A novel disruptive IgE inhibitor: Efficacy assessment in non-human primate and human precision-cut lung slices

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Introduction

- Asthma affects 334 million people worldwide. To counteract IgE-dependent asthmatic symptoms, the new disruptive inhibitor DARPin bi53_79 was developed. It binds free and releases receptor-bound IgE.
- To test new therapeutics specific for human IgE, suitable animal models are needed.
- Precision-cut lung slices (PCLS) offer a promising tool to assess early airway reaction (EAR) ex vivo.

Hypothesis:

- It is possible to sensitize mast cells in PCLS of humans, cynomolgus macaques (Macaca fascicularis), and common marmosets (Callithrix jacchus).
- Early airway reaction can be counteracted by omalizumab and the novel, disruptive IgE inhibitor DARPin bi53_79.

Methods

- Lungs were filled with 1.5 % agarose and medium and processed with a Krumdieck tissue slicer to achieve precision-cut lung slices (PCLS) containing central airways.
- For passive sensitization, PCLS duplicates were incubated in human allergic plasma containing IgE directed against house dust mite (HDM). Control PCLS were placed in human non-allergic plasma.
- Omalizumab was incubated overnight. Additionally, Omalizumab, DARPin bi53_79, or control DARPin were incubated for 2 h before HDM challenge. After HDM exposure, the airway area of PCLS was recorded for 20 minutes and supernatants were analyzed for histamine release.

Results

1A Human 1B Marmoset 1C Cynomolgus

![Figure 1. Allergen provocation induces bronchoconstriction in PCLS.](image)

Incubation in human allergic plasma resulted in HDM-induced reduction of initial airway area from 100% to 31.9 ± 8.0% in human (1A), 74.0 ± 7.8% in marmoset (1B), and 70.7 ± 7.9% in cynomolgus PCLS (1C). In comparison to PCLS incubated in non-allergic plasma, histamine levels were only increased in sensitized PCLS (1D) (human: 30.8 ± 18.0 ng/ml; marmoset: 2.0 ± 0.7 ng/ml; cynomolgus: 21.4 ± 5.6 ng/ml; mean ± SEM).

2A Omalizumab overnight incubation 2B Omalizumab 2 h incubation

![Figure 2. Anti-IgE inhibitor Omalizumab does not prevent bronchoconstriction in pre-sensitized human PCLS.](image)

5 µM Omalizumab prevented bronchoconstriction when incubated during sensitization of human PCLS (2A). In contrast, an incubation for 2 h did not inhibit bronchoconstriction (2B). n=3, mean ± SEM.

3A Human PCLS + DARPin 2h 3B Marmoset PCLS + DARPin 2h 3C Cynomolgus PCLS + DARPin 2h

![Figure 3. Preincubation with DARPin bi53_79 prevents bronchoconstriction and reduces histamine release.](image)

Figure 3. Preincubation with DARPin bi53_79 prevents bronchoconstriction and reduces histamine release. Bronchoconstriction was prohibited at a concentration of 5 µM in human (3A) and cynomolgus (3C) PCLS and 0.5 µM in marmoset PCLS (3B), respectively.

Conclusion

- Non-human primate PCLS and human PCLS can be passively sensitized with human allergic plasma and are a model for early airway reaction (EAR).
- Omalizumab only inhibits EAR if inoculated during sensitization.
- The anti-IgE DARPin bi53_79 prevents EAR in human, marmoset, and cynomolgus PCLS.
- Data of cynomolgus PCLS are comparable to those of human.

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