REPORT
DELIVERABLE 6.12

outPHit WP6 – Monitoring

Innsbruck, 18/08/2024,

LIVING QUALITY IMPROVEMENT VERIFICATION (TASK 6.5)

D.6.12 REPORT ON MICROBIOLOGICAL ASSESSMENT OF INDOOR AIR QUALITY IN CASE STUDY PROJECTS

Last updated 18/08/2024 by University of Innsbruck

CONTACT

Rainer Pfluger
University of Innsbruck
Unit of Energy Efficient Building
rainer.pfluger@uibk.ac.at

Sascha Hammes
University of Innsbruck
Unit of Energy Efficient Building
sascha.hammes@uibk.ac.at

OUTPHIT – DEEP RETROFITS MADE FASTER, CHEAPER AND MORE RELIABLE

outPHit pairs such approaches with the rigour of Passive House principles to make deep retrofits cost-effective, faster, and more reliable. On the basis of case studies across Europe and in collaboration with a wide variety of stakeholders, outPHit is addressing barriers to the uptake of high-quality deep retrofits while facilitating the development of high-performance renovation systems, tools for decision making and quality assurance safeguards. outphit.eu http://www.klimabuendnis.org/

This project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 957175. The presented contents are the author's sole responsibility and do not necessarily reflect the views of the European Union. Neither the EASME nor the European Commission are responsible for any use that may be made of the information contained therein.

TABLE OF CONTENTS

INDOOR AIR QUALITY AND THE ROLE OF BIOAEROSOLS

INTRODUCTION

Heat loss via the building envelope is one of the largest energy consumers of a building. Accordingly, measures to increase the energy efficiency of the building façade are essential to fulfil climate policy objectives. Comprehensive refurbishments offer the advantage of reducing energy consumption to such an extent that, in the best-case scenario, the remaining demand can be covered by renewable energies on site, e.g. EnerPHit Plus or Premium [1]. However, it is not only an improvement in energy efficiency that can be achieved by refurbishing the thermal envelope, but also a holistic improvement in indoor air quality (IAQ).

The refurbishment of the building envelope is associated with improved airtightness. This also means that the pollutants in the air (e.g. indoor air contaminants and water vapour) must be actively removed to a greater extent. This is done either manually or by using mechanical ventilation concepts. The improved thermal performance of the building envelope leads to greatly increased interior surface temperatures. Ultimately, this minimises the risk of mould and maximises thermal comfort. To ensure that the planned energy standards have been achieved, on-site measurements can be used to monitor success. This data is important not only to check whether the refurbishment measures meet the desired performance standards, but also to identify areas that require further improvement. This includes checking whether new insulation materials are working as expected and whether heating, ventilation and air conditioning (HVAC) control systems are operating efficiently. In addition to the energy assessment, the measurement of temperature, relative humidity, carbon dioxide levels and, where possible, spot checks of microbiological indicators of air quality are important for indoor air quality.

Due to the length of time people spend indoors (up to 90 % of their lives), the increasingly ageing population and the simultaneous rise in the number of more sensitive people, indoor air quality has become much more important [2], [3], [4]. While many studies focus primarily on the CO_2 concentration in the field of IAQ in the context of monitoring the success of retrofitted buildings, this report aims to highlight the methodology and significance of microbiological testing.

This is because microbiological sampling is essential for assessing indoor air quality, as it provides a detailed understanding of the types and concentrations of microorganisms present. These include, in particular, bacteria and fungi that can affect human health and the integrity of building structures.

LIMITATIONS FROM CONVENTIONAL ROOM CLIMATE SENSORS

Indoor climate sensors can provide real-time data on indoor air quality. The most commonly measured variables include CO_2 concentration, temperature, relative humidity and VOC (Volatile Organic Compounds). CO_2 and VOC values are used as a control variable for mechanical ventilation and as an indicator of its effectiveness. Temperature and humidity sensors provide insight into comfort levels and the humidity level can indirectly influence microbial growth. However, these sensors do not detect the presence, diversity or concentration of microorganisms in the air or on surfaces, which can significantly affect IAQ and health. Microbiological sampling of indoor air involves collecting and analysing airborne microorganisms, including bacteria, viruses, fungi and their by-products. Microbiological sampling provides direct insight into airborne components that are often associated with health effects such as respiratory infections, allergies and asthma [4]. Regular microbiological sampling can detect mould spores even when electronic sensors indicate acceptable humidity levels, allowing for a more accurate assessment of potential mould problems.

OBJECTIVE OF THIS REPORT

Microbiological sampling in outdoor and indoor air and on selected surfaces of walls, windows and other surfaces of the building fabric as well as in ventilation systems can provide insights into aspects of indoor air quality that cannot be easily captured by electronic sensors. The large number of projects across a wide geographical area that have been studied as demonstration projects of the outPHit project provides a unique opportunity to apply a standardised sampling and assessment methodology and derive comparable results. The sampling and assessment methodology will be documented in this report. The survey was carried out by the local partners with the help of the commissioned laboratories, where available. The results were analysed and compared and documented in this report.

STRUCTURE OF THE REPORT

This document first presents the different types of microbiological samples, their influence on human health and the influence of ventilation on individual fungal and bacterial genera and their growth rates. Subsequently, the methodology for the collection and evaluation of the individual fungal and bacterial genera is described and their representation in the monitoring with interpretation of values. The results of real microbiological assessments from field studies from the outPHit project are then presented and discussed in the context of the scientific literature. In particular, the importance of ventilation and legal requirements are emphasised. Finally, a conclusion is drawn on the significance of microbiological testing in the context of energy-efficient building refurbishment to the EnerPHit standard.

BIOAEROSOLS – FUNGI AND BACTERIA

Bioaerosol particles are pollutants that can have a negative impact on indoor air quality [5]. Bioaerosols can account for up to a third of indoor air pollution [6], [7]. A bioaerosol is a colloidal suspension consisting of liquid droplets and solid particles which, due to their small size, can float in the air (aerosol) and to which fungi, viruses and/or bacteria adhere [5], [4]. Due to their small size (size range from 0.01 µm to 100 µm [8]), these airborne particles of biological origin can spread accordingly. Due to their small size, these particles are also known as microbial fine dust.

The concentrations of fungi and bacteria in the outdoor air are determined in particular by the interaction of seasonal and short-term weather conditions and circadian patterns of light, humidity and temperature. The microorganisms in indoor air originate both from the outdoor air entering a building and from the occupants of a building and their activities as well as from contaminated building materials and interior furnishings [9].

Bioaerosols usually consist of a mix of fungi, viruses and bacteria as well as pollen. Indoors, however, fungi make up an average of two thirds of the total concentration of bioaerosols, while bacteria account for almost a third and viruses for only a small proportion. Outdoors, the proportion of fungi and their spores is even higher (up to three quarters). There is even a significant difference between the proportions of airborne bacteria and fungi outdoors and indoors, which is due to the fact that conditions are better for bacteria indoors [9].

Airborne bacteria and fungi have been linked as the cause of non-specific diseases such as sick building syndrome (SBS) and specific conditions such as respiratory allergies and asthma [4] especially in early childhood. It has been shown that poor ventilation of rooms in particular can lead to an increase in the accumulation of pollutants (such as bioaerosols) and thus also increase the risk of sick building syndrome occurring among building occupants [10]. Accordingly, one of the most important factors influencing IAQ and the proportion of bioaerosols in the air is the operation of heating, ventilation and air conditioning (HVAC) systems [4], [11]. It has also been shown that most types of fungi or bacteria require a relative humidity of at least 60% in order to survive [12], [13]. It has been shown that the three main growth requirements “temperature, humidity and substrate” must be present simultaneously for a certain period of time to enable fungal growth. The work of Sedlbauer (cf. [14]) makes it possible to determine based on isopleth systems development of spore germination times and mycelium growth, whereby the influence of the substrate is also considered. As temperature and relative humidity (so called activity of water) at the surface of the building fabric can be measured, at least two parameters of the isopleth system can be easily recorded.

The particles that bacteria and fungi transport outdoors are usually somewhat larger than those indoors. In addition, meteorological parameters such as wind direction and relative humidity can be used to predict fungal concentrations in particular [9].

Besides these particles, it has been shown that certain microorganisms produce characteristically unique microbial volatile organic compounds also known as MVOCs. The odours that microorganisms produce may be attributable to distinct MVOCs produced by certain species of microorganisms. The methods of analysis of MVOCs by cryogenic preconcentration coupled with gas chromatography and mass spectrometry (GC/MS) is not part of this report, as it was found in [15] that no evaluation criteria for assessment of mould infestation indoors through the measurement can be derived from MVOC.

FUNGAL SPORES AND FUNGI

Aspergillus (with subspecies fumigatus, niger, flavus) is a type of fungus that is spread via bioaerosols and can cause respiratory problems. More frequently, the fungi Penicillium and Alternaria as well as Chaetomium species can be found as indoor pollutants, especially in humid areas such as bathrooms and wall surfaces with high humidity. Penicillium can produce mycotoxins. These are secondary metabolites that can affect human health. Penicillium species can also be associated with sick building syndrome [16]. Alternaria and Chaetomium species can cause respiratory problems. Stachybotrys is another fungus that can be found in particularly damp living areas and produces harmful mycotoxins.

BACTERIA

As with fungi, which thrive particularly well in high humidity, the bacteria Pseudomonas aeruginosa also grows well under such conditions. This bacterium can lead to respiratory infections. Bacillus species are often found in dust and indoor air and can cause opportunistic infections. Other bacteria that are frequently detected in bioaerosols are Staphylococcus aureus, Actinomycetes and Streptomyces. As bioaerosols are usually absorbed via the respiratory tract, this is where most infections occur.

FREQUENTLY TESTED MICROORGANISMS

The bacterial genera Micrococcus and Staphylococcus were most frequently identified in indoor air in the study by Bonetta et al [4], [17].

Penicillium, Aspergillus and Cladosporium are among the most widespread genera of fungi [17], [18], [19], [20]. However, seasonal differences can be observed here. Penicillium is detected as more dominant in indoor air in winter and spring, while Cladosporium is usually the most widespread genus in summer [4]. This trend was similar to that frequently observed in outdoor air [21]. Macher et al. found that Cladosporium was more abundant outdoors in autumn and indoors in winter [9].

Because of this seasonal variation of species and to detect whether the source of microorganisms is from indoors or outdoors, samples from indoors and outdoors (as reference) must be taken at the same time during field measurements.

AIR CONDITIONING SYSTEMS

Air conditioning systems can provide favourable breeding grounds for the colonisation of bioaerosols if the system is operated and specified incorrectly [22]. Condensate forms in the air conditioning systems because of cooling below the dew point. Microorganisms can multiply if maintenance and cleaning are inadequate. The situation is different with simple mechanical ventilation systems with heat recovery (MVHR). In ventilation systems, outside air is heated via the heat exchanger, the relative humidity, which is already low in the outside air in winter, is only heated further and thus becomes even drier. This means that there is no risk of microorganisms growing in the fresh air path.

For this reason, the aim of the study by Bonetta et al. was to assess the content and composition of bacteria and fungi in the indoor air of an office building with an Air conditioning system in Turin, Italy. Microbiological assessments were carried out in the outside air, in the supply air diffusers, in the air flow of the fan coil unit and in the humidification water tank of the air treatment unit. These results indicate that bacteria and fungi could proliferate in the ATU filter (air treatment unit) and in the air condition ducts if the air condition system is switched off, particularly overnight. By cooling down, the surface temperatures inside the air conditioning system fall below the dew point and humidity conditions can prevail that favour growth. Restarting the air conditioning systems carries the risk of spreading bacteria and mould spores [4]. For this reason, plant operation needs to be optimised in terms of energy and IAQ. Better filters (PM<2.5) would contribute significantly to the improvement [9]. Regular cleaning and maintenance work must be carried out [4].

MICROBIOLOGICAL ASSESSMENT

Among indoor air pollutants, bioaerosol is one of the most important pollutants [5]. Bioaerosols are present in the atmosphere in sizes of more than 0.2 µm and consist of living organisms such as bacteria, viruses and fungi [8]. The assessment of the number and species of individual fungal and bacterial genera for IAQ has already been emphasised. In this chapter, the measurement methodology is presented.

IMPLEMENTATION OF THE DATA COLLECTION

The most interesting period for the collection of microbiological data is the heating period. This is because windows are mainly closed during this time due to the cold outside temperatures. This means that pollutants are less easily removed without mechanical ventilation. This is therefore the least favourable period of the year for air quality parameters. In winter, outside temperatures drop considerably, making the outside surfaces of buildings very cold. These low outside temperatures lead to a corresponding drop in the surface temperatures of interior walls, especially in areas with high U-values or thermal bridges. If the internal surface temperature of a wall falls below the dew point of the indoor air, condensation can occur. This creates a damp environment that is ideal for mould growth.

Indoor surface temperature and surface humidity (so called activity of water, a_w) are the decisive factors for mould. To assess the air quality, the surface temperatures of the coldest areas can therefore be recorded with an infrared camera in the same way as for assessing energy efficiency. Room temperature and room air humidity must be recorded at the same time as the surface temperature. Assuming the absolute humidity of the air to be the same at the measurement point of the room air as well as at the surface, the relative humidity at the surface can be calculated. If the a_w-value is higher than 80%, mould growth is possible, depending on the type of fungi and on the substrate. Hence, temperature and humidity measurement are helpful to indicate any mould risk, but it does not constitute proof that mould is occurring.

The microbiological examination in the context of IAQ is therefore crucial for assessing whether a set improvement in energy efficiency has a positive or negative impact on indoor air quality. The latter effect can occur, for example, if airtightness is achieved without a ventilation system and there is insufficient manual ventilation.

Penetrating moisture and condensate at cold surfaces or in building fabric (by convection or diffusion) or even water ingress can lead to mould growth and deterioration of building materials, which affects both energy efficiency and the health of the occupants. However, the greatest risk of mould arises from moisture on the surface when surface temperatures are too low due to insufficient insulation or thermal bridges. To place the measurement data in a building physics context, the performance of the HVAC system must be assessed in addition to the evaluation of the thermal envelope (moisture content of materials, thermal bridges) and air leakage measurement (effectiveness of seals and barriers). Therefore, parameters such as airflow rates and operating times of HVAC systems should be measured at the same time as microbiological sampling. Measured time series of CO2 and humidity can by themselves also be used to assess the MVHR performance and help identify inappropriate settings of the air flow rate.

Effective sampling requires comprehensive planning, including selecting the parameters to be measured, determining the sampling locations and creating a schedule for the measurements and recording sources of interference and general conditions. For example, how long a person was present at the sampling location before the sample was taken, should be noted in the protocol. Moreover, the outdoor conditions (wind, temperature, humidity) and time of year, should be documented, as Macher et al. show in their study that the concentrations of many bacteria and fungi are negatively correlated with temperature and wind speed and positively correlated with relative humidity [9].

Air sampling is the most direct method for assessing the microbial content of indoor air. This can be done by impaction as shown in Figure 1.

Figure 1: Air sampler (impact sampler MBASS30, Hohlbach) in use in the Kranichstein Passive House, Darmstadt. Source: [23].

Figure 2: Filter holder and filter for isokinetic sampling in use in the Kranichstein Passive House, Darmstadt. Source: [23].

The room air is drawn through the so-called impactor, where particles and microorganisms are collected on a solid surface, such as agar plates (defined culture medium depending on the target size). This method is effective in capturing viable microorganisms and cultivating them immediately for subsequent analyses. The impactor method is also used in [9]. As mentioned, it is important that a reference of the outdoor situation is collected at the same time as the sample is taken in the room.

Another method is filtration, in which air is passed through a filter that captures airborne bioaerosols. The filters are analysed in the laboratory. In both methods, a defined amount of air is drawn in using a pump and collected to assess the presence of individual microorganisms and their number in the room air. In the context of refurbishment, the data analysis allows the IAQ to be compared before and after improvement of the thermal envelope.

To detect mould on surfaces, a photo documentation can help. At visible places with mould, mould determination is done using the contact method. The “flap plate” is pressed onto the affected area and sent directly to the laboratory for evaluation.

However not all spores and fungi are visible with the naked eye. In this case, construction forensic using a UV- lamp and a camera with special filters for forensic photography can help to detect invisible mould.

The focus of this report is on the assessment of the indoor air, so the flap plate method and the construction forensic is not elaborated further here, but it can help to localize and identify areas affected by mould.

CULTIVATION METHODS

The impaction method is one of the most common methods for identifying and quantifying microorganisms. Cultivation of the bioaerosols aspirated is necessary. As standard, the samples are incubated on agar plates or in liquid media so that they can grow. The resulting bacterial colonies and fungal outgrowths are then quantified and identified based on their morphological characteristics and biochemical tests.

Bacteria are typically isolated on (trypticase/trypsin) soya agar [4], [5], [17]. Moulds and yeasts are isolated on malt extract agar [9]. Sabouraud dextrose agar or Rose Bengal agar are also used in the literature as a culture medium for airborne fungi (see [4], [17]). Depending on the target size, culture medium and analysis method, the culture media are then incubated for several hours (usually 48 h) with bacteria at 36 ± 1 °C and for the fungal count for 4-8 days at 24 °C [4], [5].

In the study by Macher et al. the moulds are divided into six categories in the assessment: (1) moulds with darkly pigmented hyphae and/or conidia, which include Alternaria and Cladosporium, for example (see section **Fungal spores and fungi**), (2) moulds with hyaline or light-coloured conidia (e.g. Penicillium, see section **Fungal spores and fungi**), (3) mould fungi in which spores occur within a sporangium, (4) more complex classes of moulds with unique clamp connections between the hyphal cells, e.g. wood rot moulds, (5) yeasts and (6) others.

INVESTIGATION

For the detection and counting of mould fungi in the air, a measurement according to ‘DIN ISO 16000-18: Sampling by impaction’ is suggested. The entire measurement procedure and the required materials are described in DIN ISO 1600-18. The air samples are analysed in a microbiological laboratory. The required equipment and the timing of the measurements should be discussed with the laboratory at an early stage.

The air samples can also be collected independently. In the outPHit project, the sampling was carried out by the individual project partners themselves where possible. The measurement was carried out at the same time as the installation of the data loggers for temperature, relative humidity and CO_2 concentration. This not only reduces the workload of the outPHit project partners, but also provides the necessary framework information to relate the microbiological results. A reference measurement of the outside air is just as obligatory as the thermography-based measurement of surface temperatures. Optionally, as mentioned, mould can be made visible on the walls using a forensic UV lamp. The room must be darkened for this.

For sampling in outPHit, the tenants were asked to keep the windows in the analysed rooms closed for at least 8 hours before the measurement. Occupant presence and ventilation times immediately prior to microbiological sampling must be recorded to interpret the data adequately.

The workflow has already been described in detail in Deliverable 6.1 (**D6.1 Measurement of the Air Quality - Moulds, Cultivable Airborne Fungal Spores**). This includes in particular:

- Instructions for tenants (rules of behaviour before and during sampling)
- Details on air sampling (cleaning the air sampler, inserting new Petri dishes with culture media, preparation for the laboratory)
- Collection of additional data (temperature, humidity, visual damage, indoor orientation, outdoor conditions (current weather conditions, including wind), date and time, ventilation (mechanical/manual))
- Reference measurement of the outside air
- Evaluation of the laboratory results (interpretation of germ density, genera)

In principle, it is often up to the laboratory to make an expert assessment as to whether the measured concentration of mould and bacteria in the indoor air is an indicator of mould growth in the home.

MICROBIOLOGICAL TESTING AS PART OF THE OUTPHIT MONITORING

The average concentration of microorganisms in indoor spaces collected in one day is usually given in units of colony-forming units per cubic metre (CFU/m³). Values above 500 CFU/m³ are reported as moderate contamination, while values above 2000 CFU/m³ indicate high contamination [2], [24]. In addition to determining the number of airborne colony-forming units per cubic metre, it is advisable to identify the predominant taxa [2].

INDIRECT MOULD EVALUATION ON THE HISTORY OF INDOOR CLIMATE MEASUREMENTS

The growth of bacteria and fungi is favoured by poor ventilation, resulting in indoor air relative humidity levels of more than 60% [12], [13]. The growth of specific types of mould and bacteria is also linked to specific temperatures. As already noted in Section **Limitations from conventional room climate sensors**, conventional room climate sensors cannot be used to gain any dedicated insights into the microbiological components and their frequency. However, data logging of indoor climate values (relative humidity and room air temperature) and knowledge of thermal bridges and ventilation rates can be used to make potential assumptions about the risk of mould, but hardly about the type and extent, as this depends on several factors. This is solved using probabilistic models based on seasonal frequency distributions of certain fungi and bacteria in the outdoor air. Ventilation rates can be recorded for window ventilation via window contact sensors, for example, and for mechanical ventilation via CO_2 logging and/or logging of the operating states of the ventilation system (time and speed).

Simulation software can also be used to determine sufficiently accurate assumptions about the risk of mould growth on interior wall surfaces. The software used here is e.g., Delphin-Plugin from Bauklimatik-Dresden [1] and WUFI® Bio - Assessment of Mould Growth Risk from the Fraunhofer Institute for Building Physics (IBP) [2].

[1] **https://bauklimatik-dresden.de/**, 07.2024
[2] **https://wufi.de/de/2017/03/31/wufi-bio/**, 07.2024

FIELD MEASUREMENTS

As an example for this report, a valid microbiological investigation of an outPHit case study was carried out, which is representative of an energy-efficient building refurbishment. In addition, the microbiological results of a new energy-efficient building constructed to the passive house standard are presented. For the latter, data from a preliminary project by the Passive House Institute in Darmstadt was used.

VAND (SPAIN)

Building Information

As a case study in Spain, there is a multi-party residential building in Teruel, in the autonomous region of Aragon in the eastern centre of Spain (cf. Figure 3). The climate is temperate to warm; rain (approx. 415 mm/year) falls throughout the year [3]. It is a social housing building and part of a residential complex with two other apartment blocks that are to be renovated in the next few years. The building was erected in 1970 using traditional construction methods: a concrete frame with a ceramic, double-skin façade, an uninsulated pitched roof, balconies, an unheated attic and basement, steel frames and single-glazed windows with shutters. The building has numerous comfort problems and is characterised by high energy consumption (406 kWh/m^{2}a according to PHPP calculations). After implementing the refurbishment measures, the energy requirement was reduced by 350 kWh/m^{2}a to 66 kWh/m^{2}a (see: **https://outphit.eu/en/case-studies/list/detail/CS17/**, 07.2024). A Decentralised supply and extract air ventilation system separate for each flat (Ventilation standard air flow rate: 0.4 m^3/h) was used for ventilation. New ventilation units with heat recovery have been placed in suspended ceiling in bathrooms. All rooms in the building are ventilated[4].

---

[3] **https://de.climate-data.org/europa/spanien/aragonien/teruel-2152/**, 07.2024

[4] **https://outphit.eu/media/outphit_case_studies/outPHit_D5.2._CS17_RenovationApproachDocuments.pdf**, 07.2024

Figure 3: Social housing Parque Maquinaria de Teruel, outPHit CS17. Image source: Nuria Díaz Antón / VAND.

Data collection

The measurement of the outdoor situation (HUMLOG 20 data logger, period April 2023 to July 2024) shows a very wide range of outdoor temperatures in Teruel, Spain (min: -6.8°C, max: 43.2°C). The average is 15.4°C + 8.5. Figure 4 and Figure 5 show the temporal distribution of the temperature and the frequency distributions of individual temperature levels.

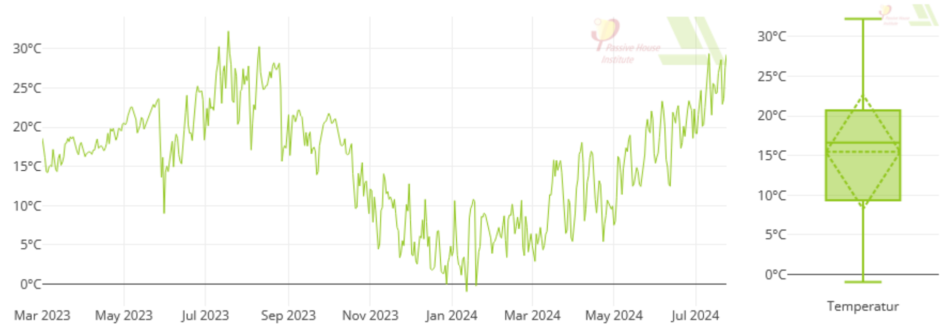

Figure 4: Outdoor Air-Temperature for outPHit CS17, Part A, one value per day. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

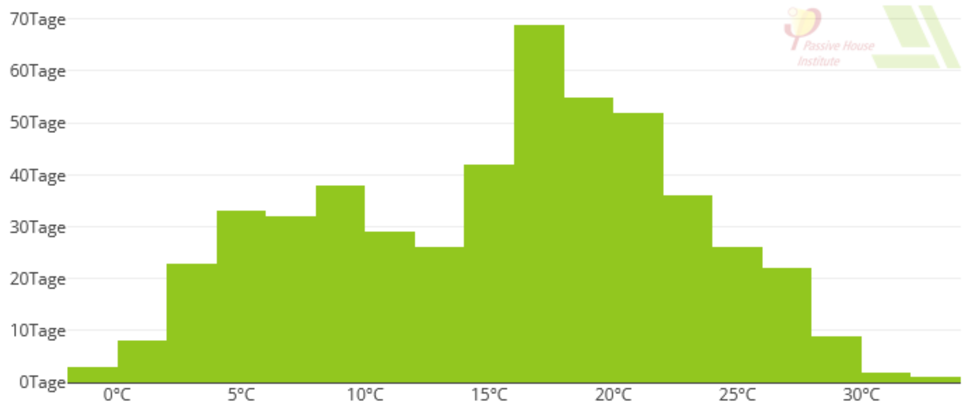

Figure 5: Outdoor Air-Temperature for outPHit CS17, Part B, one value per day. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

The relative humidity outside is on average 59.6% ± 13.1. The following figures show the temporal progression over the measurement period. The relative humidity was in the range of 45% to 70% on most days.

Figure 6: Outdoor Air-Temperature for outPHit CS17, Part A, one value per day. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

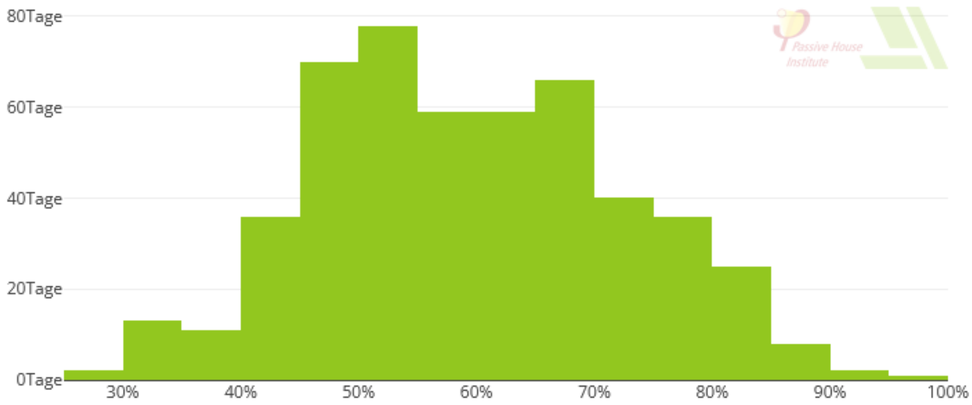

Figure 7: Outdoor Air-Temperature for outPHit CS17, Part A, one value per day. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

The microbiological examination shows that all values are within the normal range (Figure 8 and Figure 9). Sampling was carried out before and after the renovation (before renovation: Feb 2022, after renovation: Jan 2024). No outliers were detected before the renovation either.

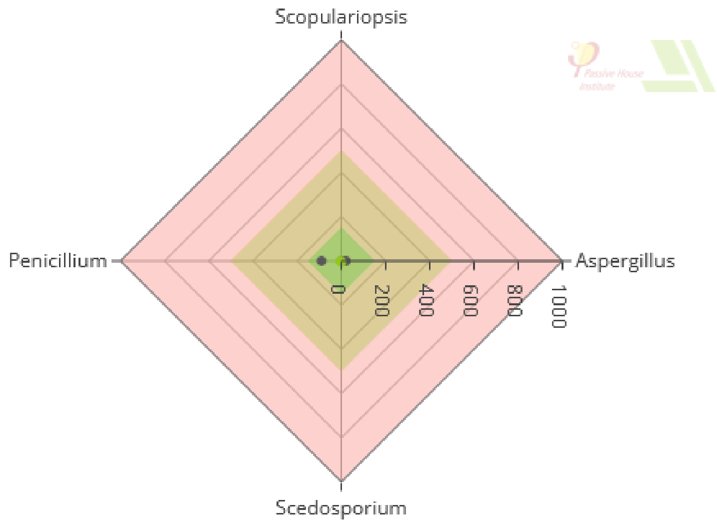

Figure 8: Indoor Microbiological Assessment for outPHit CS17, Room 1 (Bajo_Dch), unit: CFU. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

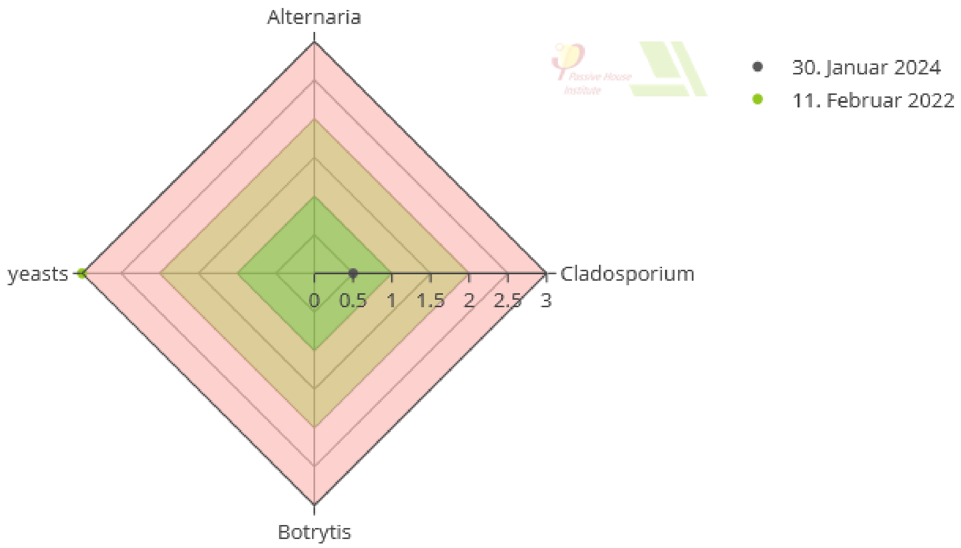

Figure 9: Indoor Microbiological Assessment for outPHit CS17, Room 2 (Bajo_Dch), Part A, unit: CFU. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

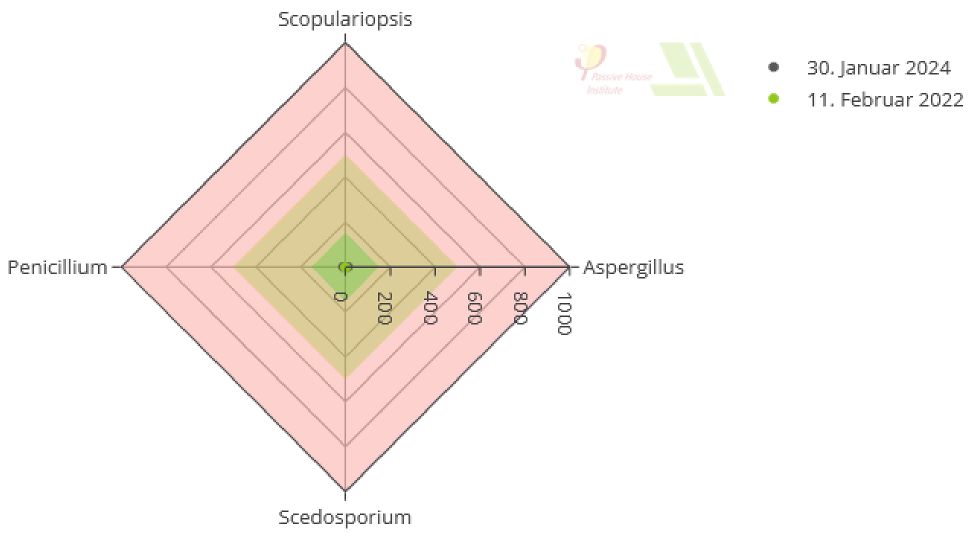

Figure 10: Indoor Microbiological Assessment for outPHit CS17, Room 2 (Bajo_Dch), Part A, unit: CFU. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

A comparison of the pre- and post-monitoring data for an example room in the Spanish case study in Teruel regarding temperature, CO_2 concentration and relative humidity shows how the EnerPHit standard reduces the risk of mould. Effective ventilation is a key element here. Implemented in the case study via MVHR. The effect is evaluated via the relative threshold deviation (RTD), i.e. the excess or shortfall of the comfortable corridor. While Figures 11 to 13 are representative of the data collected during pre-monitoring (Jun 2021-Dec 2021), Figures 14 to 16 are representative of post-monitoring (Mar 2023-Jul 2024).

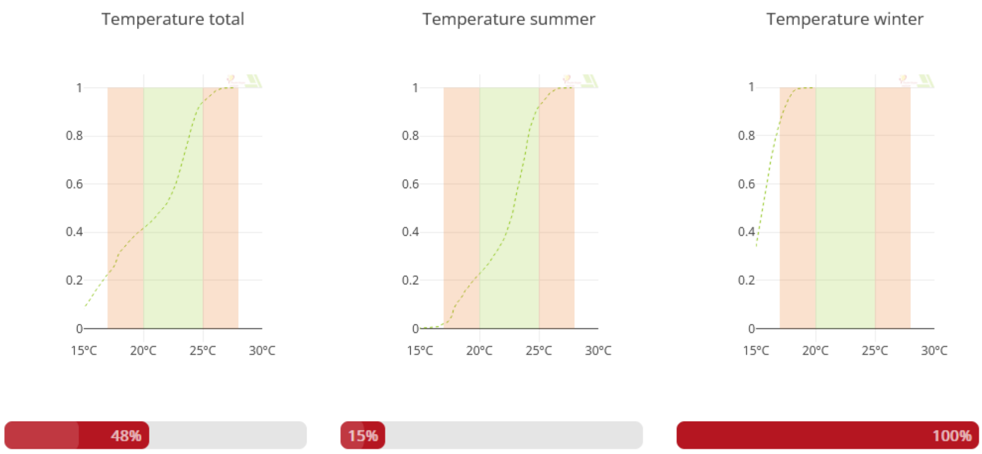

Figure 11: Relative threshold deviation (RTD) for temperature over the entire monitoring period before the refurbishment (measured in room "Room 1" in zone "Bajo_Izq"). Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

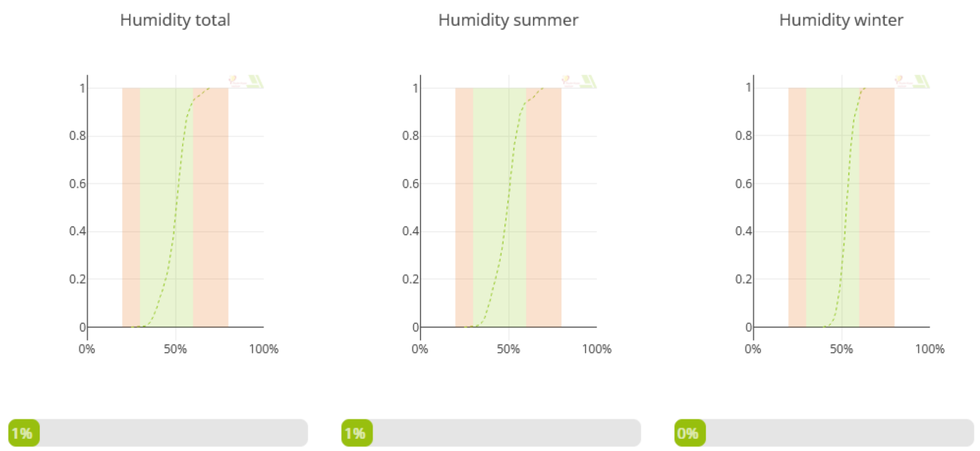

Figure 12: Relative threshold deviation (RTD) for relative humidity over the entire monitoring period before the refurbishment (measured in room "Room 1" in zone "Bajo_Izq"). Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

Figure 13: Relative threshold deviation (RTD) for CO_2 concentration over the entire monitoring period before the refurbishment (measured in room "Room 1" in zone "Bajo_Izq"). Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

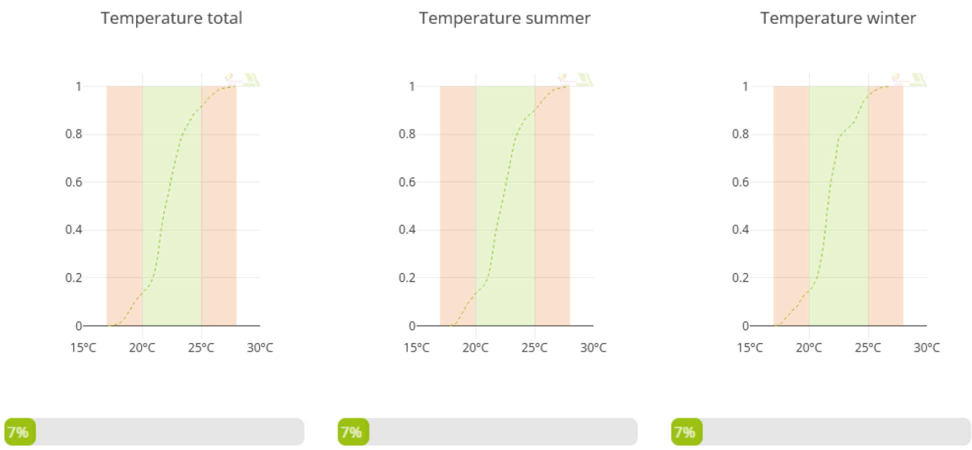

Figure 14: Relative threshold deviation (RTD) for temperature over the entire monitoring period after the refurbishment (measured in room "Room 1" in zone "Bajo_Izq"). Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

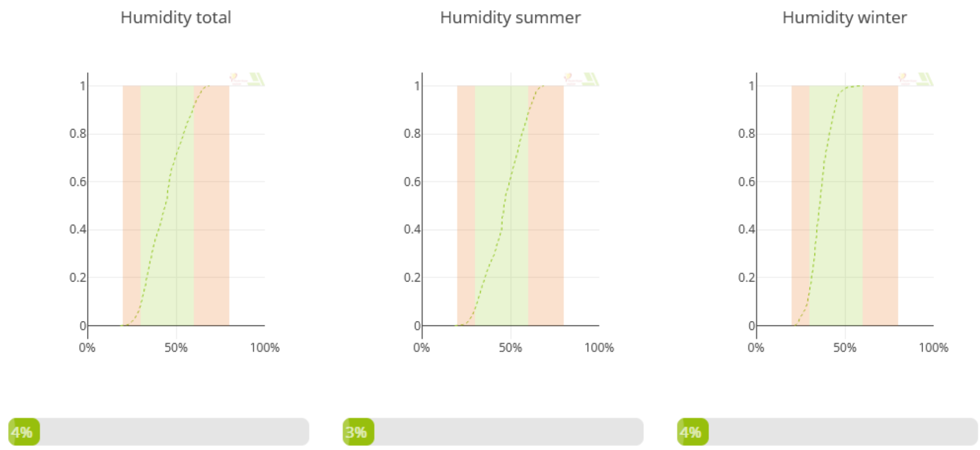

Figure 15: Relative threshold deviation (RTD) for relative humidity over the entire monitoring period after the refurbishment (measured in room "Room 1" in zone "Bajo_Izq"). Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

Figure 16: Relative threshold deviation (RTD) for CO_2 concentration over the entire monitoring period after the refurbishment (measured in room "Room 1" in zone "Bajo_Izq"). Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

PASSIVE HOUSE INSTITUTE (GERMANY)

Building Information

In addition to the microbiological examination for the Spanish case study, the building renovation in the Passive House standard is assessed in terms of microbiological evaluation from a preliminary project (cf. Figure 17). It is the first passive house in Darmstadt-Kranichstein (10.5 kWh/m²a) [5]. The indoor climate measurements were taken over a year (Jun 2020 - Jun 2021) at several locations in the building on different floors (attic, upper floor and ground floor). The outside climate was also logged. The data result from the period 25 years after the completion. The microbiological examination was carried out in Feb. 2016, this means outside the indoor climate measurement, which makes the assessment difficult in relation to the reference. Since no change in use occurred during this time, similar values are assumed. The building functions with a controlled ventilation system with highly efficient heat recovery.

Figure 17: "Passivhaus-Endhaus Kranichstein". Image source: Wolfgang Hasper, Passive House Institute.

5 https://passipedia.de/beispiele/wohngebaeude/mehrfamilienhaeuser/das_erste_passivhaus_in_darmstadt-kranichstein_deutschland, 07.2024

Data collection

The outdoor situation shows relatively high relative humidities of 75.7% (median) in the measurement period from June 2020 to June 2021 – see Figure 18 and Figure 19. Higher relative humidities are achieved in the winter months (Table 1).

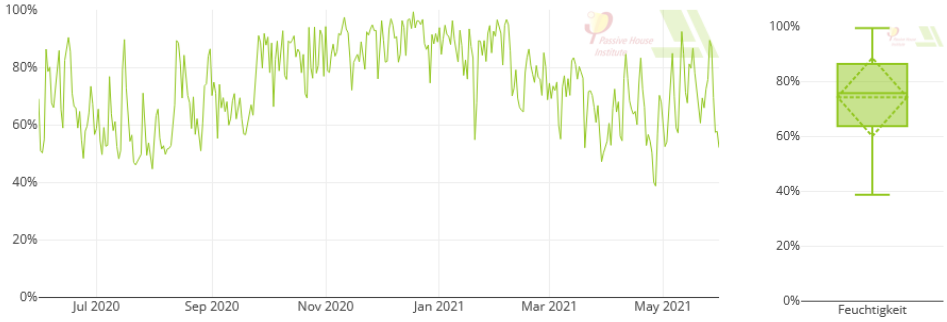

Figure 18: Outdoor Humidity for "Passivhaus-Endhaus Kranichstein", Part A, one value per day. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

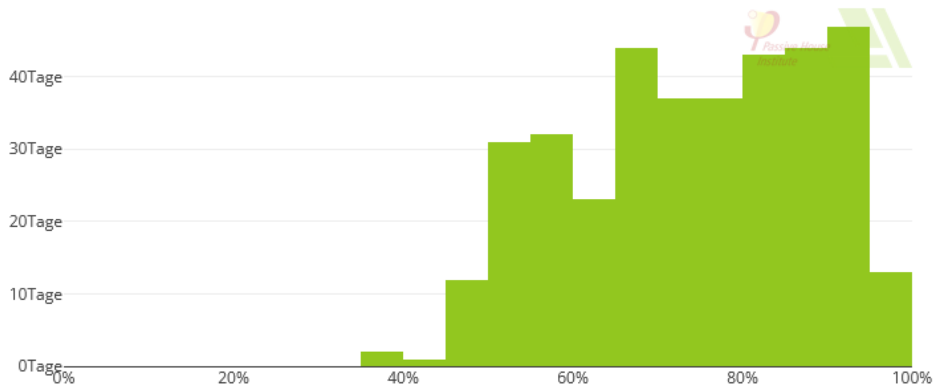

Figure 19: Outdoor Humidity for "Passivhaus-Endhaus Kranichstein", Part B, one value per day. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

Table 1: Outdoor Humidity for "Passivhaus-Endhaus Kranichstein" per month. Data source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

Month	Relative Humidity (Outside) in %
Jan	86.4
Feb	80.0
Mar	68.6
Apr	63.0
May	70.5
Jun	68.2
Jul	57.5
Aug	65.6
Sep	69.8
Oct	83.4
Nov	87.1
Dec	90.1

The outside temperatures are subject to strong fluctuations, especially in the summer-winter comparison (Max: 27.9°C, Min: -8.3°C) – see Figure 20 und Figure 21.

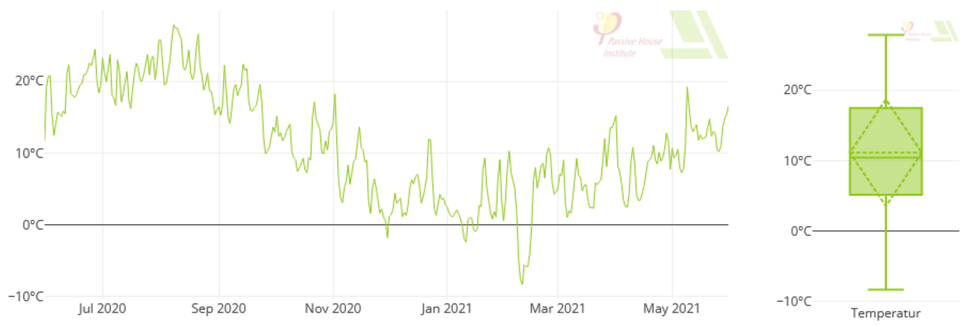

Figure 20: Outdoor Air-Temperature for "Passivhaus-Endhaus Kranichstein", Part A, one value per day. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

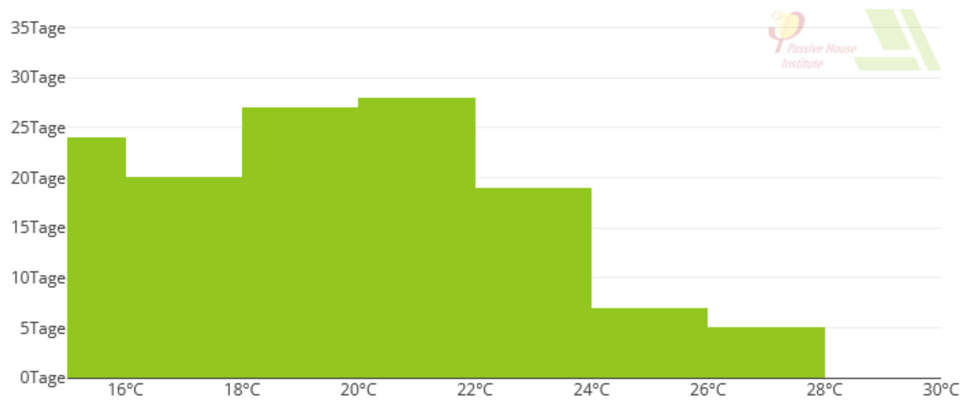

Figure 21: Outdoor Air-Temperature for "Passivhaus-Endhaus Kranichstein", Part B, one value per day. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

The average temperature inside (top floor) is 23.4°C ± 1.84 (details in Figure 23). The average relative humidity is 42.8% ± 7.4 (details in Figure 22). Humidity levels of over 60%, which are conducive to mould, are only reached on very few days. The average CO_2 concentration is 621.7 ppm ± 232 (details in Figure 24). Only at a few points in time are critical CO_2 concentrations of over 1500 ppm or even over 2000 ppm reached, which suggests good ventilation. It is therefore not surprising that the proportions of fungi and bacteria are within the normal range (Figure 25 and Figure 26). The values on the other floors hardly deviate. This is the microbiological reference in the outdoor situation (Figure 27). The values for the other floors are very similar and are therefore not listed separately here.

Figure 22: Indoor Humidity for "Passivhaus-Endhaus Kranichstein", attic, one value per day. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

Figure 23: Indoor Air-Temperature for "Passivhaus-Endhaus Kranichstein", attic, one value per day. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

Figure 24: Indoor CO_2 concentration for "Passivhaus-Endhaus Kranichstein", attic, one value per day. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

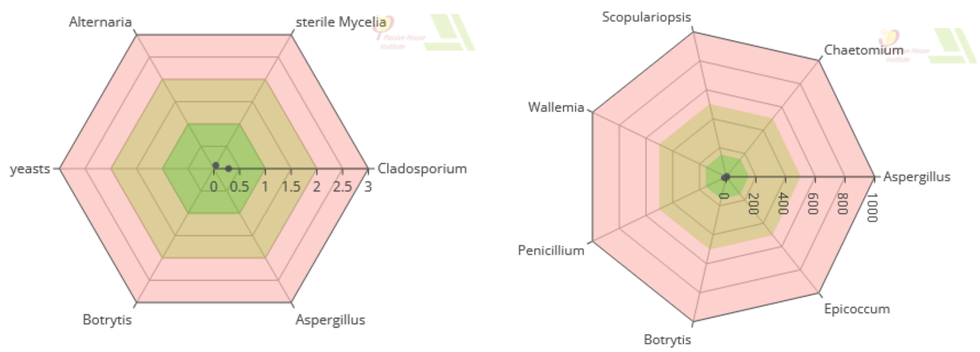

Figure 25: Indoor Microbiological Assessment for "Passivhaus-Endhaus Kranichstein", attic, Feb. 2016, Part A, unit: CFU. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

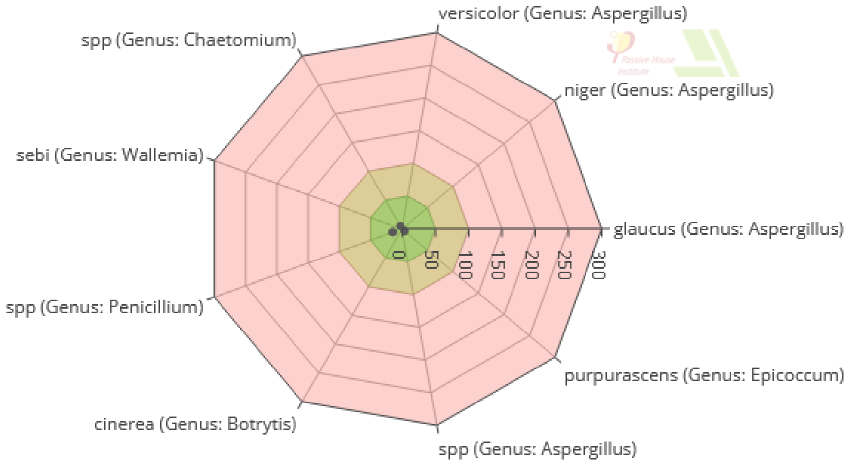

Figure 26: Indoor Microbiological Assessment for "Passivhaus-Endhaus Kranichstein", attic, Feb. 2016, Part B, unit: CFU. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

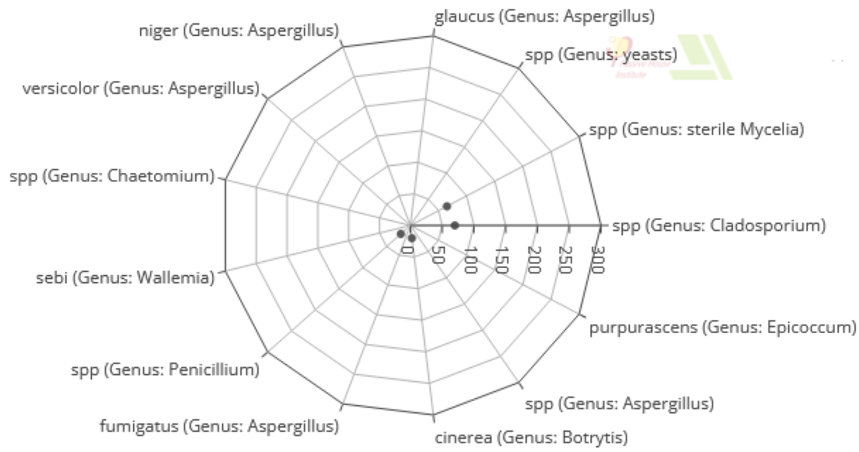

Figure 27: Reference Microbiological Assessment (Outdoor) for "Passivhaus-Endhaus Kranichstein", attic, Feb. 2016, unit: CFU. Image source: outPHit-Monitoring-Platform by Passive-House-Institute Darmstadt.

DISCUSSION

Identifying and quantifying microorganisms in the air helps to assess potential health risks, especially for children and people with respiratory diseases, allergies or weakened immune systems. This can be followed by appropriate analyses to identify critical moisture in building materials and take targeted remedial measures. The most appropriate one is to install a heat recovery system with supply and exhaust air. If it already exists, it can be used to optimize ventilation control curves in mechanical ventilation. Regular monitoring and sampling help to ensure that the IAQ is sufficiently guaranteed, with benefits for the comfort and health of the occupants and increased value of the property.

The work of Macher et al. shows that wind direction and relative humidity are good predictors of microbiological air concentrations. The concentrations of some bacteria and fungi are negatively related to temperature and wind speed and positively related to relative humidity [9].

Penicillium and Cladosporium species were consistently found in dust collected on the windowsill in the study by Macher et al. They were also detected in both investigations for this report, indicating their high natural presence.

Besides mould, which was the focus of investigation in this report, high indoor air humidity can create favourable growth conditions for house dust mites (faeces and decomposition products). Like mould, remedial measures include the subsequent installation of mechanical ventilation in the home too keep the level of indoor air relative humidity blow 55 %[6].

THE NEED FOR MECHANICAL VENTILATION

Ventilation and air conditioning systems are highlighted as a good technical solution to keep the IAQ high, free from pollutants. In addition, temperature and

[6] https://www.umweltbundesamt.de/hausstaubmilbe#vorbeugende-und-alternative-bekampfungsmassnahmen, 07.2024

humidity can be well controlled, the factors influencing the growth of bacteria and fungi [5]. The two case studies from the field measurements also show the importance of good ventilation (each with a controlled ventilation system with highly efficient heat recovery).

Mucha et al. investigated the influence of poor ambient air quality on indoor air quality (IAQ) in a single-family home. They also compared natural ventilation with mechanical ventilation. Overall, mechanical ventilation led to an improvement in many IAQ parameters. However, it is important to consider the filter performance for fine dust with a diameter of <2.5 µm. In addition, the installation of PM2.5 sensors to control the ventilation system during periods with high PM2.5 concentrations in the ambient air proves to be valuable to limit the ventilation air flow when it occurs. This is because under conditions of high PM2.5 concentrations in the outdoor air, mechanical ventilation can contribute to the deterioration of IAQ due to high infiltration rates [2]. In the IAQ context, this sensor-based control mechanism proves to be an advantage for ventilation systems compared to manual window ventilation.

LEGAL REQUIREMENTS

Regular sampling can serve as a success check and help identify problems such as air leaks, inadequate insulation or HVAC malfunctions before and after remediation. Promptly addressing these problems prevents energy inefficiencies and potential long-term damage to building systems.

Despite the advantages of the outPHit project, the sampling proved to be difficult to carry out, as there were little to no lab services to analyse the data, especially in some countries such as Greece. Therefore, more support, e.g. through funding, from the EU and at national level would be needed to support the development of the necessary infrastructure.

CONCLUSION

Energy-efficient building refurbishment aims to improve the quality of a building by reducing energy consumption and improving environmental sustainability. However, achieving these goals goes beyond simply improving thermal insulation or installing new windows. The improved insulation effect of the building envelope leads to greatly increased interior surface temperatures compared to unrenovated old buildings. Ultimately, this minimises the risk of mould and maximises thermal comfort. As this is directly linked to the physical properties of the building envelope, a great deal of attention is therefore required during the design phase of the refurbishment. To check afterwards whether the refurbishment measure was effective and whether the building's performance matches its design objectives, sampling of the indoor air and metrological monitoring of the surface temperatures can be used.

Microbiological sampling can identify specific types of bacteria or fungi and their concentrations, revealing potential health risks that cannot be detected by conventional indoor climate sensors for temperature, relative humidity, CO_2 and VOC concentration alone. Microbiological sampling is an important part of indoor air quality assessment. By identifying and quantifying various fungi, spores and bacteria, it provides important insights into the health and safety of indoor spaces and derives effective operational and remediation strategies.

Mould growth is often caused by high humidity due to inadequate ventilation. Exposure to bioaerosols has become a significant public health concern. The case studies emphasise the use of mechanical ventilation and high energy efficiency standards to minimise the risk of mould growth and ensure comfort and health requirements are met.

LIST OF FIGURES

REFERENCES

[1] Passive House Institute Darmstadt, 'Energiestandards', Passive House Institute - official website. Accessed: Jun. 18, 2024. [Online]. Available: https://passiv.de/de/03_zertifizierung/02_zertifizierung_gebaeude/04_enerphit/04_enerphit.htm

[2] W. Mucha, A. Mainka, and E. Brągoszewska, 'Impact of ventilation system retrofitting on indoor air quality in a single-family building', *Building and Environment*, vol. 262, p. 111830, Aug. 2024, doi: 10.1016/j.buildenv.2024.111830.

[3] S. G. Tringe *et al.*, 'The Airborne Metagenome in an Indoor Urban Environment', *PLoS ONE*, vol. 3, no. 4, p. e1862, Apr. 2008, doi: 10.1371/journal.pone.0001862.

[4] Sa. Bonetta, Si. Bonetta, S. Mosso, S. Sampò, and E. Carraro, 'Assessment of microbiological indoor air quality in an Italian office building equipped with an HVAC system', *Environ Monit Assess*, vol. 161, no. 1–4, pp. 473–483, Feb. 2010, doi: 10.1007/s10661-009-0761-8.

[5] E. Brągoszewska, I. Biedroń, B. Kozielska, and J. S. Pastuszka, 'Microbiological indoor air quality in an office building in Gliwice, Poland: analysis of the case study', *Air Qual Atmos Health*, vol. 11, no. 6, pp. 729–740, Jul. 2018, doi: 10.1007/s11869-018-0579-z.

[6] Z. Gizaw, M. Gebrehiwot, and C. Yenew, 'High bacterial load of indoor air in hospital wards: the case of University of Gondar teaching hospital, Northwest Ethiopia', *Multidiscip Respir Med*, vol. 11, no. 1, p. 24, Dec. 2016, doi: 10.1186/s40248-016-0061-4.

[7] P. Srikanth, S. Sudharsanam, and R. Steinberg, 'BIO-AEROSOLS IN INDOOR ENVIRONMENT: COMPOSITION, HEALTH EFFECTS AND ANALYSIS', *Indian Journal of Medical Microbiology*, vol. 26, no. 4, pp. 302–312, Oct. 2008, doi: 10.1016/S0255-0857(21)01805-3.

[8] W. Mücke and C. Lemmen, *Bioaerosole und Gesundheit: Wirkungen biologischer Luftinhaltsstoffe und praktische Konsequenzen*, 1. Aufl. Landsberg am Lech [Heidelberg]: Ecomed, 2008.

[9] J. M. Macher, F.-Y. Huang, and M. Flores, 'A Two-Year Study of Microbiological Indoor Air Quality in a New Apartment', *Archives of Environmental Health: An International Journal*, vol. 46, no. 1, pp. 25–29, Feb. 1991, doi: 10.1080/00039896.1991.9937425.

[10] D. Norbäck *et al.*, 'Endotoxin, ergosterol, muramic acid and fungal DNA in dust from schools in Johor Bahru, Malaysia — Associations with rhinitis and sick building syndrome (SBS) in junior high school students', *Science of The Total Environment*, vol. 545–546, pp. 95–103, Mar. 2016, doi: 10.1016/j.scitotenv.2015.12.072.

[11] O. Seppänen and W. J. Fisk, 'Association of ventilation system type with SBS symptoms in office workers: **SBS symptoms in office workers**', *Indoor Air*, vol. 12, no. 2, pp. 98–112, Jun. 2002, doi: 10.1034/j.1600-0668.2002.01111.x.

[12] N. N. Abdul Rahim, J. Jalaludin, E. S. Junaidi, and A. Sumantri, 'Association Between Air Microbiological Exposure With Sick Building Syndrome (SBS) among College Dormitory Students in Public University', *MJMHS*, vol. 19, no. s14, pp. 36–46, Dec. 2023, doi: 10.47836/mjmhs.19.s14.5.

[13] T. Verdier, M. Coutand, A. Bertron, and C. Roques, 'A review of indoor microbial growth across building materials and sampling and analysis methods', *Building and Environment*, vol. 80, pp. 136–149, Oct. 2014, doi: 10.1016/j.buildenv.2014.05.030.

[14] K. Sedlbauer, 'Vorhersage von Schimmelpilzbildung auf und in Bauteilen, Dissertation University of Stuttgart', Stuttgart, 2001. Accessed: Jul. 26, 2024. [Online]. Available: https://www.ibp.fraunhofer.de/content/dam/ibp/ibp-neu/de/dokumente/dissertationen/ks_dissertation_tcm45-30724.pdf

[15] H.-D. Neumann and M. Buxtrup, 'MVOC-Konzentrationen in Klassenräumen und mögliche Einflussfaktoren/MVOC concentrations in classrooms and possible influencing factors', *GrdL*, vol. 80, no. 06, pp. 234–245, 2020, doi: 10.37544/0949-8036-2020-06-24.

[16] C. J. Schwab and D. C. Straus, 'The Roles of Penicillium and Aspergillus in Sick Building Syndrome', in *Advances in Applied Microbiology*, vol. 55, Elsevier, 2004, pp. 215–238. doi: 10.1016/S0065-2164(04)55008-6.

[17] Z. Asadgol, A. Nadali, H. Arfaeinia, M. Khalifeh Gholi, R. Fateh, and M. Fahiminia, 'Evaluation Of Negative Air Ions In Bioaerosol Removal: Indoor Concentration Of Airborne Bacterial And Fungal In Residential Building In Qom City, Iran', Feb. 2018, doi: 10.5281/ZENODO.1316177.

[18] A. Hyvarinen, T. Reponen, T. Husman, J. Ruuskanen, and A. Nevalainen, ‘Characterizing Mold Problem Buildings - Concentrations And Flora Of Viable Fungi’, *Indoor Air*, vol. 3, no. 4, pp. 337–343, Dec. 1993, doi: 10.1111/j.1600-0668.1993.00017.x.

[19] A.-L. Pasanen, M. Niininen, P. Kalliokoski, A. Nevalainen, and M. J. Jantunen, ‘Airborne Cladosporium and other fungi in damp versus reference residences’, *Atmospheric Environment. Part B. Urban Atmosphere*, vol. 26, no. 1, pp. 121–124, Mar. 1992, doi: 10.1016/0957-1272(92)90044-S.

[20] S. Sampǒ and A. M. Luppi Mosca, ‘A study of the fungi occurring on 15th century frescoes in Florence, Italy’, *International Biodeterioration*, vol. 25, no. 5, pp. 343–353, Jan. 1989, doi: 10.1016/0265-3036(89)90014-6.

[21] W.-K. Jo and Y.-J. Seo, ‘Indoor and outdoor bioaerosol levels at recreation facilities, elementary schools, and homes’, *Chemosphere*, vol. 61, no. 11, pp. 1570–1579, Dec. 2005, doi: 10.1016/j.chemosphere.2005.04.103.

[22] A. K. Y. Law, C. K. Chau, and G. Y. S. Chan, ‘Characteristics of bioaerosol profile in office buildings in Hong Kong’, *Building and Environment*, vol. 36, no. 4, pp. 527–541, May 2001, doi: 10.1016/S0360-1323(00)00020-2.

[23] W. Feist, W. Ebel, S. Peper, W. Hasper, R. Pfluger, and M. Kirchmair, ‘25 Jahre Passivhaus Darmstadt Kranichstein’, Passive House Institute. Accessed: Jul. 26, 2024. [Online]. Available: https://passiv.de/downloads/05_passivhaus_kranichstein_25_jahre_endbericht.pdf

[24] I. Colbeck and C. Whitby, ‘Biological Particles in the Indoor Environment’, in *Indoor Air Pollution*, R. M. Harrison and R. E. Hester, Eds., The Royal Society of Chemistry, 2019, pp. 127–157. doi: 10.1039/9781788016179-00127.