New imaging approach to evaluate the in vivo administration of nanostructured mycophenolic acid in a preclinical model of SSC-ILD

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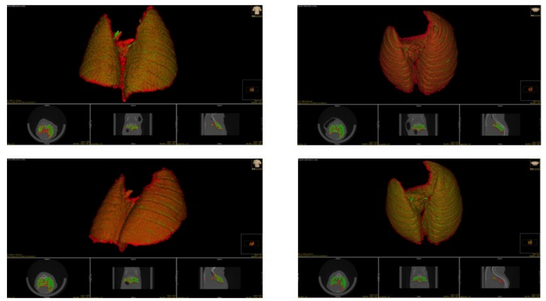
Background and Aim

Systemic Sclerosis (SSc) is an uncommon disease affecting several tissues and internal organs including lungs (ILD), causing gas exchange impairment due to pulmonary fibrosis.

To date ILD management include immunosuppressors such as oral administration of high daily doses of mycophenolate mofetil (MMF) . This systemic treatment can lead to side effects such as bacterial or viral infection, bruising and gastrointestinal bleeding [1, 2].

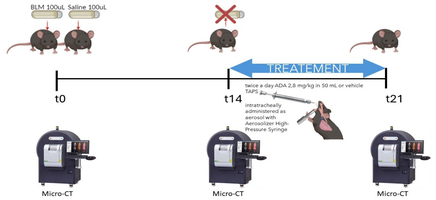
To avoid these systemic side effects, A.D.A srl developed a treatment with a nanostructured prodrug of MMF (named ADA). The efficacy of ADA was tested in a mice preclinical model of mild ILD.



Lung's parenchyma oxygenation, green: well aerated; red: poorly aerated.

Materials and Methods

To induce ILD, 6-weeks old C57BL/6 female mice were implanted with two osmotic micro pumps (Alzet model 1007D DUCRET™) loaded with Bleomycin (BLM) or with saline (Sham) for 14 days. On day 14 micro pumps were explanted and mice were further divided into 4 groups to be treated twice a day with 2,8 mg/kg in 50 µL for the experimental compound ADA or relative vehicle (TAPS) as follows: SHAM + TAPS; SHAM + ADA; BLM + TAPS; BLM+ ADA.



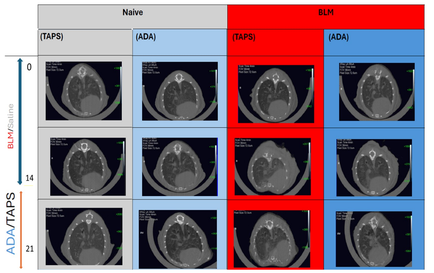
Obtained 3D images were segmented and analyzed with Analyze 14.0 in order to evaluate lung well aerated region (>50% air/tissue ratio) and lung poorly aerated region (<50% air/tissue ratio) [3]. This method allowed us to analyze in vivo ILD onset and the potential therapeutic effect of ADA during all the experimental time course. At day 21 mice were euthanized, bronchoalveolar lavage fluid (BALF) and lungs were collected for histological and for further biological assays.

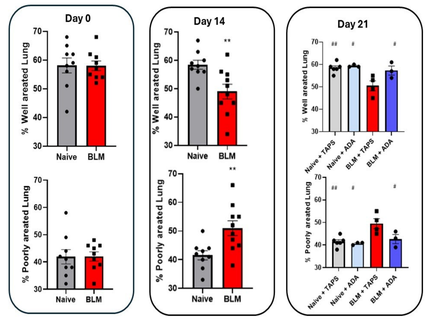
Results

At baseline (day 0) mice didn't show any difference in lung aeration status, while at day 14, BLM mice reported a body weight loss and a decrease in the percentage of well aerated area (49%) compared with sham mice (59%) indicating a proper induction of mild ILD.

When ADA compound was administered for 7 days (from day 14 to 21) to sham mice, no toxic effects were observed in terms of tissue aeration.

Notably, one weeklong administration of ADA compound to BLM mice led to complete reversion of ILD dependent lung tissue aeration impairment: at day 21, indeed, we observed a l...g well aerated percentage of 50% for BLM+TAPS while BLM+ADA group recovered to 57% (p<0,05) a value like Sham mice (58%).





All data are indicated as mean ± SEM
***=p<0,01 vs Naive (unpaired t-test).*
#=p<0,05 vs BLM + TAPS ##=p<0,01 vs BLM + TAPS (ANOVA with Dunnet Post-test).

Discussion and Future Perspective

This innovative approach in evaluating lungs aeration status *in vivo* at different time points highlights how a pulmonary administration of a nanostructured MMF prodrug can revert ILD-dependent symptoms. This preliminary data encourages us to deepen our investigation at molecular level analyzing the activation of inflammatory and profibrotic pathways in lungs and testing the efficacy of nanostructured ADA carried with low doses of antifibrotic drugs such as nintedanib.

References:
[1]Bergamasco et al. Clinical epidemiology, (2019).
[2]Pope et al. Nature reviews. Rheumatology, (2023).