patulin). The species *Aspergillus flavus* produces alphatoxins. All these types of mycotoxins have moderate to high toxicity. The severity of mycotoxins in human medicine remains largely underestimated because the material from patients with characteristic symptoms is rarely tested for mycotoxins. However, their mutagenic and carcinogenic effects are quite well known [39–41].

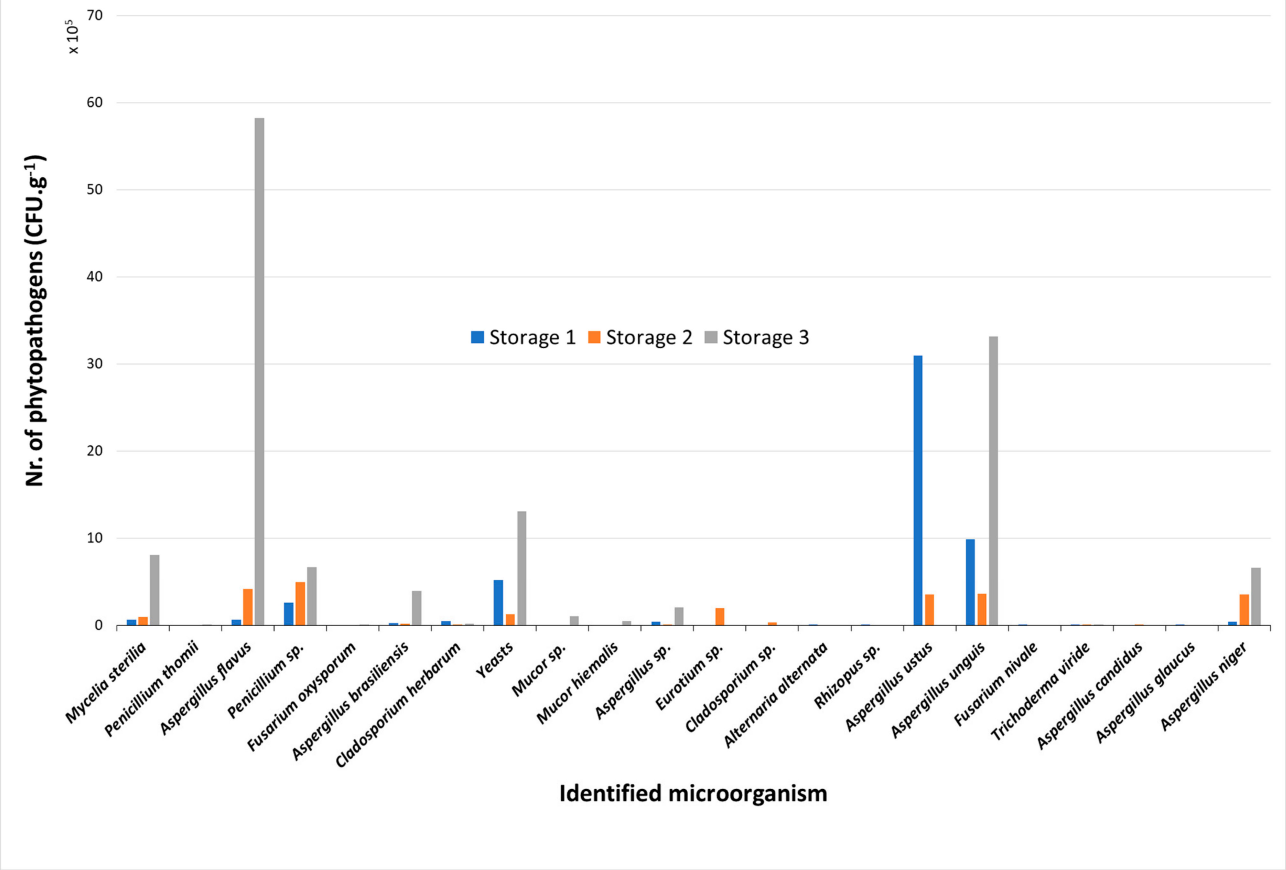

Figure 8. Identified phytopathogens and their number at individual sampling points from surface samples of stored piles.

Yeasts produce enzymes of various invasive natures and can cause serious health problems in partially immunocompromised patients. On the part of the respiratory and circulatory systems, it is shortness of breath and subjective feelings on the part of the cardiovascular system. Symptoms of candidiasis include a hard-to-remove white coating on the tongue, bad breath, allergies, eczema, nail, and skin diseases, burning and itchy scaly spots on the skin, itching all over the body, hair loss, itchy scalp, recurrent genital infections, frequent infections of the urinary tract, enlarged nodes, itching and burning eyes, joint pain, attention disorders, dizziness, headaches, and depression [42,43].

The results thus confirm that the identified microorganisms occur in biomass storage for a long time and can cause a wide range of diseases, while for people with weakened immunity or in combination with other serious diseases, they can even cause a life-threatening risk.

4. Discussion

Most works identify risks associated with spore production during biomass storage only after a certain period of storage (e.g., 3 or more months) [20,44]. Our research confirmed that this risk also arises with the regular rotation of biomass in storage areas with a larger volume, where the total storage period may not even be 3 months. Scholz et al. [45] found two to four species of phytopathogens in the wood chip samples of large fractions and four to eight species of potential human pathogens, poison-causing, or allergenic species in small fraction chips, which occurred at least temporarily during one year of storage. Our results show that, even with a multi-year operation and regular circulation of biomass, at least two to three types of phytopathogens in high concentrations, which can have a negative impact on human health, are permanently present in the stored piles of forest chips. Numerous colonies of phytopathogens can be identified after 30 days of storage in biomass storage in industrial conditions [46].

Barontini et al. [17] evaluated the concentration of spores of phytopathogens during biomass storage per 1 m^3 of air. The highest incidence was recorded in the genera *Alternaria* spp. and *Cladosporium* spp. The exposure of the operators who handled the chips was at the level of 4864 ± 580 CFU.m^{-3}. Based on this, it can be concluded that lower concentrations of phytopathogens are released into the air when handling wood chips than are found in the biological material itself. However, the exposure is still significant enough to cause serious health problems. Garstang et al. [47] determined the number of spores from air samples collected during wood chip handling. Based on the analysis, they identified 267 CFU.m^{-3} for thermophilic actinomycetes and 50 CFU.m^{-3} for *Aspergillus fumigatus* species. This study also confirms the lower concentration of spores in the air than in the stored material itself.

In most operations that use wood chips as the main energy source, the storage method also plays a role in the development of phytopathogens. Since these are mostly large volumes, storage takes place on paved or unpaved surfaces, mostly without coverage. If the chips were stored in a closed-covered space (e.g., silo), the number of phytopathogens could be significantly lower [48]. Laitinen et al. [49] found high exposure levels of actinobacteria, bacterial endotoxins, and fungi mainly during the unloading of wood chips in thermal and biomass power plants in Finland. In addition, workers were exposed to mechanical irritation from organic dust and chemical irritation from volatile organic compounds and diesel exhaust components. During operation, workers were also exposed to endotoxins, actinobacteria, and molds, especially when cleaning and handling wood chips in silos and when working near screens or crushers. The measured concentrations exceeded the limit values proposed for these substances.

Similarly, several works state the absence of globally recognized safety procedures in the storage and handling of stored biomass [44,50]. In addition, safety procedures for handling wood chips should be established and continuously monitored, as there are known cases of health damage resulting in death [51].

5. Conclusions

Long-term monitoring of large-scale biomass storage in urban-type heating plants confirmed the constant presence of phytopathogens with a potential threat to human health. Currently, adequate attention is not paid to this threat in industrial conditions and legislation does not specify exact exposure levels and only recommends the use of personal protective work equipment in these operations. The emergence of serious illnesses and occupational diseases as a result of the action of these risk factors may already increase in the near future. Since relatively high concentrations of phytopathogens in the air during the handling of wood chips have been confirmed in other works, they represent a potential risk not only for the workers of the given operation, but also for residents who have their homes near such plants.

Another subject of the research should therefore be exposure levels at different distances from biomass piles as well as the potential of their air transport depending on atmospheric conditions.

The first experiments with the application of chemical substances are already underway, which have the task of reducing the degradation of biomass and also reducing the production and growth of colonies of phytopathogens. However, the application of calcium hydroxide $Ca(OH)_2$ did not prevent biomass degradation and only small differences in the composition of microbial communities were found. This confirms the tolerance of microbes to changing environmental conditions and adaptation to them [52,53].

The greatest health risk in this type of operation is represented by the species of the genus *Aspergillus* and Yeast infections. In most cases, they can cause serious respiratory diseases, skin diseases, and allergies. Several species produce dangerous mycotoxins, which belong to carcinogens and mutagens. This is also why in the future it will be necessary to establish relevant legislative frameworks for these risks and, at least at the national level, generally accepted work procedures for individual work activities, so that the health and life of the workers of plants, as well as residents in the vicinity of this type of plant, are not endangered. Existing recommendations for permitted concentrations are more for indoor spaces. According to the working document of the European Commission for Health and Consumer Protection, the recommended limits for Aerobic Plate Count are <105 CFU/g or mL; Anaerobic spore-formers < 105 CFU/g or mL; Yeast and Mold Count < 1000 CFU/g or mL [54]. Based on analyzed infectious disease studies, the recommendations of the World Health Organization for indoor air quality recommend permitted concentrations according to the type of phytopathogen from 500 CFU/m^3 to 1000 CFU/m^3 for airborne fungal spores in indoor air in urban areas [55].

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