RESEARCH PROGRAMME

Department of Respiratory Diseases and Allergology

Aarhus University Hospital

Aarhus University

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Content

Introduction

The research department is a part of the department of respiratory medicine and allergology at Aarhus University Hospital. Its major goal is to do research in clinical respiratory medicine and allergology especially in the area of diagnostics and treatment. Basic research of disease mechanisms in respiratory medicine and allergology also have a place and is often done in collaboration with other institutions e.g. institutes at Aarhus University and other research institutions nationally or internationally.

The chief of the department is the professor (chair) in clinical respiratory medicine (at present vacant) in conjunction with the professor in basic allergology and pulmonary medicine. The staff comprises the two professors, 3 full time research nurses, 2 lab technicians, 1-2 post docs, the 6 associate professors from the clinical department, 1 secretary, phd students and medical students.

At present 6 phd and 4 medical students are working in the research department.

Department of respiratory medicine and allergology

The department comprises the ward named LUB6, 14 beds, LUB5 6 beds and a outpatient unit. The outpatient clinic divided into 5 units: cancer diagnostics, pulmonary medicine, allergology, respiratory-physiological laboratory and a endoscopic unit .

In the unit for cancer diagnostics

Lung cancer: Outpatients clinic

LUCA (LUng CAncer Out Patient Clinic) is a highly specialised department for state-of-the-art investigation of patients under suspicion of thoracic malignancy. 2000 patients are referred every year.

In a dynamic out-patient setting our dedicated staff is working to establish diagnosis and refer patients to oncological or surgical treatment within 28 days from referral, according to national standards.

Diagnostic procedures span from non-invasive imaging such as CT and PET-CT to highly advanced invasive endoscopic ultrasound investi-gations, EBUS and EUS, to which patients are referred from the whole region. Transthoracic, abdominal and bone needle aspirations are performed by highly specialised invasive radiologists.

Treatment decision for every individual patient is made at the MultiDisciplinary Team Lung conference (MDT) held twice weekly with participation of specialists in Pulmonary Medicine, Oncology, Thoracic Surgery, Thoracic Radiology, Nuclear Medicine and Pathology. Regional hospitals participate via video-link for discussion of patients.

RFA (Radio Frequency Ablation) treatment of patients with malignant lung nodule(s) not found suitable for surgery is performed in the department of Radiology with postoperative observation and treatment in our department.

Research in LUCA focuses on improving standards for investigation and treatment of patients with lung cancer and is done in close collaboration with collegues from the Department of Oncology and the

Department of Radiology at Aarhus University Hospital, and from the Research Unit for General Practice under the Department of Public Health at Aarhus University.

Among current studies are:

A study with the aim of optimizing the oncological treatment of patients with non-small cell lung cancer (NSCLC) with mutations in the epidermal growth factor receptor (EGFR). Inhibition of EGFR is associated with prolonged survival in some patients with NSCLC and patients with mutations in EGFR are among the responders. But it is unsettled which importance expressions of the other members of the EGF system have and it is the aim of this study to explore this.

All patients who are undergoing investigation for lung cancer are asked to participate. Those who participate give blood samples and samples from needle aspirations from the tumor and lymph nodes if such samples are taken as part of the work up.

A randomised controlled study aimed to evaluate the diagnostic strategy for patients referred to the lung cancer fast track pathway. Lung cancer is the most common cause of cancer death in Denmark, and triaging patients through fast track pathways is recommended to improve patients experience and outcome. While such pathways are likely to lead to earlier diagnosis and treatment, data on the most efficient organization of the pathways are limited.

The randomised intervention is to have a chest CT scan done before chest physician evaluation. The study outcome is effectiveness, as measured by spent physician time per patient, number of CT-scans, and staff acceptability.

A study evaluating the use of Microwave Ablation as an alternative local treatment of lung tumours - primary or metastatic - in patients ineligible for surgery. Microwave Ablation is a new, careful, fast and cheap treatment, which seems to be an effective alternative in local treatment of lung tumours. The ablation is performed as a single treatment in anesthetized patients, where an antenna build in a needle is CT-guided placed in the lung tumour. The antenna will for 10 minutes emit electromagnetic impulses, which heat and destroy the tumour. Half the patients will be discharged from the hospital the following day, the majority of the rest within 2-3 days.

Respiratory medicine: Out-patient clinic

In the respiratory out-patient clinic, there are approximately 16000 visits per year.

The clinic receives patients from the city of Aarhus (350000 citizens) with common pulmonary diseases and from the region with highly specialized function in pulmonary medicine (1.2 mio indbyggere).

The most common conditions and diseases examined and treated are:

Dyspnea

Cough

Tuberculosis, aspergillosis and other infectious pulmonary diseases.

COPD patients are by far the biggest group. In trying to prevent them from being admitted to hospital, an acute clinic has been developed, offering the possibility of day-to-day consultations.
The sickest COPD patients are invited to participate in the Pulmonary Rehabilitation program.

Research in Tuberculosis and various aspects of COPD is taking place in collaboration with the Department of: Psychology, Healthcare Technology, Public Health, Advance Care Planning (involving numerous departments), the Pharmaceutical Industry as well as projects of our own.

Allergology: Outpatients clinic

3000 new patients are referred to the out-patient clinic of allergology every year. The clinic is a part of **ALLERGY CENTRE WEST** one of three centres in Denmark (the others in Odense and Copenhagen) serving as a highly specialized unit for allergology for 1.9 mio citizens.

The departments taking part in the centre comprise: department of respiratory diseases and allergology, department of occupational medicine, department of dermatology, department of ENT and department of Pediatrics.

The most common conditions and diseases treated are asthma and airway allergies.

The unit is also center for difficult treatable asthma and biological treatment is offered.

Urticaria and food-allergies are also examined and treated.

The unit has main research in mast cells, basophil activation test, and immune-therapy.

Respiratory medicine unit B5.

B5 is highly specialized in diagnosing and treating patients with lung fibrosis and other rare respiratory diseases. It is also responsible for lung transplantation evaluation and follow-up after transplantation.

The staff is composed of a senior consultant and specialist in Respiratory Diseases, a head of nurses and 7 nurses with experiences in lung diseases and specifically pulmonary fibrosis and transplantation. There are also 2 nurses help's, 2 service assistants and 2 secretaries. There is 3 doctors each day, and -2 of these are specialists in respiratory diseases, while the others are trainees at different levels for becoming specialists in pulmonary medicine.

The Department of Respiratory Diseases, B5, has as highly specialised department the responsibility for diagnosis and treatment of patients with interstitial lung diseases and other rare lung diseases, complicated sarcoidosis and lung transplantation evaluation and follow-up after transplantation.

The department receives approximately 300 new referrals each year for diagnosis of interstitial lung diseases and approximately 30 referrals for lung transplantation evaluation or other surgical treatment for COPD such as lung volume reduction surgery or bronchial valves. Also a smaller number of patients with rare lunge diseases such as lymphangioleiomyomatosis, alveolar proteinosis etc. are referred.

The department has a close cooperation with the transplant unit at Rigshospitalet in Copenhagen, including video conferences each week. Each week there is a radiology conference, mostly on high resolution computered tomography scans of the lungs ((HRCT) and each month multidisciplinary interstitial lung conferences with participation of radiologists, pathologists and pulmonologists. Every 6th week there is a pulmonary vascular conference with participation of cardiologists, radiologists, pathologists, rheumatologists and pulmonologists on patients with pulmonary hypertension. Two times each year, there is a conference with the ophthalmologists on patients with uveitis, mostly sarcoidosis patients.

The research interest of the department is mainly in interstitial lung diseases such as idiopathic pulmonary fibrosis, rare lung diseases and lung diseases complicated with pulmonary hypertension. The department has developed the "Registry of Interstitial Lung Diseases in Western Denmark", a registry that includes all newly referred patients with interstitial lung diseases between2003 and 2009. Based on the experiences from the retrospective registry and in close cooperation with the Danish Technical University in Copenhagen, a prospective registry has been developed for the registration of patients with idiopathic pulmonary fibrosis (IPF) but also other interstitial lung diseases and rare lung diseases. The department has also taken a Nordic initiative to develop a Nordic registry for these patients based on our experiences.

Based on the all the registries, research on epidemiology including incidence, prevalence and characteristic of different subtypes of interstitial lung diseases are performed. Also, research on the prognostic value of HRCT scoring, the connection of smoking and lung fibrosis, socioeconomic studies etc are performed.

In a close cooperation with the Department of Cardiology and Pharmacology, the department contributes to research on the incidence/prevalence of pulmonary hypertension among lung patients (COPD, lung fibrosis, sarcoidosis), the value of different biomarkers and echocardiographic screening.

The department participates actively in several ph.d. studies on pulmonary fibrosis, pulmonary alveolar microlithiasis, tracheomalacia and pulmonary hypertension.

Research in patients with pulmonary fibrosis and sarcoidosis initiated by pharmaceutical companies are also carried out.

Respiratory medicine unit B6

The unit is mainly receiving acutely ill pulmonary patients from the county of Aarhus. It also serves as a highly specialized unit for acute referral for 1.2 mio. citizens in the region and as specialized unit for very lung diseases for 2.1 mio. citizens. The staff of the department consists of one consultant, one charge nurse, 20 nurses, three social/health assistants, two service- assistants and one secretary. There are three doctors in the department every day. At least one of them is specialized in pulmonary medicine, while the remaining doctors are on different stages in their supplementary training.

The patients being received typically have COPD-exacerbation, active tuberculosis, exacerbation in pulmonary fibrosis, severe pneumonia and lung infections, acutely lung-transplant rejection, severe respiratory insufficiency and rare lung diseases.

The yearly admission is apprx 800 patients. 10-15 patients will be treated with a respirator within the department, 30-50 with non-invasive ventilation.

The department works close together with respiratory medicine unit B5 when it comes to transplantation of the pulmonary and pulmonary fibrosis.

The department is an important unit in the medical education at AUH, as well as in common internal medicine and in pulmonary medicine.

In addition the education of the nurses is an important part of the activities going on in the department.

The research that is being done in the department includes research about rear pulmonary diseases, palliation and inhalation-treatment of different infectious diseases**.**

Research in tuberculosis is also one of the main areas in focus in the unit.

Respiratory Physiologic Laboratory and X-ray unit

The unit has a wide range of equipment for examination of the airways and lung function. Together with the outpatient clinics more than 25.000 spirometries are performed every year. The unit has 4 body-plethysmographs and is doing more than 8000 lung function tests including diffusion capacity and static volume measurements every year. Forced ocillometry testing. Equipment for fully automatized ergospirometry. Endoscopic equipment for diagnosing exercised induced larynx disorders. Challenge facilities. More than 6000 conventional chest x-rays are performed every year.

Research profile

Two main research areas:

Interstitial Lung Diseases

Allergology: Basophils and diagnostics

Respiratory Medicine

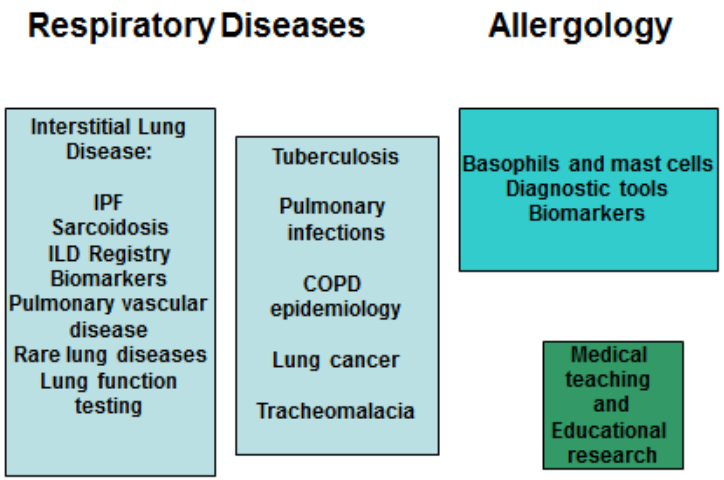
- Intersititial lung diseases
  - o IPF
  - o Sarcoidosis
  - o Database
  - o Biomarkers
  - o Vascular diseases
  - o Rare Lung diseases
  - o Lung function testing

    - Tuberculosis
    - Infectious diseases in the lung
    - COPD-epidemiology
    - Lung cancer
    - Tracheomalacia

Allergology

- Basophils and mast cells
- Diagnostic tools
- Biomarkers

Medical teaching and educational research



Troels Johansen

M.Sc.Eng., ph.d. student

Pulmonary gas exchange and ventilation/perfusion inequalities in Dysfunctional Breathing and asthma

Background

Symptoms of chronic and/or acute dyspnea can be caused be a range of conditions and are sometimes misdiagnosed as asthma. In some case, Dysfunctional Breathing (DB) is the underlying cause of the dyspnea, The Hyperventilation Syndrome being one of the most common manifestations of DB. Characterized as dyspnea without an apparent organic cause, DB is generally hard to diagnose and treat, though the impact on quality of life and functional level can be severe.

DB is common among all age groups and can overlap with organic and psychiatric disease. Furthermore, there are strong indications that DB may be a common cause of idiopathic cardiac pain.

Very few quantitative studies of DB have been performed. Consequently, DB's impact on pulmonary ventilation, perfusion, gas exchange and blood gas levels are not well known.

Purpose of the ph.d. project

To contribute to a quantification of the effect of DB on pulmonary ventilation, perfusion, gas exchange, blood gas levels and acid/base status, in particular regarding the following research questions:

- What methods and sensor technologies are best suited to measure DB and it's physiological effects?
- How can we most efficiently distinguish DB and asthma?
- How can the severity of DB be reliably quantified? (this is especially important in order to obtain an objective method of measuring the effect of interventions to alleviate DF)

Methods

1) Systematic review and meta-analysis of blood gases in asthma, in order to distinguish DB and asthma on the basis of pulmonary gas exchange
2) Computer modeling and simulation of pulmonary gas exchange in DB (collaboration with Center for Model-based Medical Decision Support (Aalborg University))
3) Measurement of pulmonary ventilation, perfusion and arterial blood gases in DB (collaboration with The Pulmonary Functional Imaging Research Laboratory (Harvard Medical School)).

Perspectives

Increased understanding of pulmonary gas exchange in asthma and other obstructive lung diseases may contribute to the development of more effective treatments.

Junjing Su, MD, ph.d. student, dept. Respiratory Medicine AUH, dept Cardiology AUH , Dept Pharmacology AU, NHLI, Imperial College London,

Background

Pulmonary hypertension (PH) is a severe disease defined as a mean pulmonary arterial pressure (MPAP) above 25 mmHg. Several types of PH exist, such as pulmonary arterial hypertension (PAH) and chronic tromboembolic pulmonary hypertension (CTEPH).

The pulmonary circulation is highly compliant. Therefore, PH leading to right ventricular (RV) dysfunction is a relatively late manifestation of disease. Pressure separation techniques to subtract the reservoir pressure and wave intensity analysis (WIA) to quantify wave reflection have been applied in the aorta and the coronary arteries . However, WIA has not been conducted in the pulmonary circulation except in animal models. Moreover, WIA has not been introduced to hospitals in Denmark. Pilot experiments performed in two patients at the National Heart and Lung Institute (NHLI), Imperial College London, have shown that it is feasible and safe to perform measurements of pressure and flow in the pulmonary arteries.

Hypothesis

Wave reflection in the normal human pulmonary arterial bed assists forward flow by "sucking" blood down the vessels, i.e. that the dominant reflected waves will be expansion waves resulting from reflection from an "open-ended" vessel. In subjects with PAH and CTEPH, there will be a backward travelling reflection wave in mid-systole which decelerates blood flow whilst increasing pulmonary pressure.

Methods

Study 1

This part will be a two-armed observational cross-sectional study carried out at NHLI, Imperial College London, among adult patients due to undergo right and left heart catheterisation for clinical reasons. 18 subjects with established PAH and 18 subjects being clinically investigated for shortness of breath are required. During cardiac catheterisation, simultaneous pressure and flow velocity within the pulmonary artery will be measured using a purpose-designed pressure and Doppler flow sensor tipped guide wire.

Study 2

A three-armed observational cross-sectional study will be carried out at the Department of Cardiology, Aarhus University Hospital. 18 patients in each group are required. Patients with established or suspected PAH and preoperative CTEPH will be recruited for right heart catheterisation, during which, simultaneous pressure and flow velocity within the pulmonary artery will be measured based on the techniques described above and data will be used for WIA. As controls, cardiac transplant recipients will be invited.

Study 3

This part will be a follow-up study among the patients with CTEPH from study 2 that are scheduled to go through PTE. So far, right heart catheterisation is not routinely performed postoperatively. These patients will be asked to undergo right heart catheterisation and WIA two months after PTE and the measured MPAP and wave intensity patterns will be compared to preoperative measurements. Furthermore, functional capacity assessed by the 6-mintue-walk-test, lung function tests, the Borg Dyspnoea scale and patients' perception of life quality will be assessed preoperatively and two months after PTE.

Perspectives

Increased understanding of the nature of the pulmonary vessel stiffness and dynamics in normal and in PAH contribute to the development of more effective treatments PAH.

Elisabeth Bendstrup, Chief Physician, Associate Professor, Ph.D.

Elisabeth Bendstrup is responsible for and participating in several research projects in the department. There are three main areas at the moment.

A National Registry for Interstitial Lung Diseases: THE NATIONAL ILD DATABASE

A prospective observational cohort study in interstitial lung disease patients in the Denmark

Background: Interstitial Lung Diseases (ILD) is a heterogeneous group of more than 200 different rare diseases affecting the lung tissue. Even if many factors such as environmental and occupational agents including drugs, aspiration, radiation therapy, genetic predisposition and collagenous diseases have been implicated, the majority of these diseases are considered idiopathic. Idiopathic pulmonary fibrosis (IPF) is the most common idiopathic lung disease representing about 50% of

idiopathic cases. IPF is a serious lung disease that affects mostly adults, particularly after 65 years of age and incidence the increases with increasing age. IPF is associated with a high mortality rate that is higher than many cancers with a median survival of 2-3 years. Data on incidence of IPF show an eightfold variation from 3.62/100,000 person-years in Spain to 31.5/100.000 person-years in males and 26.1 in females in New-Mexico. There a no valid incidence data on other sub types of ILD.

Aim: A national prospective registry, The National ILD Database, has been developed in cooperation with the Danish Technical University in Copenhagen and is based on Aarhus experiences with a retrospective database. The aim of the registry is

- To describe patient demographics, working/living status and clinical characteristics at diagnosis
- To estimate the incidence and period prevalence of ILD and subtypes
- To estimate the distribution of ILD and subtypes according to severity at diagnosis
- To describe natural history, disease course and outcomes in a longitudinal real world setting
- To describe known and suspected risk factors of ILD and its subtypes associated with outcomes and death.

Perspectives: There are no valid Danish data of the incidence of ILD and subtypes. Knowledge will allow better organization of ILD diagnosis and treatment in the future and will give us important knowledge on the course and prognosis. This will make it possible to establish better patient programmes including treatment.

Sarcoidosis

Background: Sarcoidosis is a multiorgan, non-nekrotizing granulomatous diseases with a high incidence in Scandinavian countries. Studies from 1990'es have found that the incidence in Denmark is 7.2/100,000 corresponding to 400 new cases per year. There are no new studies on the incidence or extrapulmonary manifestations of sarcoidosis. The disease is characterized by an often benign course but approximately 10-20% of patients have a severe disease course with progressive pulmonary fibrosis, respiratory insufficiency and ultimately death or, is possible, lung transplantation.

Aim: To develop a National Sarcoidosis Registry based on the experiences from the National ILD Database and the tuberculosis registry.

Methods: All incident sarcoidosis patients and their data on symptoms, clinical characteristics, radiology, blood samples and pathology will be registered. A biobank will be created allowing for blood and tissue samples to be kept for analysis on genetics, HLA-type and biomarkers.

Perspectives: The registry is aimed to become national and will give an opportunity to perform epidemiological and clinical research in these patients.

Common variable Immonodeficiency CVID

Background: Common variable immunodeficiency (CVID) is one of the most frequent primary immunodeficiencies affecting approximately one in 50,000 northern Europeans. The condition is characterized by decreased serum levels of immunoglobulin G (IgG) and reduced serum IgA and/or IgM levels, and recurrent infections especially of the respiratory and gastrointestinal tracts. The aetiology of is unknown. 25-58% of CVID patients have non-infectious pulmonary manifestations termed granulomatous-lymphocytic interstitial lung disease (GLILD). GLILD is a group of diseases with clinical and radiological similarities, but different histopathological patterns i.e. lymphocytic interstitial pneumonia, follicular bronchiolitis, non-specific interstitial pneumonia and granulomatous disease are described.

Hypothesis: The incidence of GLILD in Danish CVID patients is higher than in other cohorts because of the genetic composition in ethnic Danes.

Aim:

- To obtain valid estimates on the true prevalence of CVID and to characterize GLILD in a Danish cohort of CVID patients
- To evaluate differences in immunologic parameters in CVID-GLILD and CVID-non-GLILD patients in bronchoalveolar lavage fluid, blood and tissues.
- To evaluate differences in HLA type and possible mutations in patients CVID-GLILD and CVID-non-GLILD

Perspectives: Since the introduction of intravenous immunoglobulin treatment for CVID patients, the severity and frequency of infections have declined. Non-infectious complications of CVID are therefore becoming more important and contribute increasingly to the morbidity and mortality in CVID. Knowledge of the characteristics of CVID patients could result in higher awareness of GLILD for clinicians, possible regular screening programs and ultimately improve the course of disease and the prognosis for the patients.

Ole Hilberg, Chief Physician, Associate Professor, Dr.Med.

Ole Hilberg is taking part in different research projects in the department. Three areas are during development at the moment.

A Study of lung and plasma voriconazole concentrations after Inhalation and Oral Administration

Background

Voriconazole is a broad-spectrum antifungal drug effective against *Aspergillus fumigatus*. It inhibits the cytochrome P450 dependent 14-α–lanosterol demethylase preventing the conversion of lanosterol to ergosterol. This results in accumulation of toxic methylsterols in the fungal wall and inhibition of fungal growth. Studies of inhaled voriconazole in rodents have been shown to reduce histological

manifestations of invasive aspergillosis and it has been proposed that a favourable lung tissue to plasma concentration ratio is obtained through this route of administration. In a recent case report it has been shown that inhaled voriconazole in humans has a remarkable effect against invasive aspergillosis in humans without any side effects. Consequently, inhalation of voriconazole may achieve higher concentrations at the site of infection without increasing the risk of systemic side effects, this has not been investigated in humans.

Hypothesis

Compared to orally administered voriconazole, inhaled voriconazole will have a higher lung tissue/plasma concentration ratio.

Aim

To investigate the concentrations of voriconazole in bronchoalveolar lavage fluid (BALF) and serum after inhaled and orally administered voriconazole, respectively. 12 patients inhale voriconazole, 40 mg once dailyt for three days. Last inhalation is given 8 hours before a planned bronchoscopy for other reasons. Voriconazole is measured in the BALF and serum. 12 patients are administered oral voriconazole (400 mg twice daily for one day, followed by 200 mg twice daily for two days) . Last dose is given 8 hours before a planned bronchoscopy for other reasons, and voriconazole is measured in the BALF and serum.

Perspectives:

A preliminary study has already shown extraordinary effects of inhaled Voriconazole. Therefore, the results of the present study will contribute to a better and safer treatment of fungal infections in the airways.

Socioeconomic aspects of interstitial lung diseases

Objective: Interstitial Lung Disease (ILD) is among the leading causes of morbidity and mortality in lung disease worldwide, but longitudinal studies of the economic consequences of ILD are scarce. This study will evaluate the economic consequences of ILD in Denmark before and after initial diagnosis.
Methods: Using records from the Danish National Patient Registry (2000-2010), apprx 11000 patients with will be identified and compared with 44000 randomly selected controls matched for age, gender, educational level, residence, and marital status. Direct and indirect costs, including frequency of primary and secondary sector contacts and procedures, medication, unemployment benefits and social transfer payments will be extracted from national databases. Furthermore, the patients from a minor cohort of a retrospective patient with ILD of aprx 450 patients, classified into specific diagnosis of IPF and other ILS will be included.
Results: Patients survival rate will be compared with controls as well as rates of health-related contacts, medication use and socioeconomic costs. The employment and the income rates of employed ILD patients will be compared to controls.
The annual net costs, including social transfers will be calculated.
Perspectives: This study is going to provide unique data on direct and indirect costs before and after initial diagnosis with ILD in Denmark as well as mortality, health and economic consequences for the individual and for society.

Galectin-3, an important mediator in lung fibrosis.

Background

Lung fibrosis is a rare disease. Many mediators are thought to be involved. Galectin-3 has been shown to be an important mediator in many fibrotic diseases in other organs. Recently, animal models and (human) studies seem to indicate that anti-Galectin-3 may provide a new option in treatment of fibrotic diseases.

Hypothesis

Galectin-3 is an important mediator in lung fibrosis. Anti-galectin-3 may inhibit fibrosis in animal models of lung fibrosis, and in humans in IPF.

Methods

1. Literature review
2. Animal studies
3. Phase 1+2+3 human studies

Perspectives

Recently, Pirfenidone has been approved in the treatment of idiopathic pulmonary fibrosis (IPF). The drug has shown to have significant effects in clinical trails but has only a limited effect against the decline of lung function and survival in IPF. Animal studies of anti-galectin-3 show superior effects compared to Pirfenidone and due to low toxicity it may become an important treatment in the future of IPF

Projects

Interstitial lung diseases

Charlotte Hyldgaard, MD, PhD student, Dept of Respiratory Diseases, Aarhus University Hospital

A retrospective cohort study of West Danish patients with interstitial lung diseases; burden, severity, treatment and survival

Introduction

Interstitial lung diseases (ILDs) form a heterogeneous group of diseases causing varying degrees of inflammation and fibrosis. The diseases typically result in a restrictive ventilatory defect and impaired diffusion capacity. The majority of cases are idiopathic, but many exogenous factors, such as organic dust and certain drugs, may cause interstitial lung diseases. International consensus on diagnostic criteria was reached only ten years ago, and epidemiological data based on the current diagnostic criteria are sparse.

Aim of the study

To characterize ILDs incidence rate in Danish ILD patients diagnosed at a tertiary referral hospital and to compare classification of patients with idiopathic pulmonary fibrosis (IPF) based on the 2001 and 2011 ATS/ERS/JRS/ALAT guidelines.

Methods

The project is based on the West Danish ILD Registry which was established as a part of this study. The registry includes all incident patients diagnosed with interstitial lung diseases at the Department of Respiratory Diseases, Aarhus University Hospital, between 2003 and 2009. Details of referral, diagnostics, and clinical status have been recorded, and all diagnoses have been re-evaluated according to current diagnostic criteria for interstitial lung diseases.

Results

A total of 431 patients were included in the registry. ILD incidence was 4.1 per 100,000 central Jutland inhabitants/year. Idiopathic pulmonary fibrosis (IPF) was the most common diagnosis (28%) followed by connective tissue disease-related ILD (14%) and hypersensitivity pneumonitis (7%). Almost one fourth of the patients were diagnosed with either end-stage fibrosis (10%) or unclassifiable fibrosis (14%). Lung biopsy was performed in 40% of the patients. In half of the cases, IPF could be diagnosed by high resolution computed tomography (HRCT) scan alone. The median survival of patients having IPF was 3 years.

In this well-characterised cohort, 25% of patients had unspecific ILD diagnoses, even after a systematic re-evaluation. Although re-

evaluation raised the number of IPF diagnoses, a diagnostic "grey zone" was evident in patients with UIP features not qualifying the patients to be diagnosed with IPF.

Research in progress

Prognostic factors in IPF and other ILDs with focus on the impact of comorbidities on disease course and survival. Disease course and prognostic determinants in unclassifiable ILD

Smoking-related ILDs.

Pulmonary Fibrosis and Sexual Function

Introduction

The relationship between erectile dysfunction and chronic diseases, most notably diabetes and atherosclerosis, is well established. A number of studies have shown a relationship between COPD and erectile dysfunction classified by *International Index of Erectile Function* (IIEF). The pathogenesis is not clearly established, but 3 relatively small studies have shown a significant correlation between chronic hypoxia and erectile dysfunction among men. Other proposed courses are systemic inflammation and hormonal disturbances.

As to our knowledge, no studies have evaluated the association between sexual dysfunction and interstitial lung disease (ILD), or the association between objective lung function tests and sexual dysfunction.

Hypothesis

Interstitial lung disease is independently associated with sexual dysfunction.

Objective:To evaluate whether an established diagnosis of interstitial lung disease is associated with sexual dysfunction as classified by IIEF, and to evaluate if there is an association between lung function tests and/or hypoxia and sexual dysfunction among this patient group.

Study population: The study is performed at Department of Pulmonology (Lungemedicinsk afd. LUB), Aarhus University Hospital. Being a highly specialized department, a relatively high number of patients with the insterstitial lung disease, mainly from Western Denmark, are seen here on a regular basis. In this highly selected setting, we aim to include 50 patients with ILD in our study.

Methods: Patients will be selected for enrolment based on a diagnosis (ICD-10) of interstitial lung disease (DJ84, DJ84.1A-X, DJ84.8), documented on basis of radiologic manifestations on high-resolution computed tomography (HR-CT) and/or a histopathologic pattern supporting ILD in a lung biopsy.

Patients with a diagnosis of diabetes, ischaemic heart disease or manifest atherosclerosis will be excluded. Enrolled patients will fulfil a questionnaire (IIEF, Danish version, appendix 1), covering impact of sexual dysfunction during a 4 week period. The latest lung function test (max. 3 months) performed in Department of Pulmonology, Aarhus University Hospital, will be registered, to evaluate the association between IIEF-results and lung function. If possible, a comparable lung function test 1 year earlier will be registered for comparison of results. Upon enrolment, an arterial puncture (1 ml) will

be performed, to assess the relation between hypoxia and sexual dysfunction.

The study has been approved by the regional Research Ethics Committee.

Charlotte U.Andersen, MD, PhD, postdoc, Dept of Respiratory Diseases and dept of pharmacology, Aarhus University Hospital

Pulmonary arterial hypertension in lung disease: Biomarkers

Background: Pulmonary hypertension (PH) is considered as a complication to chronic lung disease that worsens the prognosis. The epidemiology and use of biomarkers of PH are not well described in patients with chronic lung disease.

Aim: To investigate the prevalence, impact, and role of proposed biomarkers of PH in COPD and ILD patients.

Methods: Echocardiographic screening for PH was performed in stable COPD patients previously admitted to hospital with an acute exacerbation, and in ILD patients at a tertiary referral center. Patients with signs of PH on echocardiography were asked to undergo right heart catheterization to confirm PH. Demographic data, lung function parameters, and results of six minute walk tests were collected, and N-terminal pro-brain natriuretic peptide (NT-proBNP) were measured.

Results: The main findings of the studies were that 14% of COPD patients screened positive for PH by echocardiography. Patients who screened positive had a poorer prognosis and a lower exercise capacity. Fourteen percent of ILD patients had PH. PH patients had a markedly worse prognosis and a lower exercise capacity than non-PH patients after correction for lung function parameters. In both COPD and ILD, a value of NT-proBNP below 95 ng/l effectively excluded the presence of PH

Conclusion: Echocardiographic screening for PH yields prognostic information in COPD patients, and PH in ILD patients is a common finding and severely affects prognosis per se. This suggests a rationale for specific treatment of PH in ILD patients. NT-proBNP may serve as a first line screening test to exclude PH in ILD and COPD patients.

Research in progress: NT-proBNP as a rule-out test for PH will be further validated in incident ILD patients at AUH.

Lung function testing

Lars Kristensen, MD, PhD student, Dept of Respiratory Diseases, Aarhus University Hospital

What is a normal lung function?

Introduction:

Lung function tests (LFT) are used in the diagnosis of most respiratory diseases, among them COPD and asthma. They are used in the classification of the severity of the disease, in monitoring the progression of the disease, and monitoring the effect of the treatment. Patient's lung functions values are compared with a set of *reference values*, which is the expected lung function for a person with the same sex, age and stature as the patient. In Denmark we use reference values from foreign countries and common for them is a lack of data for people older than 70 years. Therefore, we use extrapolated values for the elderly and this is a significant problem, because different studies have shown different results of how the lung function decline with increasing age. This problem will become of even greater importance in the future, as the general population gets older. For lung diffusion capacity and body pletysmography there are few data sets with reference values and common for these data is that they are based on very few test persons. (Less than 500)

Purpose:

To establish Danish reference values for three different types of lung function testing: Spirometry, lung diffusion capacity and body pletysmography.

Methods:

We will examine 3.000 randomly selected healthy, never smoking Danish citizens of Caucasian origins, aged 18-90 years with three different minds of LFT. The potential test persons are selected by a random sample from the Danish Central Public Register.

The tests will be performed at approximately 16 different centres in Denmark.

Before including the test persons, each centre will participate in intensive quality control programs which include both education of the staff that performs the procedures, and quality control and calibration of the equipment. The education and the calibration program will meet the criteria's from ERS and ATS.

Perspectives:

By examining about 3.000 persons this project will globally be the largest study of its kind.

Especially when it comes to the quantity of data for diffusion capacity and pletysmography where earlier studies has examined less than 500 persons and most studies less than 200.

This project will result in a quality boost in the performance of lung function testing across the country:

1) We will have contemporary reference values for the three types of lung function tests most often used in Denmark based on the current population.

2) Unlike today where different departments use different reference values we will have a nationwide consensus about the choice of reference values that we use. It ensures a consistent evaluation of any given lung function value no matter where it is performed.

3) We will ensure that the high standards for quality control established before the beginning of the project will continue after the end of the project.

Peter Kenney, Medical student, Dept of Respiratory Diseases, Aarhus University Hospital

A novel nasal filter, as treatment for hay fever.

Introduction:

Allergic rhinitis is characterised by an inflammation of the nasal mucosa as a result of early- and late-phase IgE-mediated responses to allergens.

It is estimated that more than 20 % of the world's population suffer from IgE-mediated diseases, of which allergic rhinitis is considered most common of all.

The most widespread symptoms of allergic rhinitis include but are not limited to nose blowing, runny nose, sniffles, sneezing, blocked nose and itchy throats.

Principally three courses of treatment exist: 1) allergen avoidance/reducing exposure, 2) pharmacotherapy and 3) immunotherapy. The former is albeit logical in effect often impractical and cumbersome in reality. The latter requires a high degree of patient compliance over a long time period.

Pharmacotherapy is currently the main course of treatment.

My work focuses on evaluating a method for allergen avoidance that might prove more practical than the options available today.

Purpose:

To assess the hypothesis that reducing exposure to allergens in patients with seasonal allergic rhinitis by means of a dedicated nasal filter will significantly minimize the patients' symptoms and improve their quality of life.

Method:

The hypothesis is assessed through a single-centre, randomised (1:1), double-blind placebo-controlled crossover clinical trial conducted in an Environmental Exposure Unit (EEU) in Aarhus, Denmark in December 2012 through January 2013 on 24 participants with seasonal allergic rhinitis.

Results:

None available (01.08.2013).

Conclusions:

None available (01.08.2013)

Perspectives, future research:

Future research will of course depend on the outcome of the study.

On the assumption that the study will show an effect other areas of interest for further research could be 1) larger multi-centre studies on the same patient population, 2) studies on the effect of using nasal filters on other respiratory diseases caused by allergens or other particles, 3) studies investigating any association between minimizing exposure during sleep and allergic reactions during day time.

Mette Nygaard Christensen, MD, PhD student, Dept of Respiratory Diseases, Aarhus University Hospital

Introduction: Dyspnea and cough are symptoms that often send patients to their general practitioner, but the cause of the symptoms is often difficult to establish. Tracheomalacia, a collapse of the central airways, leads to similar symptoms. We hypothesize that tracheomalacia is a common trigger or cause of chronic cough and that tracheomalacia is underdiagnosed.

Purpose:

1. To determine the difference in prevalence of collapse of the central airways during expiration based on 4 different diagnostic tools for MDCT.
2. To develop a confident but yet practical and executable method for MDCT scan of patients undergoing investigations for central airway collapse.
3. To determine the collapsibility of the airways in 3 groups of patients, with suspected high incidence of tracheomalacia, by measuring pulmonary function test, doing bronchoscopy and MDCT.

Method:

1. A retrospective study based on 345 consecutive high-resolution computed-tomography (HRCT) scans with both inspiratory and expiratory imaging. The patients of this cohort were primarily under suspicion of emphysema. Image interpretation: 4 different diagnostic tools for MDCT: 1: manually tracing of the inner-wall giving a cross-sectional area of the airway starting at the carina and then measuring the cross-sectional area in steps of one centimetre to the thoracic outlet in both inspiratory and expiratory imaging. 2: The smallest area in the expiratory imaging will be localized and compared to the area at the same location in the inspiratory imaging. The change will be calculated in percentage of the inspiratory image. 3: Comparison the area 1 cm above the carina in the inspiratory and expiratory imaging finding the change in percentage. 4: The volume of the trachea given by the CT software in inspiration and expiration will be used to calculate a difference in percentage. Each of the four methods for evaluation of collapse of the trachea will be compared and correlated to pulmonary function tests and symptoms in order to identify the most valid method. The prevalence based on each of the four different modalities will be determined.

2. A prospective study. 10 patients undergoing examinations for severe asthma or COPD and 10 patients suspected of tracheomalacia judged on previous scans will be included. Imaging technique**:** Patients will be scanned both at end inspiration and

during forced expiration. The respiratory pattern of the patients will be trained before scanning and during the scan with respiratory manoeuvres to ensure maximal expiration at image acquisition.

3. 3 groups of patients of approximately 30 subjects will be studied in this prospective study. 1. group: patients with COPD, 2. Group: patients with chronic dyspnea, cough and recurrent infections, 3. Group: patients suspected to suffer from laryngeal diseases.

All the patients will be examined according to the established algorithms with pulmonary function tests, bronchoscopy and MDCT.

Results:

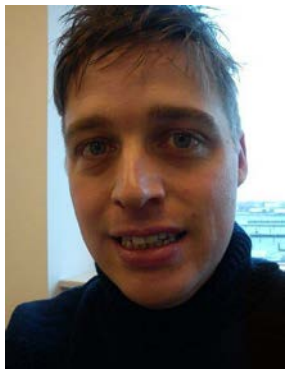
1. The preliminary data from the first study are showing a good correlation between the volumes obtained through manually tracing and the volumes given by the CT software. In the future it might be possible to use the values given by the software for detecting tracheomalacia but further studies and evaluations are still necessary.

The other studies are not yet executed or ongoing.

Perspective/future research: Tracheomalacia is an overlooked condition especially in Denmark. This study will lead to a clarification of the diagnosis of tracheomalacia. Studies from abroad show a high prevalence of this disease in patients with asthma and COPD and in Denmark there is possible a large group of patients suffering from this disease. These patients may suffer from unnecessary medication and benefit from other treatments targeted at tracheomalacia. Thus, this study might change the treatment for asthma and COPD as we know it today.

Tuberculosis

Andreas MF Nielsen, MD, PhD student, Dept of Respiratory Diseases, Aarhus University Hospital



Improving control of tuberculosis in a low-incidence country – observational studies of isolation procedures, contact investigation and social determinants.

Background:

Tuberculosis (TB) remains a major health problem globally. Denmark is a low-incidense country – but active disease transmission is still going

on, and in high-risk environments – especially immigrants and socially marginalized groups – TB is significant course of morbidity. In low-incidense settings, an important tool in fighting the disease is contact investigation procedures: Every time a TB case is detected, contacts of this patient must be evaluated for TB, aiming to find secondary cases as well as the source of transmission to the index case. In USA and Holland, studies have found that actual contact investigation procedures are inconsequent and suboptimal, and not always focused on relevant risk groups.

The relatively stable incidence in Denmark is mainly carried by two mechanism: Immigration of patients from high-incidence countries, and TB transmission among socially marginalized people in Denmark. Poverty is a factor for TB, both globally and within low-incidence countries, reflecting factors like crowding, under/malnutrition, smoking, immune suppression and migration. But little is known about how social and economic variables are influenced by diagnosing and treating TB. Also, we do not know the actual health costs of TB in western countries.

When evaluating and treating TB cases in hospital wards, contagious patients must be isolated to prevent disease transmission to other patients and to the staff. This involes isolation of a major number of TB-suspects, who will turn out not to have TB. Current guidelines for ceasing isolation require microscopy of 3 consecutive sputum samples. PCR-analysis for TB has higher sensitivity for detecting *mycobacterium tuberculosis* than microscopy. If isolation of TB-suspects could be ceased on basis of single-sample PCR analysis, the time spent under stressful isolation regimens could be significantly reduced, when this analysis is negative

Studies:

Study 1: Improving guidelines for ceasing isolation of patients suspected of having TB

Objective: To evaluate whether single-sample PCR result can be used as a parameter to cease isolation of patients suspected of having TB.

Material: All Danish samples investigated for *mycobacterium tuberculosis complex* at Statens Serum Institut, Copenhagen, through a 10-year period (approx. 55.000 samples). Data are obtained from the National Reference Laboratory of Mycobacteriology, Statens Serum Institut.

Method: Patients having at least 3 samples in a 14-day window around the first culture-positive sample, will be selected. Main outcome is percentage of smear-positive, PCR negative patients. Secondary outcomes are the numbers of tests pr. patient, and sensitivity/specificity of PCR analysis.

Study 2: *Evaluating the socio-economic consequences of having TB in Denmark*

Objective: To assess the socio-economic burden of having tuberculosis in Denmark.

Material: The Danish registry systems offer the possibility to tease out the burden and the socio-economic consequences of acquiring tuberculosis.

Methods: TB cases through a 10 year period are obtained from the Danish National Tuberculosis Register. For each of these, four age- and sex-matched controls are obtained. Data on occupational status, receiving social transfer payments, housing conditions and income are obtained, before and after the diagnosis of TB, and after completion of treatment. The impact of TB diagnosis, of treatment for TB, and of treatment outcome on these socio-economic variables will be estimated,

Study 3: Evaluation of contact investigation procedures conducted among active TB cases

Objective: *To evaluate contact tracing procedures performed among TB cases in Denmark through a 10 year period, to quantify 1) the number of contacts identified per index patient, 2) the rate of LTBI and TB among these, and 3) the completeness of data collected during contact investigations.*

Material: For all registered cases of active TB, data on performed contact investigation procedures, the number and closeness of contacts identified and screened, and the results hereof, are obtained from the contact tracing units in Denmark.

Methods: The coverage of contact investigation, as calculated from the data, will be compared to European goals for contact investigation, to assess the cost-effectiveness of these procedures in Denmark.

The study is founded in "TB-Center Vest", and will be performed in cooperation between Department of Pulomonology and Department of Infectious Diseases at Aarhus University Hospital, and Statens Serum Institut, Copenhagen.

Mette K B Dolberg (mettdolb@rm.dk)MD, PhD student, Dept of Respiratory Diseases, Aarhus University Hospital

Magnesium in asthma and chronic obstructive pulmonary disease

Aim: To assess the effect of a daily oral magnesium (Mg) supplement in the treatment of asthma and chronic obstructive pulmonary disease (COPD).

Background: Mg inhibits contraction and relaxes smooth muscles in airways owing to blocking of calcium-ion-flux across the cell membrane. Mg decreases the concentration of neurotransmitters at

motor neuron terminals, lowers the depolarizing effect of acetylcholine at the neuromuscular endplate and dampens the excitability of smooth muscle membranes. Mg deficiency increases the intracellular concentration of calcium and the secretion of histamine and inflammatory cytokines.

A high dietary intake of Mg seems to indicate a better lung function, reduced risk of airway hyper-reactivity and wheezing. The prevalence of hypomagnesemia is increased among chronic asthmatics, and chronic asthmatics with low Mg are more often hospitalized than chronic asthmatics with normal Mg.

The use of Mg is well-established in the treatment of acute asthma attacks. Hence, it seems plausible that Mg may have a beneficial effect on chronic asthma and COPD.

Materials & methods: The study falls into five parts: 1) A review of the role of Mg in the human airway; 2) A pharmacokinetic study (n=10) of oral and intravenous Mg supplementation; 3) Study of the effect of daily Mg supplementation in steroid-resistant, severe asthmatics 4) Study of the effect of daily Mg supplementation upon bronchial hyperreactivity in asthmatics; 5) Study of the effect of daily Mg supplementation in patients with moderate to severe COPD.

Parts 3), 4) and 5) are double-blinded and the participants (n=12, 12 and 18 respectively) are randomized to either 12 weeks with a daily magnesium-supplementation or placebo and cross-over after a 12-week wash-out period.

The primary endpoints in parts 3), 4) and 5) are: The Asthma Control Test (questionnaire providing a measure of disease control), PD20 for metacholine (measure for bronchial hyperreactivity) and the EQ-5D-questionnaire (questionnaire providing a measure of quality of life), respectively.

Results & conclusion: Collection of data will be completed soon. Hence, data remain to be analyzed.

Anne Sofie Bjerrum, MD, Ph.d.-student, Dept of Respiratory Diseases, Aarhus University Hospital

Learning enhancing methods for training bronchoscopy skills.

Background

Bronchoscopy is a procedure that is used in many medical specialties. Until Virtual Reality Simulators emerged, training of this procedure was mainly placed in the clinical setting, resulting in prolonged

procedure time and patient discomfort. The development of Virtual Reality Simulators marked the beginning of a new era in procedural skills training, as simulators offer the opportunity for surgical trainees to obtain basic technical skills in safe and controlled environments, with no risk for the patient and lower levels of stress for the trainee. As simulation equipment and instructor participation is costly, effective training methods are warranted.

The studies within this PhD study examine the effectiveness and efficiency of three different practice methods; Study 1: Distributed practice, Study 2: Dyad practice and Study 3: Modeling examples.

Methods

The three studies were performed as randomized, controlled trails. All participants were novices and had no previous experience with bronchoscopy.

Study 1: Participants were randomized to either distributed bronchoscopy simulation practice within a day or the same amount of training time distributed within three weeks.

Study 2: Participants were randomized to either individual bronchoscopy simulation practice or practice in dyads (participants in the dyad practice group alternated between physical and observational practice, and thereby undertook only half of the training cases physically).

Study 3: All participants were given supervised bronchoscopy simulation training with feedback from an instructor. In addition, the intervention group saw three modeling examples (a physical demonstration of the simulated bronchoscopy, performed by the instructor). These examples lasted 10.5 minutes all together.

Bronchoscopy skills acquisition was assessed immediately after training (post-test) and three weeks after training (retention-test) with previously validated bronchoscopy simulator metrics.

Results

Study 1: Improvement in performance from pre to post- and retention test was seen in both practice conditions. No significant differences were found between the daily distributed practice condition and the weekly distributed practice condition.

Study 2: Improvement in performance from pre-test to post- and retention test was seen in both practice groups. No differences in simulator metrics scores were found between the dyad- and individual practice group at post- and three-weeks-delayed retention tests.

Study 3: A clear learning curve was observed for both groups. The intervention group outperformed the control group on all parameters but one. For the primary outcome measure, the modeling group achieved a 46% higher score at post test, and a 43% higher score at retention test.

Conclusion

Study 1: We found a daily distributed practice schedule as effective for bronchoscopy skills acquisition as a weekly distributed practice schedule.

Study 2: Dyad practice and individual practice were equally effective for learning bronchoscopy skills through simulation training. However, dyad practice was more efficient than individual practice as two participants practising in dyads learned as much as one individually

practicing participant without increasing instructor resources or time scheduled for training.

Study 3: Integrating modeling examples into the curriculum of bronchoscopy simulation training optimizes the role of the instructor and enhances novices´ learning outcomes, presumably by optimizing cognitive load during training.

Research in progress

A transfer study based on the 3 studies is in progress.

Åsa Lina Jønsson, MD, Ph.d.-student, Dept of Respiratory Diseases and dept of Pharmcology, Aarhus University Hospital

Characterization and role of mutations in the SLC34A2 gene/NaPi-IIb in pulmonary alveolar microlithiasis and extrapulmonary calcification syndromes

Background

We have recently described new mutations in the SLC34A2 gene of two patients with pulmonary alveolar microlithiasis (PAM) from the Department of Respiratory Medicine and Allergology, Aarhus University Hospital. In addition to PAM, one of the patients had severe arteriosclerosis, and the other patient had aortic valve calcification, leading to a significant hemodynamically relevant aortic stenosis. Extrapulmonary calcifications have earlier been reported in different cases of PAM.

PAM is a rare lung disease characterized by deposition of spherical calcium phosphate concretions in the alveoli. Mutations in the SLC34A2 gene, that encodes a sodium phosphate (NaPi) co-transporter (NaPi-IIb), are considered to be the cause of the disease. In the lung, NaPi-IIb is expressed in alveolar type II cells. Loss of function of the gene due to mutations may lead to a decreased cell-uptake of phosphate, which in turn may lead to formation of calcium-phosphate concretions in the alveoli.

NaPi-IIb is a member of the sodium-phosphate cotransporter family SLC34. These cotransporters are expressed in several tissues, and play a major role in the homeostasis of inorganic phosphate.

Calcific aortic valve disease (CAVD) is characterized by thickening and calcification of the aortic valve leaflets. Valvular calcium deposits are shown to contain both calcium and phosphate. We have recently identified SLC34A2 mRNA in a human aortic valve.

Deposition of calcium phosphate or other calcium salts in the dermis and subcutaneous tissue are seen in a rare skin syndrome, called calcinosis cutis. The pathophysiology of the disorder is

incompletely understood. We have, as the first, recently identified SLC34A2 mRNA in human skin. However, the role of SLC34A2/NaPi-IIb in calcinosis cutis remains to be elucidated.

Hypothesis and Aim

Mutations in the SLC34A2 gene cause defect cell-uptake of phosphate, and leads to formation of calcium-phosphate concretions in the lungs, as seen in PAM. **The hypothesis of the present project is that mutations in SLC34A2 play a role in the development of calcifications in other tissues and organs.** The hypothesis is addressed by the following specific aims:

(a) Investigate the association between new mutations in the SLC34A2 gene in PAM patients and the function of the mutated NaPi-IIb, and to characterize the mutated NaPi-IIb with regards to localisation within the cell.

(b) Investigate the possible expression of SLC34A2 in extrapulmonary tissues from patients with various diseases with calcifications and healthy subjects, and to investigate if mutations in the SLC34A2 may be involved in these diseases.

Materials and Methods

Three studies will be performed to test the hypothesis:

Study 1: Mutations in the SLC34A2 gene from patients with PAM will be selected for the investigations. The mutations will be reproduced by site-directed mutagenesis and cloned into a plasmid. Opossum kidney cells and rat lung epithelial cells will thereafter be transfected with the generated mutants. Localisation of the mutated NaPi-IIb within the cell will be studied by confocal microscopy. Furthermore, the generated mutants will be microinjected into frog oocytes, with subsequent electrophysiology investigations of the Pi uptake.

Study 2: A comparative study in patients with PAM from Denmark will be performed in which pneumocytes type II cells obtained by Bronchoalveolar lavage (BAL) will be compared to skin biopsies, to investigate whether the changes in localization and function of NaPi-IIb is changed to the same degree in these cell types.

Study 3: We will investigate if calcinosis cutis is associated with mutations in SLC34A2 and impaired NaPi-IIb expression. Skin biopsies from patients with calcinosis cutis will be obtained to address, if there is an association between calcium-phosphate deposition and impaired expression of NaPi-IIb in the skin. Furthermore, the patients will be tested for possible mutations in the SLC34A2 gene.

Collaboration

Cand. Med., PhD student Åsa Lina M. Jönsson[1], B.Sc. Susie Mogensen[1], Associate professor, PhD Jane Hvarregaard Christensen[2], Chief physician, PhD Elisabeth Bendstrup[3], Chief physician, Dr. Med. Ole Hilberg[3], Chief physician, Associate professor, PhD Anne Braae Olesen[4], Professor, Dr. Med., Carsten Wagner[5], Professor, Dr. Med. Ulf Simonsen[1]

1) the Department of Biomedicine -Pharmacology, Aarhus University, Denmark

2) the Department of Biomedicine – Human Genetics, Aarhus University, Denmark

3) the Department of Respiratory Medicine and Allergology, Aarhus University Hospital, Denmark

4) the Department of Dermatology and Venereology, Aarhus University Hospital, Denmark

5) the Institute of Physiology, University of Zurich, Switzerland

Hans Jürgen Hoffmann, Professor, Ph.D., Department of Respiratory Diseases

Mechanisms of Allergy

IgE-mediated Allergy is a major chronic disease that affects up to 20% of any western society. Mast cells are the hub of the allergic response. They express the high affinity receptor FceRI that is charged with IgE. In an allergic patient, IgE is specific for allergens and allergen crosslinking of IgE activates mast cells. Our research focuses on the mechanism of activation in cultured human mast cells.

Cultured mast cells can be charged with known IgE. This gives us the opportunity to separately investigate the contribution of the cell and of IgE, and explore the significance of composition of IgE. We have developed a culture model that gives rise to mast cells with an allergic phenotype; they express FceRI in a terminal fashion, and respond strongly to allergen-crosslinking. We use this model to investigate maturation, competency to invoke an allergic response, activation and attenuation of mast cells on a large scale with tools of systems biology.

Blood basophil granulocytes also express FceRI, and respond similarly to mast cells to crosslinking by IgE and allergen. We have developed the Basophil Activation Test for clinical use as a diagnostic and monitoring tool. Key findings made with cultured mast cells can be confirmed with blood basophils, as well as with mast cells purified from biopsies.

Treatment of allergy with allergen immunotherapy induces a combination of initial desensitisation of mast cells and immune deviation as the long-term effect. We are exploring a novel treatment

variant, where allergen is injected into the lymph node, where immune deviation occurs. Advantages of this intra lymphatic immunotherapy are that it is shorter (3 visits over 2 months in contrast to 30 visits over 3 years), and that less allergen is used reducing the risk of side effects.

We are currently treating 7 patients in an open trial, and will be treating 36 patients in a prospective, randomised trial in 2014. If the treatment holds what it promises, we would be able to change the profile of allergy in Aarhus within the next 5 years similar to the way eradication of Tuberculosis changed health profile 100 years ago.

Allergy diagnosis and monitoring gives an additional, objective indicator to support a clinical decision. Allergen immunotherapy has most effect on the first allergy that is the strongest risk factor for additional allergies. The earlier allergy is treated, the more effective and preventive of further allergies will treatment be. Reduction of invasive treatment of allergy and side effects are especially important in children and adolescents, as treatment may not be their decision.

Important national and international collaborators

Professor Vibeke Backer, Bispebjerg Hospital, University of Copenhagen, DK

Senior Scientist Peter Adler Würtzen, ALK-Abelló, Hørsholm, DK

Professor Jonas Erjefält, Unit of Experimental Lung Research, University of Lund, SE

Ass. Prof. Gunter Sturm, University of Graz, Austria

Prof Werner Pichler, University of Bern, Switzerland

Ass Prof Becky Vonakis, Johns Hopkins University, Maryland, USA

Projects

1. Analysis of cultured human mast cell maturation, competency, activation and attenuation. We have developed a method to culture allergic mast cells from adult blood stem cells, and are investigating their phenotype and function in type I allergy.

2. Intralymphatic immunotherapy monitored as basophil sensitivity, plasma cell frequency and clinical effect. We are exploring intralymphatic immunotherapy as a fast alternative to conventional immunotherapy. We have observed significant plasma cell changes in individual patients.

3. Attenuation of blood basophils to investigate the effect of desensitisation. We have developed a strong model for attenuation of blood basophils that can be ported to mast cells to investigate mechanisms underlying it.

4. BasoSCIT: Changes in specific immunoglobulin and basophil activity during subcutaneous immunotherapy. We have monitored treatment of 18 patients with SCIT with BAT and other methods. Basophil sensitivity was changed throughout the observation period.

5. Diagnosis and monitoring of allergy by basophil activation. We use BAT as a diagnostic tool in addition to less demanding tools when required in complex or severe allergy, as well during treatment of severe allergy with anti-IgE.

Milestones

2007 Introduction of Basophil Activation Test as a clinical diagnostic tool

2008 Comparison of short term in vitro cultured human mast cells from different progenitors - Peripheral blood-derived progenitors generate highly mature and functional mast cells.

(Andersen HB, J Immunol Methods; 336: 166)

2010 Basophil sensitivity through CD63 or CD203c is a functional measure for specific immunotherapy (Mikkelsen et al, Clin Mol Allergy.; 8: 2)

2011 Cultured Human mast cells are heterogenous for expression of the high- affinity IgE receptor FcεRI. (Hoffmann HJ, et al. Int. Arch Allergy Immunol: 157: 246)

2013 Cultured mast cells from asthmatic patients and controls respond with similar sensitivity to recombinant Der P2 induced, IgE-mediated activation (Krohn et al, Scand J Immunol. doi: 10.1111/sji.12085)

2013 Basophil sensitivity to allergen decreases rapidly after starting subcutaneous immunotherapy but increases slowly again during maintenance therapy (Schmid et al, Allergy Supplement)

Methods

- We do clinical trials, clinical research and basic research conceptually close to patients.

- Placebo-controlled, randomised, double blind clinical trials of treatment effects in allergy

- Diagnosis of asthma, rhinitis, urticaria, food and drug allergy

- Respiratory laboratory: extended lung function analysis, exercise testing

- Allergy provocation tests of lung, nose and conjunctiva

- Cell culture and cell biological analyses

- Biomarker analyses by ELISA, ELISPOT and flow cytometry

- Functional assays and phenotypic characterisation by multicolour flow cytometry

Contact

Hans Jürgen Hoffmann, Professor, ph.d.

Contact: hans.jurgen.hoffmann@ki.au.dk

Head of lab. research in respiratory medicine and allergology

s

Ole Hilberg, Chief Physician, Associate Professor, Dr.Med.

Contact: olehilbe@rm.dk

Specialist in respiratory medicine. Research area has been focused on allergology in upper airways and development of acoustic rhinometry. Recently on interstitial lung diseases and medical education. Supervisor for phd students, in charge of postgraduate medical education.

Torben Riis Rasmussen, Chief Physician, Associate Professor, phd.

Email: torbrasm@rm.dk

Specialist in respiratory medicine, head of lung cancer outpatient clinic

Tina Skjold, Chief Physician, Associate Professor, phd.

Email: tinskj@rm.dk

Specialist in respiratory medicine and allergology, head of outpatient clinic in allergology

Elisabeth Bendstrup, Chief Physician, Associate Professor, phd.

Email: kabend@rm.dk

Specialist in respiratory medicine. Research area during the last years has mainly been pulmonary fibrosis and rare lung diseases. Member of international commitees on pulmonary fibrosis. Supervisor for phd students.

Anders Løkke, Consultant, Associate Professor

Email: aloekke@gmail.com

Specialist in respiratory medicine

Johannes Schmid, Chief Physician, Associate Professor,

Specialist in respiratory medicine and head of Respiratory Physiologic Laboratory

Email: johaschm@rm.dk

Pernille Hauschildt, Chief Physician, Associate Professor phd, Head of clinical department.

Email: pernhaus@rm.dk

Birgitte Folkersen, Chief Physician,

Specialist in respiratory medicine, head of endoscopy section

Email: birgfolk@rm.dk

Pia Pedersen, Secretary

Contact for pharmaceutical companies

piapeder@rm.dk

Information for Pharmaceutical companies

Laboratory – Department of Respiratory Medicine and Allergology, AU

Building facilities

The laboratory is located in the diagnostic building of the Department of Respiratory Medicine, in close proximity to the out-patient clinics for allergy and general pulmonary medicine, provocation chambers, the respiratory physiology laboratory and the bronchoscopy facility.

The laboratory consists of two rooms. One is primarily for sample handling and administration, the other is for cell biological research specialised in lymphocyte handling. We have access to freezers at -20C and -80C outside the lab. These are locked and monitored and can be replaced within 4 hours of instability due to freezer failure. All instruments in the laboratory are covered by maintenance plans to ensure optimal performance and minimise down time.

Staff

The chief of the laboratory is professor in basic pulmonary medicine and allergology. The laboratory is manned by four certified technicians (study nurses and lab-technicians). In addition, 2 – 4 students normally use the laboratory. A secretary is responsible for administration.

Biochemistry and cell research

The laboratory performs the cell biological tests T-spot.TB (2000 p.a.) and the Basophil activation test (70 p.a.) on a daily basis, and can perform analyses on a Phadia 100 – currently, specific IgE, total IgE and ECP are measured on demand, and tryptase is measured routinely (200 p.a). BAL CD4/CD8 ratio of T cells is performed on demand (200 p.a.). The T-spot.TB is evaluated with an AID Elispot reader to optimise quality.

In the preclinical cell biological research section, the laboratory has equipment for cell culture; a HETO sterile workbench, a CO2-controlled humidified incubator for tissue culture and two low speed centrifuges (Eppendorf).

For multicolour flow cytometry we have a Beckton Dickinson CANTO II with 3 lasers and 8 parameters and an Attune from Life Sciences with violet and blue lasers and six parameters. We have established 8 parameter protocols for the CANTO. Data is kept on the University Network, and is backed up daily.

We have a cytospin centrifuge, equipment for ELISA (plate washer, spectrophotometer) and a pH meter.

Clinical research

The present study nurses and labtech's have performed clinical trials since 1995 and are certified in GCP. They are highly professional and skilled and have conducted almost 100 clinical phase II, III and IV trials in the fields of COPD, asthma, allergy, smoking cessation, interstitial lung diseases, and physiological trials.

The equipment available comprises 3 body boxes for pulmonary function tests, spirometry equipment, ECG-equipment, ergospirometry, bronchoscopes, laryngoscopes, ultrasonic equipment, NO analysers, challenge equipment and ABL.