**Title:** Assessment of ATP degradation in bronchoalveolar lavage fluid

Dr. Zsofia 25147 Lazar zsofia.lazar@yahoo.com MD 1,2, Marisa 25148 Braun marisa.braun@uniklinik-freiburg.de 1, Anja 25149 Meyer anja.meyer@uniklinik-freiburg.de 1, Jessica 25150 Beckert jessica.beckert@uniklinik-freiburg.de 1, Prof. Ildiko 25151 Horvath hildiko@elet2.sote.hu MD 2 and Prof. Marco 25193 Idzko marco.idzko@uniklinik-freiburg.de MD 1. 1 Dept. Pulmonology, University Clinic Freiburg, Freiburg, Germany, 79106 and 2 Dept. Pulmonology, Semmelweiss University, Budapest, Hungary, 1125.

**Body:** Introduction: Extracellular adenosine triphosphate (ATP) via purinergic signalling plays a role in the development of airway inflammation. Elevated ATP concentration in airway samples was reported in inflammatory diseases. Airway ATP level is under tight control by enzymatic degradation, which could also affect ATP concentration measured in ex vivo biological samples. Aim: To assess ATP degradation in bronchoalveolar lavage (BAL) fluid. Methods: ATP was measured in BAL collected from 20 subjects (6 patients with interstitial lung diseases, 4 with asthma, 2 with COPD, 1-1 with extrinsic allergic alveolitis / Wegener granulomatosis / microaspiration / pleuritis / pulmonary hypertension / pneumonia and 2 healthy controls) within an hour after collection using luminometry. ATP degradation was assessed as the recovery of 1 µM ATP after 30 minutes. Seven samples were also collected on chelating solutions (0.32% sodium citrate or 2 mM EDTA+2 mM EGTA). Data were analyzed with non-parametric tests (median / interquartile range /). Results: BAL fluid ATP concentration was 18 nM /4-107 nM/, and recovery of added ATP was 36% /29-82%/.

ATP concentration was higher in samples treated with citrate (530 nM /375-715 nM/) or EDTA+EGTA (420 nM /234-828 nM/) compared to untreated samples (186 nM /39-302 nM/; p<0.05). ATP degradation was inhibited by both citrate (recovery: 99% /50-100%/ and EDTA+EGTA (recovery: 80% /53-100%/ in comparison with no treatment (p<0.05). Conclusion: Extracellular ATP is degraded at a variable speed in BAL fluid, which is effectively inhibited by chelating agents. The analysis of pre-treated BAL fluid might more precisely reflect airway ATP concentration.