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MEETING ABSTRACTS

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## Pirfenidone inhibits TGF- $\beta$ 1-induced extracellular matrix production in nasal polyp-derived fibroblasts

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**Background:** Pirfenidone has been shown to have anti-fibrotic and anti-inflammatory effects in the lungs. The purpose of this study was to evaluate the inhibitory effects of pirfenidone on transforming growth factor (TGF)- $\beta$ 1-induced myofibroblast differentiation and extracellular matrix accumulation. We also determined the molecular mechanism of pirfenidone in nasal polyp-derived fibroblasts (NPDFs).

**Methods:** NPDFs were isolated from nasal polyps from eight patients with chronic rhinosinusitis with nasal polyp. Pirfenidone was used to treat TGF- $\beta$ 1-induced NPDFs. Cytotoxicity was evaluated using a 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-tetrazolium bromide assay. Fibroblast migration was evaluated with scratch assays. Expression levels of  $\alpha$ -smooth muscle actin (SMA), fibronectin, and phosphorylated Smad2/3 were determined by western blot and/or reverse transcription-polymerase chain reaction and immunofluorescence staining. Total collagen production was analyzed with the hydroxyproline assay and contractile activity was measured by a collagen contraction assay.

**Results:** Pirfenidone (0 – 2 mg/ml) has no significant cytotoxic effects in TGF- $\beta$ 1-induced NPDFs. Migration of NPDFs was significantly inhibited by pirfenidone treatment. The expression levels of SMA and fibronectin were significantly reduced in pirfenidone-treated NPDFs. Collagen contraction and production were also significantly decreased by pirfenidone treatment. Finally, pirfenidone significantly inhibited phosphorylation of Smad2/3 pathway in TGF- $\beta$ 1-induced NPDFs.

**Conclusion:** Pirfenidone has an inhibitory effect on TGF- $\beta$ 1-induced myofibroblast differentiation ( $\alpha$ -SMA), extracellular matrix accumulation, and collagen contraction by blocking the phosphorylation of Smad2/3 pathways in NPDFs. Thus, pirfenidone may inhibit extracellular matrix by regulating Smad2/3.

## Efficacy of a 2-week course of oral steroid in the treatment of chronic spontaneous urticaria refractory to antihistamines

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**Background:** Antihistamines are the mainstay in the treatment of chronic spontaneous urticaria (CSU), some CSU patients have not responded to antihistamines. Many clinicians have accepted the efficacy of steroids in the treatment of CSU. There is, however, little evidence supporting steroid use in antihistamine-resistant CSU. The

purpose of this study was to demonstrate the efficacy of, and suggest a regimen for, oral steroid in the treatment of CSU patients who were refractory to a high dosage of antihistamines. We conducted a retrospective chart review of all patients diagnosed with urticaria between February 1, 2012, and December 31, 2014. A total of 98 patients with CSU were included. Of these, 16 patients (16.3%) were antihistamine-resistant and prescribed a 2-week course of steroid. Thirteen patients (81.2%) were successfully controlled with antihistamines only after stopping the first course. Second course of steroid induced remission additionally in two patients (12.5%). No adverse events and complications associated with oral steroid were observed over the study period. This study demonstrated the excellence of a 2-week course of oral corticosteroid in antihistamine-resistant CSU and propose standardized corticosteroid treatment regimen.

## A3

### The altered distribution of follicular t helper cells may predict a more pronounced clinical course of primary sjögren's syndrome

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Recent studies emphasized the important role of follicular T helper (T<sub>FH</sub>) cells, which contribute to B-cell differentiation, as well as antibody production. The aim of our study was to investigate the possible role of T<sub>FH</sub> cells in the pathogenesis of primary Sjögren's syndrome (pSS). In the first part of the study, we focused on the periphery by analyzing immune-competent cells and serological markers. We enrolled 50 pSS patients and 16 healthy controls in the study. Patients had elevated ratio of peripheral T<sub>FH</sub> cells, however, when dividing patients into two groups defined by the presence of extraglandular manifestations (EGMs), only patients with EGMs differed from controls significantly. Moreover, T<sub>FH</sub> cell percentages correlated positively with both activated T cell and Tr1 cell values, but T<sub>FH</sub> cell percentages showed negative correlation with both IgM and IgG memory B cell proportions. Elevated T<sub>FH</sub> percentages were observed in the anti-SSA/SSB positive patients. In the second part, we concentrated on the site of the inflammation, and determined the composition of lymphocyte infiltration in labial salivary gland (LSG) biopsies with special emphasis on T<sub>FH</sub> cells. We selected tissue blocks obtained from 10 patients at the time of disease onset. LSGs were graded based on the organizational level of periductal lymphocytic infiltrates. T<sub>FH</sub> cell markers occurred predominantly in more organized structures with higher focus scores. The co-expression of CD3 and Bcl-6 markers identified T<sub>FH</sub> cells close to Bcl-6<sup>+</sup>B cells with the typical formation of germinal centers. Systemic features were developed later in the disease course only in patients with more structured infiltrates.

Our results indicate that the presence of T<sub>FH</sub> cells in LSGs at the disease onset may predict a more pronounced clinical course of pSS. We expect that the further understanding of the regulation of T<sub>FH</sub> cells will provide new potential therapeutic targets in the treatment of pSS patients with EGMs.

with corticosteroid for 4 weeks. **Results:** The sensitivity of methacholine and adenosine bronchial provocation test in diagnosis of bronchial asthma are 65% and 60%, respectively. Both PC20 methacholine and PC20 adenosine are improved after corticosteroid treatment. Divided into two groups according to severity, in mild group, improvement in PC20 methacholine ( $p<0.05$ ) after corticosteroid treatment was found but not in PC20 adenosine. In moderate group, improvement in PC20 adenosine ( $p<0.05$ ) after corticosteroid treatment was found but not in PC20 methacholine. Pre-treatment [pre-] MMP-9 was correlated with PC20 methacholine and PC20 AMP ( $p<0.05$ ;  $p<0.05$ ). Strong correlations between pre- IL-8 and pre-MMP-9 ( $Rho=0.655$ ,  $p<0.05$ ) and between pre- IL-8 and pre-TIMP-1 ( $Rho=0.815$ ,  $p<0.001$ ) was observed. We found that changes in PC20 methacholine and PC20 adenosine after treatment with inhaled corticosteroid are correlated with eosinophil percentage in blood and pre-TIMP-1 in sputum, respectively ( $Rho=0.488$ ,  $p<0.05$ ;  $Rho=0.464$ ,  $p<0.05$ ). All inflammatory markers such as IL-5, IL-8, MMP-9, TIMP-1 and ECP decreased after treatment but no significance was found. **Conclusions:** Our findings show that PC20 adenosine and PC20 methacholine are useful markers to assess the efficacy of anti-inflammatory therapy. In moderate persistent asthma, PC20 adenosine may be more valuable to evaluate the efficacy of treatment than PC20 methacholine.

## A661

#### S100 Calcium Binding Protein A9 in Sputum of Patients with Steroid Naive Asthma: Relation with Airway Obstruction and Neutrophilic Inflammation

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**Background:** We previously reported elevation of S100 calcium binding-protein A9 (S100A9) protein in sputum of neutrophilic severe uncontrolled asthmatics in spite of high dose inhaled corticosteroid treatment compared with stable asthmatics. The aim of this study was to evaluate the relation of S100A9 protein with neutrophilic inflammation in the steroid naive - mild to severe asthmatics and lung function. **Materials and Methods:** Sputum were obtained from 132 never-smoking or ex-smoking asthmatics (<10 pack-years). S100A9 proteins were measured in inhaled or systemic corticosteroid naive asthmatic (SNBA, n=103), inhaled and systemic steroid treated asthmatic (STBA, n=29) and normal controls (NC, n=35) using an enzyme-linked immunosorbent assay. Correlations between S100A9 levels and inflammatory cells, FEV1, and annual rate of exacerbation were analyzed. **Results:** The S100A9 levels were significantly higher in sputum of both SNBA and STBA than that of normal controls ( $P=0.008$ , and  $0.001$  respectively) and were comparable between SNBA and STBA ( $P=0.734$ ). In bronchial asthma (BA), S100A9 levels were significantly correlated with the percentages of neutrophils ( $r=0.267$ ,  $p=0.002$ ) and those of eosinophils ( $r=-0.195$ ,  $p=0.025$ ) in sputum. The correlations were persistently observed in SNBA. When BA was divided into 4 groups: Neutrophil dominant (70% or more), eosinophil dominant (3% or more), co-dominant and pauci-granulocytic group. The S100A9 levels was the highest in the neutrophilic group (13.51 pg/ug) compared to the eosinophilic (9.06 pg/ug), co-dominant (5.96 pg/ug) and pauci-granulocytic (7.87 pg/ug) groups. S100A9 levels was significantly increased in the subject experiencing exacerbation compared to those not experiencing exacerbation. **Conclusion:** Our data suggest that S100A9 protein was elevated in sputum of asthma compared to normal control. The levels were correlated with percentage of neutrophil in sputum and were regardless of steroid treatment.

## A662

#### Risk Factor Asthma in Pediatric Pneumonia Patients

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## Abstract

**Background:** Asthma is a risk factor of pneumonia in children. Children with asthma are 40% more likely to have pneumonia, especially hospitalizations, especially children under five years old. Childhood pediatric pneumonia has characteristic clinical symptoms with increased need of mechanical ventilation and mortality.

**Objective:** To compare the asthma risk and the number of hospitalizations with pneumonia in Hasan Sadikin General Hospital, Bandung, Indonesia.

**Method:** Retrospective study on pediatric pneumonia with asthma as a comorbidity at Pediatric Ward Department, Hasan Sadikin General Hospital from January to December 2012. We reviewed all patient hospitalized with pneumonia and asthma using ICD-9 (J11-J18 and J45).

**Results:** Eight hundred and seventy three children with pneumonia were admitted to the hospital. Seven hundred and thirty six patients (84.3%) had comorbidities, and thirty six patients (4.1%) had asthma. Twenty pneumonia patients with asthma (2.3%), 1 patient (0.1%) had asthma and congenital heart disease, and 15 patients (1.2%) had asthma and other comorbid such as cerebral palsy, anemia, malnutrition, diarrhea, and pulmonary tuberculosis. Median age was 402 months (11 to 144 months old), with 22 of them (22.2%) were under five years old, and 22 patients were male (61.1%). Six patients didn't have history of allergy/asthma in their family, and 4 patients had one asthma exacerbation in last 4 weeks. Six patients had previous hospitalization with similar symptoms. One of them received previous controller medication. Laboratory finding revealed white blood cell 5500-24.500/mm<sup>3</sup>, only 1 bacteria was found from blood culture which was *Serratia marcescens*, and chest X-ray shows bilateral infiltrate. Eight patients (22.2%) had severe asthma exacerbation. Length of stay was 1 to 8 days. There was no patient admitted to pediatric intensive care unit or perished.

**Conclusion:** Asthma was still a risk factor for children to developing pneumonia that required hospitalization.

## A663

#### Prevalence and Risk Factors of Childhood Asthma and Allergic Disease at Exposed Area By Emission of Cement Padang Factory

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**Background.** Prevalence of asthma in children is increasing, especially in the industrial city. Padang city has Padang cement factory which is located near from community where people surrounding can inhale the dust pollutant emission from cement production, including children. We want to know the prevalence and risk factor of asthma and allergic disease in this area.

**Methods.** During May-June 2015, we conduct a cross sectional study to children around the Cement Padang factory, based on distance of house from the factory, <5 km (exposed area) and > 10 km (no exposed area). Children age from 0-15 years were selected. We used ISAAC questionnaire to determine asthma and allergic disease. Data was taken from the parent, with informed consent, including asthma, allergic rhinitis and atopic dermatitis symptoms and some risk factors. Data was analyzed with chi-square test.

**Results.** This study found 90 children, 43 (47.8%) from exposed area, most of them were female 46 (51.1%). The most age group is 1-5<sup>th</sup>(48.8%). More than half (54%) have normal nutritional status, but 33.3% undernourish. Asthma prevalence and atopic dermatitis was found higher in no exposed area than exposed area (19.5% vs 13.9% and 39.1% vs 37.2%), but allergic rhinitis was higher in exposed area (60.4% vs 58.6%), no significance difference. Atopic history of the mother was higher in exposed area (34.8% vs 17.3%). Other inhaled pollutant like pet, cigarettes smoke, kapok mattress, vapor from gasoline were higher in no exposed area, but no significance difference.

**Conclusion.** In exposed area of Cement Padang factory, the prevalence of rhinitis allergy was higher than asthma and atopic dermatitis, but no significance difference.