Executive summary:

Healthy housing and good indoor air quality are important goals of public health. Biological indoor pollution due to dampness, moisture and mould is recognized as an emerging environmental health issue. Prevalence of dampness is remarkable, and may still increase due to demands of energy savings and extreme weather periods and floods associated with climate change. While the documentation is strong on associations between building dampness and health, the role of specific biological indoor pollutants in human health and disease is less well understood. Respiratory disorders, asthma, allergy and other immuno-mediated diseases are linked with biological indoor pollution, but the causal connections and underlying pathophysiological mechanisms are still mostly unclear. Efficient control and regulation are hampered by the insufficient understanding of these causalities. An adequate risk assessment of biological indoor pollutants is hampered by the lack of European exposure data and the documentation of the long-term health impacts of the existing exposure levels. There is an urgent need for European-wide knowledge to form a basis for establishing building-associated criteria for healthy indoor environments.

The aim of the HITEA study (Health Effects of Indoor Pollutants: Integrating microbial, toxicological, and epidemiological approaches) is to clarify the health impacts of indoor exposures on children and adults by providing comprehensive exposure data on biological and chemical factors in European indoor environments, and by combining this information with extensive health data obtained from a field study and from existing population cohorts. The focus is on microbial exposures due to dampness problems of buildings; in addition, the role of allergens, chemicals, cleaning agents, and poor ventilation is being studied. Modern microbiological, toxicological and immunological techniques are used to reveal the links between the harmful exposures and long term impacts on health, and the mechanisms behind. Data on determinants and distributions of indoor microbial agents are provided for the development of avoidance measures and other dissemination for stakeholders. The study networks experts on environmental epidemiology, microbiology, immunology, toxicology and building sciences, covering the multidisciplinary field needed for adequate risk assessment.

HITEA has implemented an extensive field survey in European schools, studying the role of the dampness-related multiple exposures in school environment for children's and teachers' respiratory health. This study provides data on short term health effects which may potentially lead to long term and chronic health impacts and investigates mechanisms underlying these exposure-health interactions. More than 9400 pupils and 650 teachers from 66 schools in Spain, The Netherlands and Finland took part in the initial phase of the study. The health status of a sub-population of pupils with asthma or asthma symptoms and teachers was followed in a longitudinal study over one school year, linking to detailed exposure assessments for multiple biological and chemical agents. During a first phase of disseminating the results of this study part, the project has reported on the prevalence and characteristics of dampness problems in schools across Europe, the occurrence of microbes

and microbial compounds in the school environment, and described the adverse effect of dampness and mould on respiratory symptoms in pupils and teachers. Several more reports are currently under preparation, exploring the effects and mechanisms of microbial and chemical exposures in the school environment on pupils' and teachers' respiratory health.

The second major part of HITEA integrates several ongoing European population cohorts and emphasizes on the long term health impacts of biological contaminants in the home environment in children and adults. HITEA provided a platform for the combined analyses of exposure and health data from children cohorts in Spain, Germany, The Netherlands and Finland. In addition, the adult cohort of the European Community Respiratory Health Survey contributed analyses to HITEA. The first scientific publications in this study part revealed associations of high microbial exposure indoors and immune development during infancy and report on the effect of environmental exposures in early life and neuropsychological development. Also, the use of household cleaning products during early life and the impact on children's respiratory health has been described. Extensive data have been provided on levels and determinants of microbial markers in homes across Europe.

Reporting and dissemination activities under HITEA are currently peaking, with multiple scientific publications being under preparation and anticipated to be released during 2013/2014. The progress of these activities can be followed on the project website http://www.hitea.eu.

Project Context and Objectives:

Abstract

Healthy housing and good indoor air quality are important goals of public health. However, biological indoor pollution due to dampness, moisture and mold is an emerging environmental health issue, as recognized in EU indoor air policy documents. Prevalence of dampness is remarkable, and may still increase due to demands of energy savings and extreme weather periods and floods associated with climate change. The exposure may lead to long-term impacts such as asthma. The documentation is strong on association between building mold and health, but the causative agents and disease mechanisms are largely unknown, which impedes recognition of a mould-affected patient in health care. Efficient control and regulation are hampered by the insufficient understanding of these causalities. Understanding of the links between building practices and health is lacking. There is an urgent need for European-wide knowledge to form a basis for establishing building-associated criteria for healthy indoor environments.

The aim of the HITEA study (Health Effects of Indoor Pollutants: Integrating microbial, toxicological, and epidemiological approaches) is to clarify the health impacts of indoor exposures on children and adults by providing comprehensive exposure data on biological and chemical factors in European indoor environments, and by combining this information with extensive health data obtained from a field study and from existing population cohorts. Modern microbiological, toxicological and immunological techniques are used that facilitate the revealing of the links between the harmful exposures and long term impacts on health, and the mechanisms behind. Data on determinants and distributions of indoor microbial agents will be provided for development of avoidance measures and other dissemination for stakeholders. The study networks experts on environmental epidemiology, microbiology, immunology, toxicology and building sciences. They cover the multidisciplinary field needed for adequate risk assessment. This approach has been successfully applied in the previous research.

Indoor air and environmental health regulation policies

Indoor environments include homes, schools, day care centers, offices, shopping centers and recreational facilities. They are diverse in their technical quality, size, location and lifestyle aspects and often beyond the control of traditional environmental regulatory tools. Therefore, control policies for indoor air pollutants are still to be developed. Implementation of this goal necessitates evidence-based practices which can only be achieved by focused research.

At present, adequate risk assessment of biological indoor pollutants is hampered by the lack of European exposure data and the documentation of the long-term health impacts of the existing exposure levels. Respiratory disorders, asthma, allergy and other immunomediated diseases are linked with biological indoor pollution but the causal connections and underlying pathophysiological mechanisms are still mostly unclear. The HITEA study aims to obtain answers to the open questions and thus provide knowledge to control the harmful exposures. Exposure data on microbial agents will be combined with health data from existing population studies in order to maximize the significance of the results and their impact. The study provides mechanistic information, especially on cytotoxicity, inflammation and oxidative stress in new field studies.

Biological and chemical agents as indoor pollutants

Biological agents include fungi and bacteria, their fragments and products, house dust mites and other allergens. Other major pollutant groups are volatile chemicals (VOC), semivolatile compounds (SVOC) and fine particles. Chemical agents include combustion products and various compounds from maintenance and cleaning. All these factors interact and have effects on humans although the causal links are not yet well understood.

The biological pollutants are commonly present; they have wide diversity and evident links to many common diseases, such as asthma and allergies. However, data about the typical exposure scenarios, both qualitative and quantitative, are lacking. Further interest on microbial exposures and their risk assessment is brought by their paradoxical role in relation to health, as microbes are not just harmful agents, but may also be protective from allergy. This has been suggested after observing a lower prevalence of allergy among farming children with high microbial exposures.

Dampness and health as an indoor air issue in Europe

Biological pollution due to indoor dampness is an emerging environmental health issue in Europe. Dampness and moisture damage are caused by failures in structures such as leakage on roofs or water pipes; condensation due to inadequate ventilation, or by occupant behaviour such as careless use of water. Water damage may also result from floods and accidental failures of water piping. Wetting of materials leads to microbial growth and biological indoor pollution.

Prevalence of dampness in buildings is remarkable; according to recent estimates 15-40 % in the housing stock. This may still increase due to additional insulation measures aiming at energy savings and by forthcoming extreme weather periods and floods associated with climate change. Indoor moisture and mould may often lead to costly remediation.

Association between building dampness and mould and human health has been extensively documented in homes, schools and other buildings. Most common are respiratory symptoms, but dampness is also a risk factor for asthma; both in onset of new asthma and exacerbation of symptoms. Intervention studies showing a positive effect of renovation on the occupants' health support the assumption of the causal link between dampness and health effects. Only part of the symptomatic individuals is atopic, mechanisms other than IgE-mediated allergy are involved. Common to all the health outcomes is the dampness and associated microbial growth as an etiological factor.

Mechanisms of the adverse health effects of biological pollutants

No single mechanism explains the wide variety of reported adverse health effects, but the responses may be allergic or irritative, or one of the toxic mechanisms may be involved: oxidative stress, cytotoxic, inflammatory or genotoxic response.

Activation of these mechanisms by exposure to microbial agents may lead to several health endpoints. IgE-mediated mold allergy is rare, but several fungal species have been associated with mold allergy, e.g., Aspergillus, Cladosporium and Penicillium. Microbial agents may also stimulate innate immune system triggering responses, and recurrent activation of the immune defence may lead to prolonged production of inflammatory mediators. Microbes or their toxins may have immunosuppressive effects mediated by toxic effects in the airways or other cells of the immune system.

Potential causative agents and their assessment

Dampness related exposures involves a number of organisms and agents: (a) fungi, bacteria, amoebae, house dust mites and other biological agents that proliferate due to excess water, and (b) chemical pollutants, including products of the microbiological activity (microbial volatile organic compounds (MVOCs), microbial secondary metabolites) or compounds released by chemical degradation of materials initiated by moisture. All these pollutants end up in the indoor air environment as a result of dampness, moisture damage or other presence of excess water. The diversity of the health outcomes of indoor dampness, moisture and mould suggest that the harmful exposures are multi-factorial and vary from case to case.

Exposure assessment to microbial agents in the indoor environment is a key issue in research of their health effects. Lack of specific biomarkers for these exposures makes exposure assessment complex and laborious. Components of microbial cells are often used as surrogates for microbial exposure. Endotoxin, a common surrogate of gram negative

bacteria, and (1-3) and (1-6)-ß-glucans, cell wall components of fungi, are causing airway irritation and inflammation, but their exposure levels in damp buildings may not explain all the effects reported. Other, specific constituents in the environmental microbial exposures, such as certain species or genera of bacteria or fungi, as well as their bioactive, secondary metabolites are less well studied in well-controlled epidemiological settings.

Assessing indoor microbial exposures is complex not only due to the multitude of possibly relevant exposures and their potential to interact. Epidemiological studies on health effects resulting from exposure to dampness and moulds are limited by the lack of valid methods to quantify exposure, which may contribute to the fact that qualitative assessment of visible water damage and mould has shown more consistent associations with health outcomes than quantitative estimates. Earlier studies have mostly focused on viable microbes using cultivation based techniques and short term samplings, while we know today that the reproducibility of such assessments is low and also non-viable microbial compounds are immunologically active and health relevant. Exposure to environmental microbes has rarely been considered over extended periods, but may – as other environmental exposures – be highly variable over time, so that a single measurement of short duration gives a limited or even wrong impression of human exposure. In general, timing (eg. childhood versus adulthood) and persistence of the exposure are highly relevant in addition to dose, when considering health outcomes. Sampling is a crucial factor in microbial exposure assessment, with different sample types being subject to different limitations, requiring careful selection of sampling approaches and considerate interpretation of results. The HITEA study represents a major effort to advance the knowledge in indoor microbial exposure assessment and provide more valid tools for future studies.

Specific objectives of the HITEA study

The overall aim of the HITEA study is to identify the role of indoor (biological) agents that lead to long term respiratory, inflammatory and allergic health impacts; children's health is emphasised in the project, and focus is put on microbial exposures due to dampness problems of buildings.

The major objectives in HITEA can be summarized as follows:

(1) to characterize the microbial, immunological, toxicological, and dampness-related properties of indoor air in school buildings in a new field study in three climatic regions across Europe

(2) to study the associated long term respiratory, inflammatory and allergic health impacts on children and adults

(3) to study the short term health effects, potentially leading to long-term impacts, associated with indoor air exposures in schools with special emphasis on markers of inflammation

(4) to identify causal links by combining measured exposure characteristics to the observed health effects among children and adults in ongoing European child cohorts and an existing adult cohort

(5) to reveal the mechanisms underlying the health effects

(6) to apply new and innovative approaches in exposure assessment

(7) to develop guidelines for health professionals on how to study indoor microbial pollution.

Project Results:

1. Overall strategy and general description of the HITEA study

The essential elements of the strategy and the approach followed in the HITEA study are:

a) to focus on those indoor pollutants and health effects where information is most urgently needed;

b) to deal with indoor air issues that have European-wide relevance and environmental health importance;

c) to focus on a particular vulnerable population group, i.e., children;

d) to exploit previously developed technologies, methodologies and design approaches to obtain the maximum productiveness in achieving the goals of the study; and

e) to maximize the possibilities to find the links between exposing agents and health by exploiting the previously established population cohorts.

The HITEA study networks experts on environmental epidemiology, microbiology, immunology, toxicology and building sciences that together cover the multidisciplinary field needed for adequate risk assessment. With these strategic choices, we have developed a study which will provide European-wide knowledge on the health effects of biological indoor exposures at reasonable costs.

The HITEA study consists of three closely linked parts –the interdependencies of the components and the organisation of the work in 11 work packages.

Indoor Air in Schools:

In this first part, the study focuses on dampness and moisture problems and associated exposures to biological agents in schools across Europe. An extensive field study in schools was implemented during two school years in identical set-ups in Finland, The Netherlands and Spain to reveal the role of the dampness-related multiple exposures in school environment for children's and teachers' respiratory health. This study provides data on short term health effects which may potentially lead to long term and chronic health impacts.

Indoor Air in Homes:

This second part, the framework of which consists of several European population cohorts, emphasizes on the long term health impacts of biological agents in children and adults. Exposure data from different time points during life are combined with the comprehensive information on children's and adults' health already collected in these cohorts. Here, HITEA integrates on-going children cohorts from Spain (INMA-Menorca cohort), The Netherlands (PIAMA), Germany (LISA) and Finland (LUKAS), as well as the European Community Respiratory Health Survey (ECRHS) adult cohort.

Methods and Mechanisms:

The third part organizes the carrying out of extensive exposure assessment to biological and other agents of interest in indoor environments in several regions of Europe, including schools and homes (for study part one and two, respectively). Existing sample materials from on-going population cohorts are exploited, and follow-up sampling campaigns are organized in a concerted manner, so that comparable data are being produced. The laboratory analyses focus on measurement of microbial components from dust samples, and include both previously validated parameters of microbial exposure, such as endotoxin and ß-glucans, as well as novel technologies, including DNA based approaches and multi-target analyses for microbial toxins. This part also involves toxicological characterization of the multiple exposures in dust samples collected from schools, focusing especially on markers of cytotoxicity, inflammation and oxidative stress, and linking to immunological parameters measured from exposed individuals for immuno-toxicological studies and mechanistic conclusions. The contributions of this part of the study are specified in the following description under "Indoor Air in Schools" and "Indoor Air in Homes".

2. Indoor Air in Schools

2.1 The HITEA field study in schools

The HITEA school study is a longitudinal field survey that was conducted in schools in three climatic regions in Europe, focusing on dampness and moisture problems and associated exposures to biological and chemical agents. A major achievement during the first half of the project was the implementation of this study in identical set-ups and during the same time period in three European countries - Finland, The Netherlands and Spain -, representing three different climatic regions. Meeting an ambitious time-schedule, the extensive field study covering several phases and cross-sectional as well as longitudinal aspects was successfully concluded according to plan, including the environmental survey and sampling, the respiratory health assessments, and centralized data management. In the following, the study phases are detailed.

In the initial phase of the study, a questionnaire survey was conducted to assess moisture damage/dampness problems and other indoor air quality issues in primary schools in the three study countries (phase 0). The screening questionnaire was first developed in English and then translated into Spanish, Catalan, Dutch, and Finnish, using an Internet-based platform. The questionnaire contained ten key questions on current and past dampness, moisture, and mould observations and also collected background information on building and ventilation characteristics and renovation plans. The questionnaire was addressed to the principals of the schools, with support - where needed - from the person responsible for school building maintenance. Using this questionnaire, information from 236 school buildings in the three countries were collected. Information on additional school buildings was supplemented from earlier conducted questionnaire surveys including the same of similar questions in Spain and in Finland.

Of the schools that participated in the screening phase, between 20 and 24 schools per country were selected for on-site building inspections (phase Ia; summer 2008). Approximately half of these represented schools that did not report any signs of dampness problems, moisture or water damage, or mould growth in the building nor a history with such problems ('reference schools'); the other half represented schools with such problems that were considered severe (i.e. either widespread, and/or located in classrooms, and/or including reports of visible mould; 'index schools'). The inspections of in total 66 schools (95 school buildings) included walkthroughs utilizing pre-designed checklists and non-destructive measurements by trained research personnel. The research personnel were centrally trained by senior members of the research group with extensive experience in building investigations. Complementing the visible observations, measuring instruments included hand-held moisture detectors, relative humidity, temperature and CO2 monitoring devices.

Based on the location, extent and severity of moisture damage/dampness/visible mould observations derived from the school building inspections, schools were categorized ad case or control schools and selected for subsequent phases of the study. The methodology of both screening questionnaire, and selection of study schools based on walkthrough building inspections have been published in a first paper from the HITEA school study by Haverinen-Shaughnessy et al. (2012).

In autumn 2008, phase Ib facilitated the administration of an extensive respiratory health questionnaire to pupils and teachers in a minimum of eight moisture-damaged (case) and eight non-damaged (control) schools per country. For pupils aged 6-12 years, the parent administered questionnaire assessed the respiratory health status using questions from the validated International Study of Asthma and Allergies in Childhood, translated into three

languages. Information from more than 9400 pupils from 57 schools in Finland, The Netherlands and Spain was collected (Borras-Santós et al., 2013). The questionnaire developed for teachers included items on demographic characteristics, relevant exposures and respiratory health, partly based on the European Community Respiratory Health (ECRHS) protocol. In total, more than 650 teachers from 55 schools in the three countries provided information on their respiratory health in this phase (Zock et al., manuscript under preparation).

From all schools categorized as case or control schools, a minimum of 4 case and 2 control schools were selected per country, with emphasis on biggest possible contrast in observations on moisture damage, dampness and visible mould in the school buildings. In total, 23 schools were defined for more detailed exposure and health characterization in the subsequent study phases. During phase II in late winter/early spring 2009, lung function measurements were conducted in all consented pupils from the study schools, yielding lung function tests from more than 3500 pupils in the three countries. Measurements from 2736 children were considered valid and reproducible and were used for respiratory health analyses (Jacobs et al., manuscript submitted).

Phases III to V represented a longitudinal health and exposure assessment in the selected study schools. The first two of these phases were conducted before and immediately after the school vacation, to define the effect of the school exposure on pupils' and teachers' health. Lung function testing, exhaled nitric oxide measurements and health diaries were completed for more than 500 pupils with symptoms indicative for asthma. Data on sickness absence and school performance were collected where available. In addition to pupils, also 175 teachers from these schools participated in the health study, providing - in addition to lung function and lung inflammation measurements - peripheral blood and nasal lavage samples for determination of markers of inflammation and immunological parameters. Exhaled breath condensate was collected from a subset of Finnish teachers to follow changes in inflammatory markers during one school week.

In parallel to the longitudinal health assessment, extensive exposure assessments and sample collection campaigns were performed during phases 2, 3 and 5 in the study schools. The focus in these assessments was on biological agents that were measured from airborne settled dust that was collected in more than 1000 locations in the 23 study schools over three assessments. In addition, selected chemical and physical parameters were monitored in the HITEA schools, including sampling for PM2.5 and NO2, monitoring of CO, CO2 (for modelling of ventilation rates), temperature and relative humidity. Outdoor environmental characteristics as well as cleaning procedures were assessed via questionnaire.

2.2 Dampness, microbial and chemical exposures in schools

This chapter summarizes findings from the first analyses that were done based on data created under HITEA; statistical analyses are still on-going and additional results will be published during 2013 and 2014.

Moisture problems in schools in three climatic regions of Europe

An initial analysis in the HITEA school study investigated the 'Occurrence of moisture problems in schools in three countries from different climatic regions of Europe based no questionnaires and building inspections' (Haverinen-Shaughnessy et al., 2012). Based on data obtained from phase 0 and Ia , an assessment was performed utilizing both questionnaires and on-site building investigations, and the agreement between these two methods was evaluated for validation purposes. Based on the questionnaire assessment, different types of moisture problems were reported in 24-47% of all school buildings at the time of the study. In the Dutch schools, dampness was most commonly reported, moisture damage in Spanish schools, and mould odour in Finland. Upon on-site building inspections by trained research staff in a sub-sample of schools from the questionnaire survey, a good overall agreement between the questionnaire and inspection data was found (kappa-value 0.62), however, large differences (0.39-0.91) were observed between countries. Extrapolating from the inspection data, the minimum estimates for prevalence of moisture problems in school buildings are 20% in the Netherlands, 41% in Spain, and 24% in Finland. Moisture problems, such as moisture damage, dampness, and visible mould, were concluded to be relatively common in schools. A marked variation between occurrence and severity was found across geographical areas, which may partly be explained by building characteristics.

On the basis of this study, questionnaire-based surveys can be used to assess moisture problems in school buildings, but because of large variation in agreement with inspection data, the questionnaire needs to be validated by on-site inspection in a subsample of the surveyed buildings (Haverinen-Shaughnessy et al., 2012).

Microbial secondary metabolites in school buildings

Secondary metabolites produced by fungi and bacteria are among the potential agents that contribute to adverse health effects observed in occupants of buildings affected by moisture damage, dampness and associated microbial growth. However, few attempts have been made to assess the occurrence of these compounds in relation to moisture damage and dampness in buildings. This study within the HITEA school study, conducted in the context of

the school building inspections (phase Ia) aimed at providing systematic information on the prevalence of microbial secondary metabolites in a large number of school buildings in three European countries, considering both, buildings with and without moisture damage and/or dampness observations (Peitzsch et al., 2012). In order to address the multitude and diversity of secondary metabolites a large number of more than 180 analytes was targeted in settled dust and surface swab samples using liquid chromatography / mass spectrometry (LC/MS) based methodology.

While 42%, 58% and 44% of all samples collected in Spanish, Dutch and Finnish schools, respectively, were positive for at least one of the metabolites analyzed, frequency of detection for the individual microbial secondary metabolites - with the exceptions of only few compounds - was low, typically in the range of and below 10% of positive samples. In total, 30 different fungal and bacterial secondary metabolites were found in dust samples from schools. A major finding in this study was that settled dust derived from moisture damaged/damp schools contained larger numbers of microbial secondary metabolites at higher levels compared to respective dust samples from schools not affected by moisture damage and dampness. This observation was true for schools in each of the three countries, but became statistically significant only when combining schools from all countries and thus enlarging sample number in the statistical analyses.

This study concluded that a multitude of different microbial secondary metabolites, often referred to as 'microbial toxins', are part of the complex exposures encountered in indoor environments. The results suggest that moisture damage and dampness observed in school buildings link to an increase in the number of different mycotoxins present at elevated levels in settled dust; however, it is evident that microbial toxins are also present in non-damaged buildings as part of the 'normal' microbial flora indoors (Peitzsch et al., 2012). This extensive work lays the grounds for future attempts to measure indoor exposure to microbial toxins and to assess the health risks related to such exposure.

A follow-up analysis of data derived from the longitudinal study in HITEA schools confirmed the geographical dependency of indoor microbial metabolite patterns and added also indications of a seasonal effect on the occurrence of microbial metabolites indoors (Sulyok et al., manuscript under preparation).

Microbial exposures in HITEA schools

Extensive efforts in assessing exposure in the HITEA study schools during the longitudinal assessment focused on microbial agents. An earlier HITEA add-on study revealed that

endotoxin exposure in schools is high compared to home exposure and thus may contribute to the total environmental endotoxin exposure of pupils and teachers (Jacobs et al., 2012). Here, endotoxin was assayed from airborne settled dust from more than 1000 locations in 23 study school during three assessments; 645 measurements from 237 classrooms were used for the analyses (Jacobs et al., manuscript submitted). Significant difference in endotoxin levels were observed between counties, with highest levels reported for Dutch schools. Differences were also apparent within each country, between schools and between repeated samplings (sampling period). The factors that were found to determine endotoxin levels in classrooms varied between countries, but included age of the pupils (higher endotoxin levels in lower grades) and occupancy, to name the two strongest factors. The study concluded that single endotoxin measurement could provide a reasonable estimate for overall endotoxin exposure of pupils in schools and confirmed the earlier finding that school exposure may heavily contribute to total endotoxin exposure of pupils and teachers.

Focusing on molecular approaches in the microbial exposure assessment from settled dust, including DNA-based methodology, we confirmed the large geographical differences in prevalence and levels of microbial exposures in settled dust collected in schools in Spain, the Netherlands and in Finland (Täubel et al., manuscript under preparation). Exposure levels in this study were found highest for all fungal markers and general bacterial markers in Spanish schools, and lowest in Finnish schools for all of the microbes measured. The fungal and bacterial levels varied strongly and in many cases significantly in the school buildings between repeated measurements over one year. This variation could only be partly explained by the influence of season on the microbial content in schools. Moreover, changes of microbial levels over time were not consistent between countries for the different microbial groups measured, indicating that country-specific characteristics of the school buildings and their use and maintenance may be major driving factors for indoor microbial exposures. We saw some indications for clustering of source-related microbial markers in the correlation analyses. Intraclass correlations and thus reproducibility were poor for almost all of the microbial targets and indicated that microbial exposure with respect to specific microbial species/genera/groups in schools is highly variable over time and space. We observed big differences between countries with respect to how moisture and dampness conditions in school buildings affect the microbial levels measured, with a clear impact in Dutch schools, visible for some markers in Spanish schools and not visible for Finnish schools included in this study.

Immunotoxicological potential of dust samples from schools

The immunotoxicological potential of settled dust samples collected in the HITEA study schools was assessed to compare the results on the immunotoxicity of the indoor air particles both to the health status of the study subjects and information about the technical condition of the sampled building/area. The statistical analyses for this work are on-going

and have partly been presented at scientific conferences (Huttunen et al., manuscript under preparation).

The production of selected inflammatory mediators was measured in mouse macrophages. According to preliminary analyses of the results, samples collects from Spanish and Dutch schools cause clearly higher inflammatory response than the samples collected from Finnish. When the results were compared on the basis of school status (moisture damage / reference schools), the samples collected from moisture damaged schools in Spain and the Netherlands caused higher inflammatory response compared to reference schools, whereas in Finland there was no significant difference between the schools. Very similar results were obtained in the evaluation of various toxic responses upon exposure of mouse macrophages to indoor dust. Microbial components in the dust samples were measured in parallel to the immunotoxicological studies. Bacterial and fungal components (endotoxin and ergosterol, respectively) correlated significantly with both toxicity and the ability to induce inflammatory response. The correlation between toxicity and fungal ergosterol was strongest among the samples collected from damaged schools, whereas the correlation between toxicity and endotoxin was strongest within the reference schools (Huttunen et al. 2013).

Exposure to selected chemical agents in HITEA schools

The HITEA-project focuses on the evaluation of health effects of biological agents, but also considers selected chemical exposures in primary schools. Preliminary work was done to characterize exposure to fine particulate matter (PM2.5), absorbance (soot), NO2, CO2 and cleaning products in schools in Spain, The Netherlands and Finland. Indoor PM2.5, soot, NO2 and CO2 were measured during 5 consecutive school days in two periods: during winter/spring 2009 and winter/spring 2010 (phases 2 and 5). Information on frequency of cleaning and the use of cleaning products was obtained by a short questionnaire.

Average soot and NO2 concentrations were highest in Spain, intermediate in the Netherlands and lowest in Finland. PM2.5 was comparable in Spanish and Dutch schools and lowest in Finland. These results are likely to link to higher traffic and diesel related traffic loads in the Netherlands and Spain, in comparison to Finland.

CO2 as measured during school hours reached higher levels in Spain as compared to the Netherlands and Finland; there is an indication of less ventilation in Spanish and Dutch classrooms compared to Finnish classrooms, however, evaluation of air exchange rates based on CO2 logged data from classrooms in these three countries are on-going.

Frequency of cleaning was found to be comparable between countries; however the number of hours spent cleaning in each school differed by country. The most commonly used cleaning products were multi-use cleaning products in all three countries, whereas hypochlorite bleach was only used commonly in Spain.

This first crude analysis concluded that PM2.5, soot, NO2 and CO2 levels in schools differed considerably between countries and need to be taken into consideration in the analyses on respiratory health effects of exposed pupils and teachers in the HITEA study schools.

2.3 Health effects of exposure to dampness, microbes and chemicals agents in the school environment

The majority of analyses on health effects following exposure to dampness, microbes and chemical agents in HITEA study schools are currently on-going; scientific publication of study results are expected for mainly for 2013/2014. The following chapter summarizes first results from the statistical analyses and preliminary findings.

Dampness and moisture problems in schools and respiratory health in pupils and teachers

While adverse respiratory health effects of dampness and mould in the home have extensively been reported, these effects have much less bee studied in the school environment. The HITEA study evaluated associations between dampness and mould in school buildings and respiratory symptoms among 6 to 12 years old pupils in Finland, Spain and The Netherlands (Borràs-Santos et al., 2013). Questionnaire information from 9271 pupils from 56 schools were used and related to observation of dampness, moisture damage and visible mould made during standardized school building inspections (phase Ia). Country specific associations between moisture problems and parent-reported respiratory symptoms were explored. Dry cough at night was associated with moisture damage in schools in all three countries. Finnish children attending a moisture damaged school had significantly more often wheeze, nasal symptoms and respiratory related school absence, but these associations were not seen in Dutch or Spanish school children. The study concluded that moisture problems in schools may cause adverse respiratory effects in pupils; Finnish pupils were found to be at higher risk for adverse health outcomes due to dampness problems in schools (Borràs-Santos et al., 2013).

Analysis on dampness and mould in school buildings and respiratory symptoms in teachers is on-going (Zock et al., manuscript under preparation). Preliminary analyses of self-reported respiratory symptoms in 645 teachers from 53 schools in three European countries showed an overall tendency of more respiratory symptoms in teachers from schools with moisture damage. The analysis revealed a consistent increase in nasal symptoms in teachers working in moisture damage schools in all three countries. Dutch and Finnish teachers from moisture damaged schools had on average higher asthma symptom score and reported more work-related respiratory symptoms. In conclusion of this study, teachers working in schools with moisture problems have a higher risk for upper and lower respiratory tract symptoms, but not for respiratory infections. This work underlines the public health relevance of damp and mould problems in indoor workplaces.

Microbial exposures and respiratory health in pupils and teachers

A first, cross-sectional study was carried out to explore the associations between school dampness, microbial exposures and respiratory health in children (Jacobs et al., manuscript submitted). Respiratory health of pupils from 25 schools in Finland, The Netherlands and Spain was assessed via questionnaire and lung function measurements (phase II). In total, 3843 pupils provided respiratory health questionnaire data, and 2736 pupils provided acceptable, good quality lung function measurements. Settled dust was repeatedly sampled in 232 classrooms and levels of 14 different microbial markers were determined. Earlier found associations between respiratory symptoms and school dampness (Borràs-Santos et al., 2013) were confirmed in this study; however, no significant effects on pupils' lung function were found. The observed effects were strongest in Finland, although microbial levels in Finnish schools were generally much lower compared to Spanish or Dutch schools. The results of this study indicate that the associations between moisture, microbial exposure and health may vary between countries and particular exposure patterns may be relevant. Associations between microbial exposure and health were unclear.

Earlier, and add-on study to HITEA explored associations of endotoxin in schools and in the home in relation to asthma and sensitization (Jacobs et al., 2012), since population studies that would include microbial pollutant exposure in both indoor environments are more or less non-existent. Pupils from 10 Dutch schools with and without moisture problems were followed to their homes; case and control pupils were selected based on asthma-like symptoms of the children. This study found clearly higher endotoxin exposure in classrooms compared to homes. The results of this study suggested that endotoxin in schools may be associated with non-atopic asthmatic symptoms in pupils, although the results require reproduction because of the modest sample size (Jacobs et al., 2012).

Referring to the analyses of self-reported respiratory health in teachers described above (Zock et al., manuscript under preparation), associations were studied using total load of microbial secondary metabolites (often referred to as 'microbial toxins') in settled dust of the study schools. While in Spanish and Dutch teachers there were no clear trends in asthma symptom score or prevalence of nasal symptoms, Finnish teachers from schools with higher microbial toxins levels were more likely to have nasal symptoms in the previous year and there was some indication that also lower respiratory tract symptoms were more prevalent with increasing toxin load. These preliminary findings indicate a potential role of fungal toxins in explaining some of the adverse respiratory health outcomes in teachers working in schools with moisture problems. More detailed, longitudinal analyses on the exposure of teachers to dampness and microbial agents and the effects on respiratory health, assessed via objective measures (i.e. lung function, inflammatory markers in blood, nasal lavage and exhaled breath condensate) are currently being conducted (Zock et al., manuscript under preparation).

Exposure to chemical and physical parameters in schools and association with respiratory health

Using respiratory health questionnaire data from 3877 school children and information on indoor concentrations of PM2.5, soot, NO2 and CO2, as well as cleaning policies in the 25 study schools, associations between chemical exposures in the school environment and respiratory symptoms were explored. As mentioned above, chemical pollutant levels were lowest in all cases in Finnish schools, and highest in Spain for soot and NO2, and comparable between Spain and Netherlands for PM2.5. Initial analyses of these data indicated a tendency for PM2.5, soot and NO2 levels to be associated with an increased risk of having respiratory symptoms in the Netherlands and Finland, but inverse associations with respiratory symptoms were found in Spain. We concluded that chemical agents in the study schools may play a role in occurrence of potential respiratory health effects of occupants and need to be taken into consideration in future analyses. In addition, the effect of ventilation as well as of cleaning agents on respiratory health will be explored.

3. Indoor air in homes

3.1 Microbial exposures and health in the ECRHS adult cohort

The European Community Respiratory Health Survey (ECRHS, 1990-1993) was designed as a multicentre study to estimate variations in the prevalence of asthma, asthma-like symptoms, airway responsiveness, and allergy and their known or suspected risk factors in

adults living in Europe (Burney et al., 1994). In 1999-2001 a random and a symptomatic samples of the participants were followed-up as a multicenter cohort (ECRHS II) (2002) in 29 European centres to determine the incidence of and risk factors for the development of allergic disease, atopy and rapid loss of lung function. 22 centres from 10 European countries agreed to take part in a detailed assessment of home exposures in the course of ECRHS II, including collecting dust samples from mattress. In the HITEA project, a total number of 999 house dust samples from the ECRHS II random sample of all 22 participating study centres were randomly selected for the analyses of microbial agents in house dust. The ECRHS II cohort contributed extensive health and exposure data to the HITEA project to facilitate evaluation of microbial agents as risk factors for allergic diseases in adults.

An initial analysis from Chen et al. (2012) studied the geographical variation and the determinants of domestic endotoxin levels in mattress dust in Europe. Endotoxin exposure is known to have manifold effects on human health. Results from this analyses revealed that high summer temperature, cat or dog keeping, a high household crowding index, and visible damp patches in the bedroom were significantly associated with a higher endotoxin concentration. The study found a large variability of domestic endotoxin level across Europe, partly linking to meteorological determinants of endotoxin in house dust. The observed variation needs to be taken into account when evaluating health effects of endotoxin exposures in international contexts.

In a second effort to characterize the microbial exposures in mattress dust from homes across Europe in more detail, several bacterial and fungal species or groups were measured with DNA based methodology. The analysis revealed a large variation in concentrations of microbial levels across Europe, with major variations of up to 30-fold median levels observed for fungal groups (Cladosporium herbarum, Penicillium spp./Aspergillus spp. group). Levels of bacterial groups (Mycobacterium spp., Gram positive and Gram negative bacteria) varied as well, but typically showed less than 10-fold difference in median concentrations. When exploring determinants of microbes in mattress dust, some home characteristics indicating dampness/moisture problems (such as damp spots on surfaces, condensation of moisture in the bedroom), as well as the season of sampling and having the bed in the living room had significant effect on fungal levels. In addition to some of the same determinants, bacterial levels were affected by the age of the home and mattress, the presence of a cat or dog at home, and increased with reported indoor smoking.

In a large epidemiological study in 972 adults of the ECRHS II cohort, association of endotoxin levels in mattress dust with respiratory health in adults were explored (Bakolis et al., 2012). This study concluded that there was no evidence that mattress endotoxin level was associated with respiratory symptoms or IgE sensitisation; however, the study found evidence that the association of lung function with mattress endotoxin may be modified by certain genetic factors (i.e. the CD14/-260 genotype).

An on-going analysis in the same study population will provide information on the distribution and the health effects of current exposure to fungal and bacterial species in mattress dust, determined by molecular techniques (Tischer et al., manuscript under preparation). The study shows that higher exposure to fungal and bacterial components assessed via DNA-based, quantitative PCR technique in mattress dust from 22 study centres throughout Europe was positively associated with shortness of breath and with experiencing at least two asthma symptoms within the past 12 months. These results warrant further research, particularly with respect to sources of bacteria and fungi indoors and possibly distinct effects on human health.

One dedicated effort in HITEA dealt with revealing the microbial community composition in house dust of asthmatic and non-asthmatic adults of the ECRHS II cohort, using various molecular, cultivation-independent techniques (Valkonen et al., manuscript under preparation). Mattress dust samples of 200 asthmatics and 200 control subjects from 14 European centres of the ECRHS II study were analysed for nine fungal or bacterial taxa using quantitative PCR (qPCR), as well as for quantitative markers for fungal and bacterial content in the house dust. In addition, DNA fingerprinting technique was applied to illustrate bacterial community profiles. Some of the measured bacterial and fungal markers showed a trend to be protective or a risk factor for asthma, however, none of these associations was statistically significant. Similar results were seen for cough, atopy and lung function measurements. Statistically significant differences in microbial profiles in mattress dust of asthmatics versus non-asthmatics were visible when comparing bacterial community profiles, where several bacterial signals could be associated with an increased risk for asthma. The work on following up these indicative findings is on-going and will be on-going beyond the lifetime of the HITEA project.

3.2 Exposure to microbes and chemicals and children's health

As described under point 1 in the general strategy of HITEA, the studies on children's health integrate several on-going, European population cohorts, emphasizing on exploring the long term health impacts of exposure to biological agents in children. HITEA umbrellas children cohorts from Spain (INMA-Menorca cohort), The Netherlands (PIAMA), Germany (LISA) and Finland (LUKAS). An inventory of existing health data in four children cohorts and the creation of a database containing common variables allowed the combined statistical analysis of LISA (Ge), INMA (Sp) and PIAMA (Nl) cohorts; the Finnish LUKAS cohort was handled separately in the health analyses due to differences in timing, age of the study participants and characteristics of the cohort. The following overview comprises information retrieved from statistical analyses in the different cohorts so far.

An initial effort to describe microbial exposures in the different cohorts was published under the title "Endotoxin, extracellular polysaccharides, and beta(1-3)-glucan concentrations in dust and their determinants in four European birth cohorts; results from the HITEA project" (Casas et al., 2013 (1)). It is known that geographical differences in the prevalence of asthma and allergy exist, but the differences in early life indoor microbial agent levels and their determinants were hardly studied, even though microbial exposures are suggested to play a role in the development of these diseases. A total of 1572 dust samples from living rooms of participants were analyzed for general markers of bacterial and fungal exposure. Also here, differences in microbe concentrations were found across cohorts. Season of sampling, dog ownership, dampness, and the number of people living at home were significantly associated with concentrations for at least one microbial agent, with heterogeneity of effect estimates of the determinants across cohorts. The study concluded that both early life microbial exposure levels and exposure determinants differ across cohorts derived from diverse European countries. These results raised the hypothesis that the variations observed between cohorts in this study may play a role in the differences observed in asthma and allergies prevalence across countries.

The possible link between exposure to microbes and microbial components and the maturation and functions of immune system was studied in the LUKAS2 cohort by examining the association of house dust microbial content with cytokine -producing capacity at birth and at age 1 year (Lappalainen et al., 2012). Production of a set of cytokines (TNF-α, IFN-γ, IL-5, IL-8 and IL-10) at birth (number of subjects=228) and at age 1 year (n=200) following whole blood stimulation were measured. Concentrations of ergosterol (marker for fungal biomass), muramic acid (marker for Gram-positive bacteria) and 3-hydroxy fatty acids (C10:0-C14:0, marker for Gram-negative bacteria) in floor dust were analyzed. In addition, five single microbial species or groups were determined using quantitative polymerase chain reaction method from the house dust samples. High levels of Gram-positive bacteria in general and specifically Mycobacterium spp. in house dust, as well as total fungal levels were associated with decreased IFN-γ production, especially at age 1 year, though not significant for fungal exposures. The conclusion from this work is that high indoor microbial exposures may affect immune development in early life by reducing Th1 –cytokine secretion capacity. The observed hypo-responsiveness may reflect the adaptation of the immune system against environmental antigens. The study also indicates that more attention should be paid especially to the immuno-modulatory role of Gram-positive bacterial exposures.

A second study from the LUKAS cohort looked at the quantity and diversity of environmental microbial exposure and development of asthma. The objective of this study was to characterize microbial exposure that predicts the risk of asthma, and other respiratory conditions. 410 children were followed-up with questionnaires until the age of 6 years, with a large set of surrogate markers as well as specific microbial components measured from early life house dust samples. The results from this on-going study suggest that an index score for quantity of environmental microbial exposure would be better suitable to predict

asthma than measurement of single microbial markers (Karvonen et al., manuscript under preparation).

A study in three prospective birth cohorts (LISA, PIAMA, INMA) aimed to evaluate the associations between early life microbial exposures and airway inflammation at school age (Casas et al., manuscript submitted). Microbial exposures were assessed via general microbial markers (endotoxin, extracellular fungal polysaccharides, and β(1,3)-D-glucan); airway inflammation was measured via exhaled NO. Findings from this study indicate that early life exposure to bacterial endotoxin and owning a dog early in life are associated with lower FeNO at school age; underlying mechanisms and clinical relevance of these findings need to be further investigated.

Another, on-going study using the same (mainly urban) study population and exposure measures, did not find significant associations between microbial exposure in early childhood and respiratory and allergic health up to 10 years of age (Tischer et al., manuscript under preparation). This finding is in disagreement with previous findings in cross-sectional studies from predominantly rural areas that suggest a protective effect of priming of the immune system by early life exposure to increased levels of bio-contaminants in childhood.

Two add-on studies to HITEA were conducted in the Spanish INMA-Menorca children cohort. Study 1 explored the relation of passive exposure to domestic cleaning products, airways inflammation and lung function in school children (Casas et al., 2013(2)). Exhaled NO as a marker for airway inflammation was higher when cleaning agents and/or air refreshing sprays were used at least once a week; these finding was consistent for all types of spray under study, but was dominated by furniture spray use. There was also a tendency for lower lung function when different types of cleaning agents were used. The study suggested that the use of household cleaning agents may have an adverse effect on airways inflammation and lung function in school-age children.

Study 2 explored the impact of early life exposures to home dampness, pet ownership and farm animal contact on neurodevelopment in 4 year old children (Casas et al., 2013(3)). This study assessed the associations between early life exposure to home dampness, pets and farm animal contact and cognitive function and social competences in 4-year old children, and the associations between these indoor factors and microbial compounds measured from house dust at early childhood. Home dampness during early life significantly decreased cognitive function and social competence in the children at age 4. Cat or dog ownership and microbial levels were not associated with the psychometric tests scores. This research concluded that damp housing in early life may have adverse effects on the neurodevelopment during the first 4 years of life.

4. References - HITEA papers status published, accepted for publication, or submitted

Bakolis et al. 2012, Respiratory health and endotoxin: associations and modification by CD14/-260 genotype. European Respiratory Journal 39(3):573-81.

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Casas et al. 2013 (1), Endotoxin, extracellular polysaccharides and β(1-3)-glucans concentrations in dust and their determinants in 4 European birth cohorts: results from the HITEA project. Indoor Air 23(3):208-18. doi: 10.1111/ina.12017.

Casas et al. 2013 (2), Use of household cleaning products, exhaled nitric oxide and lung function in children. Accepted for publication in ERJ.

Casas et al. 2013 (3), Early Life Exposures to Home Dampness, Pet Ownership and Farm Animal Contact and Neuropsychological Development in 4 Year Old Children: A Prospective Birth Cohort Study. International Journal of Hygiene and Environmental Health. doi:10.1016/j.ijheh.2012.12.013.

Casas et al. 2013, Early Life Microbial Exposure and Fractional Exhaled Nitric Oxide in School-age Children". Submitted.

Chen et al. 2012, Geographical variation and the determinants of domestic endotoxin levels in mattress dust in Europe. Indoor Air 22(1):24-32. doi: 10.1111/j.1600-0668.2011.00740.x.

Haverinen-Shaughnessy et al. 2012, Occurrence of moisture problems in schools in three countries from different climatic regions of Europe based on questionnaires and building inspections - the HITEA study. Indoor Air 22(6):457-466. doi: 10.1111/j.1600-0668.2012.00780.x

Jacobs et al. 2012, Endotoxin levels in homes and classrooms of Dutch school children and respiratory health. Eur Respir J. [Epub ahead of print]

Jacobs et al. 2013, Endotoxin exposure assessment in European schools: The HITEA school study. Submitted

Jacobs et al. 2013, Dampness, bacterial and fungal components in dust in primary schools and respiratory health in school children across Europe. Submitted

Lappalainen et al. 2012, High indoor microbial levels are associated with reduced Th1 cytokine secretion capacity in infancy. International Archives of Allergy and Immunology 159(2):194-203.

Peitzsch et al. 2012, Microbial secondary metabolites in school buildings inspected for moisture damage in Finland, The Netherlands and Spain. Journal of Environmental Monitoring 14(8):2044-53. doi: 10.1039/c2em30195d

Potential Impact:

Indoor quality issues are complex in their diversity, interdependencies and difficulties in their control and regulation. The HITEA study uses a multidisciplinary approach and extensive databases to achieve its scientific goals. Its expected impacts are also diverse and will reach various target groups on both international and national scale.

1. Potential impact of the HITEA study

Impact on technical and economical aspects of indoor air quality

Many global phenomena such as climate change, increase of traffic and energy consumption and outdoor pollution, have been recognized worldwide and their inevitable effects on public health have become well-known. Within the past years, increase of prevalence and incidence of asthma and other diseases possibly attributed by environmental factors came in the focus of public debate. All of these issues have a direct link with indoor air quality although the causal connections are not yet well established.

Many European Community policies have an impact on health or health systems across Europe. Decision makers are often not well aware of potential health effects. Important health determinants cannot be influenced by health policy on its own; there is a need for coordinated actions involving other policy areas such as environmental, social and economical policies. A widely accepted principle today is the 'Health In All Policies' (Council of the EU, 2006), which emphasizes the importance of taking into account the potential health impacts in all areas of decision-making. This applies to both Community-level policies and guideline development and national, regional and local decision-making.

In order to improve this inter-sectoral collaboration of policy makers, accurate data and knowledge showing the health related aspects is of crucial importance. There is a need for new data especially on indoor air quality and health and their relation to a) technical issues such as durability of buildings, structural components and maintenance practices, and to b) economical issues, including energy use. Once the causal links are sufficiently well known, correct and effective political actions can be taken to protect the public health and the environment.

Providing knowledge for the basis of policies: IEQ in schools and health

Energy consumption is currently a strong driving force in building design, construction and maintenance. Life-cycle assessment can be used as an advanced ideology to justify selection of building components and interventions, as opposed to selection based simply on investment costs. Yet, it faces intrinsic limitation in appropriately taking into account occupant health, well-being and productivity, which is mainly due to lack of information from the environmental health point of view.

It is already known that the prevalence of dampness, poor indoor air quality and inadequate ventilation in European buildings, including schools, is relatively high. On the other hand, there is growing evidence on the positive impact of good indoor air quality and sufficient ventilation on health and human performance. There is a clear opportunity for compatible gains by improving indoor air quality by means of design and management of school facilities and other buildings.

HITEA has used an integrated approach for its complex research question: it combines epidemiology, exposure assessment and mechanistic studies in order to get a comprehensive picture of the importance of indoor air pollution on health. This integrated approach will provide knowledge for policy development and decision making.

HITEA produces objective information on the association between indoor environmental quality (IEQ) and occupant health. Focus is on the specific exposures in schools and the home environment that may be crucial with respect to occupant health, well-being and productivity. This is an important area for many reasons:

1) the existing literature indicates that IEQ may have effects on health and learning performance of school children;

2) earlier studies have failed, to a great extent, directly assess the possible association between IEQ and occupant health – including asthma and other respiratory and allergic health outcomes - using objective measures across different climates and geographic areas;

3) indoor exposures early in life - including indoor microbial exposures – are suggested to be crucial to human health later in life;

4) health and learning performance of school children may have long-lasting effects throughout their lives; and

5) IEQ in schools can be better managed if there is new evidence based motivation for the improvement of health and performance.

The improvement of indoor air quality is one of the key actions of the European Environmental Health Action Plan (EHAP). HITEA will focuses and provides scientific based information on the following aspects of needed measures of the EHAP:

-Focusing on childhood respiratory diseases (one of the four priority disease groups of EHAP),

-Produce integrated environment and health information, the importance of which is emphasized in the EHAP document.

-To provide information about the scale of the problem, how many people are affected? Development of relevant environmental health indicators

-To analyze data taking into account the multi-causality of disease and the wide range of environmental factors (physical, chemical, biological, life-style and socio-economic) that may be involved in the disease process

-Make positive contributions to education and training and to raising general levels of awareness of the nature of the environmental research undertaken and the benefits likely to accrue

-Developing networks and guidelines for factors affecting indoor air quality (e.g., dampness/mold) by using research results and national best practices.

The results of HITEA can be used to improve guidance documents and to take regulatory actions to ensure adequate IEQ in schools. A better understanding of the interrelationship between occupant health and environmental, building, and indoor air quality parameters would also help in developing management and control methods in reducing environmental and economical risks associated with school environments.

The information produced in HITEA will also be crucial for the coming programs that aim to rank environmental health issues for the future public health policies.

Providing databases on indoor exposures and related health impacts

Comprehensive databases on concentrations of biological and other indoor pollutants in schools and homes are among the most important aims of the present study. The quality of the databases is based on the harmonized sample collection techniques, sample logistics and storage, and on centralized laboratory analyses to minimize inter-laboratory variation. This is a response to the present lack of objective and harmonized data on indoor air quality in Europe. These databases will be used for assessing indoor air quality related parameters and their links to health across Europe.

The databases will be in extensive use in gathering the overall picture of indoor air quality across Europe and its potential health impacts:

1. The database will enable the assessment of indoor air quality in European schools, with focus on dampness related exposures. In addition, extensive data on the health status of the students and teachers have been collected. The design is focused on such short term health effects which potentially lead to long-term health impacts and on their immuno-toxicological mechanisms. The data set also enables the analysis of the determinants of the poor IEQ. This knowledge will have many types of next users from risk assessors to environmental policy makers.

2. The study also provides a comprehensive database on concentrations of indoor biological agents in homes across Europe. The database is unique in its extent and level of harmonization, since all the indoor environmental samples have been collected using similar, standardized protocols and the analyses have been performed according to validated procedures in a centralized way. The HITEA study will provide new microbial, immunological and toxicological parameters to complete the exposure information on biological agents. These parameters and approaches will be further developed into practical guidelines for researchers, indoor air practitioners and environmental health authorities.

3. Unique for the HITEA is also the opportunity to combine the exposure information with extensive health data collected in the participating population cohorts. The cohorts already include comprehensive data on socioeconomic aspects and other confounding factors that will be taken into account in the statistical analyses. Thus, the combination of the previously established population cohorts with novel profiling of exposures enables to achieve the maximum benefit from the study and its goal to produce data on indoor air pollution and related health impacts. This will be basic knowledge that is needed at both Community level and national and regional level for indoor air risk assessment, and indoor air policy development and in preparation of indoor air quality guidelines.

Towards more profound understanding of the mechanisms

Previous epidemiological evidence indicates that the health effects of the non-infectious microbes may develop via other than IgE-mediated allergic mechanisms. Thus, one of the main aims of the project is to provide mechanistic information on pathways via cytotoxicity, inflammation and oxidative stress. This data will help to understand the health impacts associated with particle exposures in general, and more specifically with microbial particle exposures. These exposures are closely linked to indoor air quality, which for its part is

linked to buildings, their design, construction and maintenance. This will by its part fulfill the urgent need of understanding the health effects of indoor air pollutants. This knowledge is needed for various purposes of scientific research, risk assessment, development of indoor air quality regulatory policies, and for control and risk management purposes.

Using the achieved data in regulation and for needs of stakeholders

All the above-mentioned data will be used in building up the risk assessment framework of biological pollutants of indoor environments. Biological agents have a multi-facial nature, why the traditional risk assessment methods do not lend themselves easily to these agents but the needed approach must be seen more broadly. One important impact of the HITEA results will be the raising of the awareness of various authorities, stakeholders and the general public of the health related risks of biological indoor air pollution. This is a special challenge for the dissemination of the results.

There is potential that the results of this study add pressure towards tighter regulations on indoor air quality concerning control of dampness and mould. The Consortium sees it possible that documentation of the health effects clarifies the situation that building codes should also take into account the “healthiness” of the building, not just the technical performance of the structures. 'Dampness and mould' is at the moment an indoor air issue linked strongly with health effects, but in fact without any exact metrics especially for measuring 'dampness' or 'mouldiness', the study will potentially lead to research development in this area as well.

Impact on international and national level

In large-scale visionary planning of public health, both European-wide scale and national (local) scale are needed. Production of scientifically sound information about the etiology of diseases and contribution of environmental factors to their risks is only possible by combining the best available expertise and resources internationally and using large enough study materials; therefore it is typically an action that must be carried out on European level. The main results of the study have been and will be extensively published and reported at major scientific forums, conferences and workshops (see chapter 2).

The planning of major policy trends is naturally made on European level. These policies rely strongly on relevant data produced by high quality research. Instead, planning and implementation of the practical actions, on the other hand, must be done essentially on

national and local levels. All good planning needs research data as a basis of the decision-making, therefore, there is an important and strong role within the scientific community and its organizations to disseminate scientific information to national levels through its members and dissemination tools. The partners of the HITEA study are available to expert groups and societies – such as the European Respiratory Society (ERS) or the International Society for Indoor Air Quality (ISIAQ) - and other active participation of policy development and implementation.

The expected impacts on the practices of building investigations and indoor exposure assessment

The project will improve current knowledge on the condition of school buildings and homes in different climates, and how it is linked with various environmental factors, occupant health and productivity. Anticipated results will include new methodologies in collecting data, and assessing and modeling exposures and their effects.

One important outcome is the methodology in collecting data in schools that will be utilized and validated in three different climates and geographical areas. Several reviews conducted with respect to dampness and mold in buildings conclude that the main difficulty in drawing conclusions based on hundreds of studies is that they have all used different methods and definitions, together with the fact that there are numerous variables related to the buildings and their occupants (partly driven by cultural and geographical factors). The study protocol proposed here will provide a useful assessment methodology across Europe, both from scientific and practical point of view. Potential users of the knowledge generated by this project therefore include researchers, environmental and health authorities, school management personnel, and environmental consultants. Knowledge created under HITEA - in particular considering aspects of school building investigations - is for example already contributing to a European wide survey on indoor air quality in schools as a part of ENHIS (Environment and Health Information System), guided by the WHO Regional Office in Europe.

2. Main dissemination activities and exploitation of results

Dissemination by Publications

All participants of the project will contribute throughout Europe and globally to disseminating relevant research and scientific outcomes from the project via various peer-

reviewed scientific journals. We anticipate producing position papers on the topic areas within indoor air quality, chemical and biological pollution of indoor air and their potential health impacts, and the sampling and analyzing aspects of these contaminants. Professional journals and publications are also important forums in order to reach large audiences of various stakeholder groups.

At the end of the funding period of HITEA the project has achieved 10 scientific papers that have been published or accepted for publication, with 3 more papers currently being submitted. All of these papers appear in high-quality journals with major impact and visibility in the respective research areas, such as the European Respiratory Journal or Indoor Air Journal. A list of HITEA papers with status published, accepted for publication or submitted has been provided in the results section of this summary report.

There are concrete plans and outlines for producing a minimum of another 16 scientific papers based on HITEA data, but this number is likely to increase still. The drafting of ten of the anticipated papers is currently on-going – major publication efforts are anticiapted for 2013 and 2014.

The HITEA project has furthermore been extensively introduced to the scientific community through presentations at various national and international conferences, symposia or similar. In total, more than 30 contributions to some 20 different events have been taking place during the last three years. Major conferences in the field of indoor air quality (e.g. the "International Conference on Indoor Air Quality and Climate", "Healthy Buildings Conference"), respiratory health (e.g. the "European Respiratory Society Conference"), allergy and immunology (e.g. the "European Academy of Allergy and Clinical Immunology"), and epidemiology and occupational health (e.g. "International Society for Environmental Epidemiology Conference", "International Conference on Epidemiology in Occupational Health"), to name only a few major fields HITEA has been active in. The target audience was mainly scientific, but also practitioners, stakeholders, authorities and societies, as well as media representatives were addressed in many of these events. Being currently in the most active time of publication of HITEA study results it can be expected that this type of dissemination activities will be on-going still several years to come.

Dissemination by workshops

Dissemination of knowledge has also been taking place via a number of workshops on different topics. Specific attention has been given to the innovative spin off of the project in the fields of sampling approaches, strategies for indoor exposure assessment and on

exposure characterization of microbial agents. For these workshops, representatives of several organizations have been invited from across the EU, including members of organizations like the European Respiratory Society (ERS), International Society for Indoor Air Quality and Climate (ISIAQ), the EU SCOEL committee for standard setting in indoor (work) environments, building designers, constructors and architects and Environmental and Occupational Hygiene Organizations.

A workshop addressing specifically the European Community Respiratory Health Survey (ECRHS) study group was held in Germany in June 2012. This workshop was organized by the Helmholtz Zentrum Muenchen, one of the beneficiaries in the HITEA consortium. The topic of the workshop was on microbial and chemical exposure and respiratory health effects in the ECRHS, reviewing the work conducted under HITEA for this large European survey, which includes many of the major actors in the field of respiratory health and allergic diseases in Europe. In addition to summarizing work in progress activities, identification of research gaps and suggestion future work and publications were main efforts during this workshop.

A workshop/symposium on "Moulds in indoor spaces" (April 2012, Austria) gathered scientists, stakeholders, and politicians to raise awareness of the issue of indoor biological contaminants in Austria. The event was organized by the Austrian beneficiary of the HITEA consortium, the University of Natural Resources and Life Sciences. Part of the HITEA consortium contributed as speakers to this event disseminating the project as well as the underlying topic.

A workshop on "Indoor exposures and health - New insights in indoor microbial exposure measurements and health effects from exposure to chemical and biological agents in schools and homes" was held in February 2013 in Barcelona, Spain. This event was organized by University of Utrecht together with the Centre for Research in Environmental Epidemiology (CREAL), both partners in HITEA. These 1.5 days focused on results derived from HITEA and other recent or on-going studies within this research area. It was anticipated by the organizers to make this type of workshop in "Indoor microbial exposure assessment" a biannual event, possibly linking to conferences in the area. In initial contacts with the ISIAQ – the International Society of Indoor Air Quality – this idea received very positive feedback and support.

Public project website

An external project webpage to the HITEA project has been created and can be accessed under: http://www.hitea.eu

This webpage has been created for the scientific community and various stakeholders, as well as for the general public and is acts as one major dissemination tool. The site includes information on the rationale, aims and implementation of the project, and is regularly up-dated for the progress of the study and publication of results. The relevant contact information includes links to the beneficiaries' own web-pages. Information about the school study – directed at parents, pupils and teachers – is provided also in the domestic languages of the three participating countries (Finland, Spain, The Netherlands). Information will be kept up-to-date on the website also after the end of the funding period of HITEA.

Other Dissemination aspects

The project has moreover been actively linking to other initiatives/programmes within this research field. HITEA has been presented at a Indoor Air Quality Expert Group meeting in Luxembourg at DG SANCO (October 2012). There are plans that project results will be contributed to drafting new air quality policy guidelines. On national level, the project has been disseminated to national authorities and stakeholders in the context of symposia or in print media. HITEA knowledge is furthermore contributing to a European wide survey on Indoor Air Quality in Schools as a part of ENHIS (Environment and Health Information System), linking the project to activities of the WHO Regional Office in Europe, in particular considering aspects of school building investigations in the planned survey.

Within HITEA, dissemination through technology development was furthermore anticipated. The background of this initiative was to contribute to the development of methods base on rapid direct reading devices which can be used by experts in the field without the need of backup by highly specialized laboratories. This would facilitate future surveys and studies by local health authorities and specialists for whom objective measurements are not available because of the high costs. These technology development activities have been implemented as planned. The subcontractor developing this technology utilized project results and knowledge created in order to facilitate the development of a lateral flow immunoassay targeting at selected microbial groups relevant for biological monitoring of Indoor Environments.

List of Websites:

http://www.hitea.eu