

Glycan-specific IgG anti-IgE autoantibodies contribute to protectivity against allergic diseases.

Paul Engeroff^{1,2}, Kevin Plattner^{1,2}, Federico Storni^{1,2}, Franziska Thoms⁴, Kayluz Frias Boligan⁵, Lukas Muerner⁵, Alexander Eggel^{1,2}, Stephan von Gunten⁵, Martin F. Bachmann^{1,2,6} and Monique Vogel^{1,2}

¹University Hospital for Rheumatology, Immunology, and Allergology, University of Bern, Bern, Switzerland; ²Department of BioMedical Research, University of Bern, Bern, Switzerland; ⁴Department of Dermatology, Zurich University Hospital, Schlieren/Zurich, Switzerland; ⁵Institute of Pharmacology, University of Bern, Bern, Switzerland; ⁶Nuffield Department of Medicine, The Jenner Institute, University of Oxford, Oxford, UK.

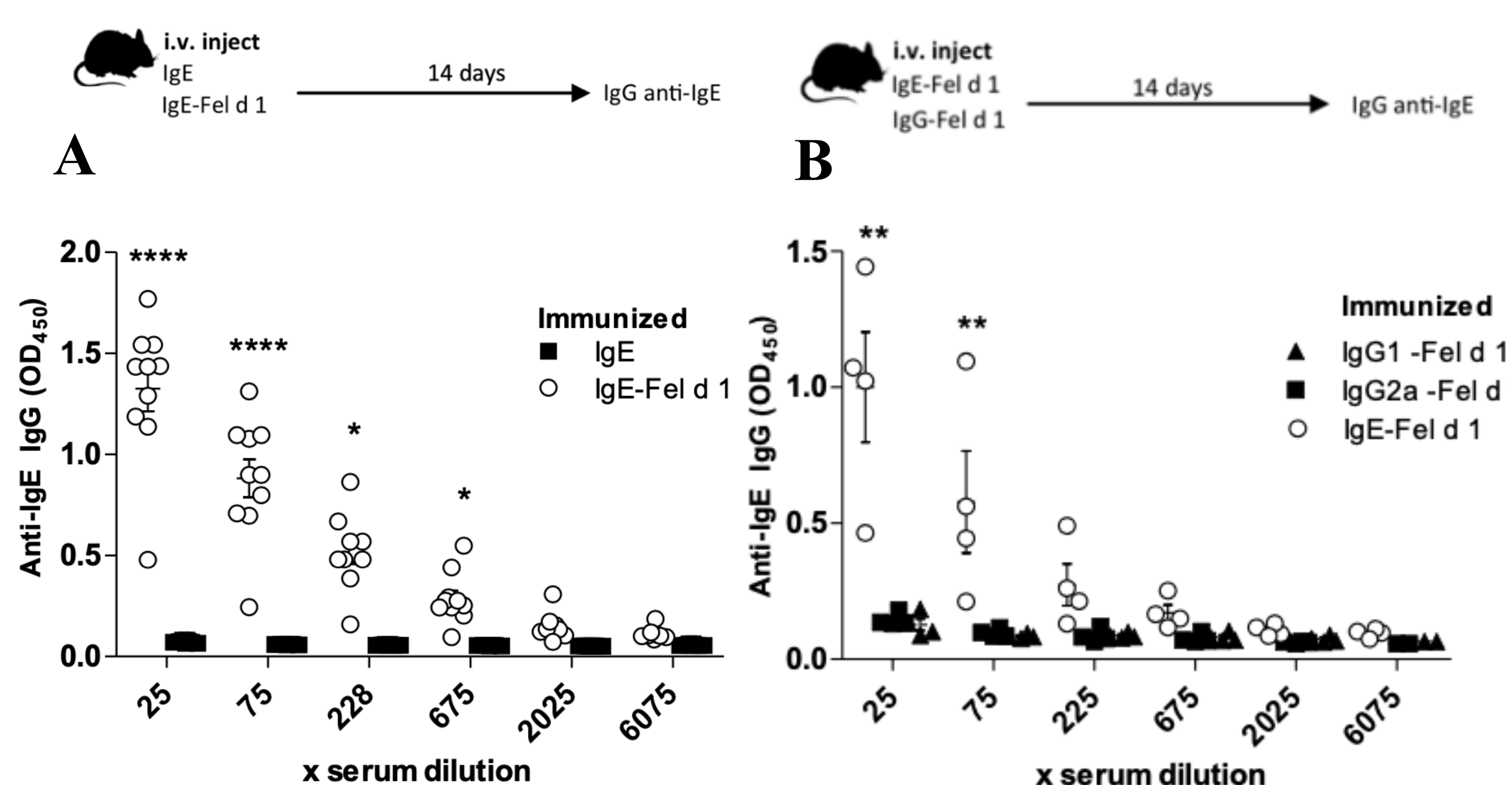
INTRODUCTION

Allergies have become a severe problem worldwide. They are mainly driven by the sensitization of effector cells with IgE. Studies have shown that IgE-specific IgG autoantibodies exist and that they may also exert regulatory functions. It is, however, still not clear how these anti-IgE antibodies are induced.

To investigate this, naïve mice were immunized with different forms of IgE either as IgE alone or as IgE in form of complex with the major cat allergen Fel d 1. We then tested their anti-IgE responses and analyzed the effect of those antibodies on their *in vitro* and *in vivo* impact on anaphylaxis. Finally, we investigated anti-IgE autoantibodies in human sera.

RESULTS

INDUCTION OF ANTI-IgE ANTIBODIES



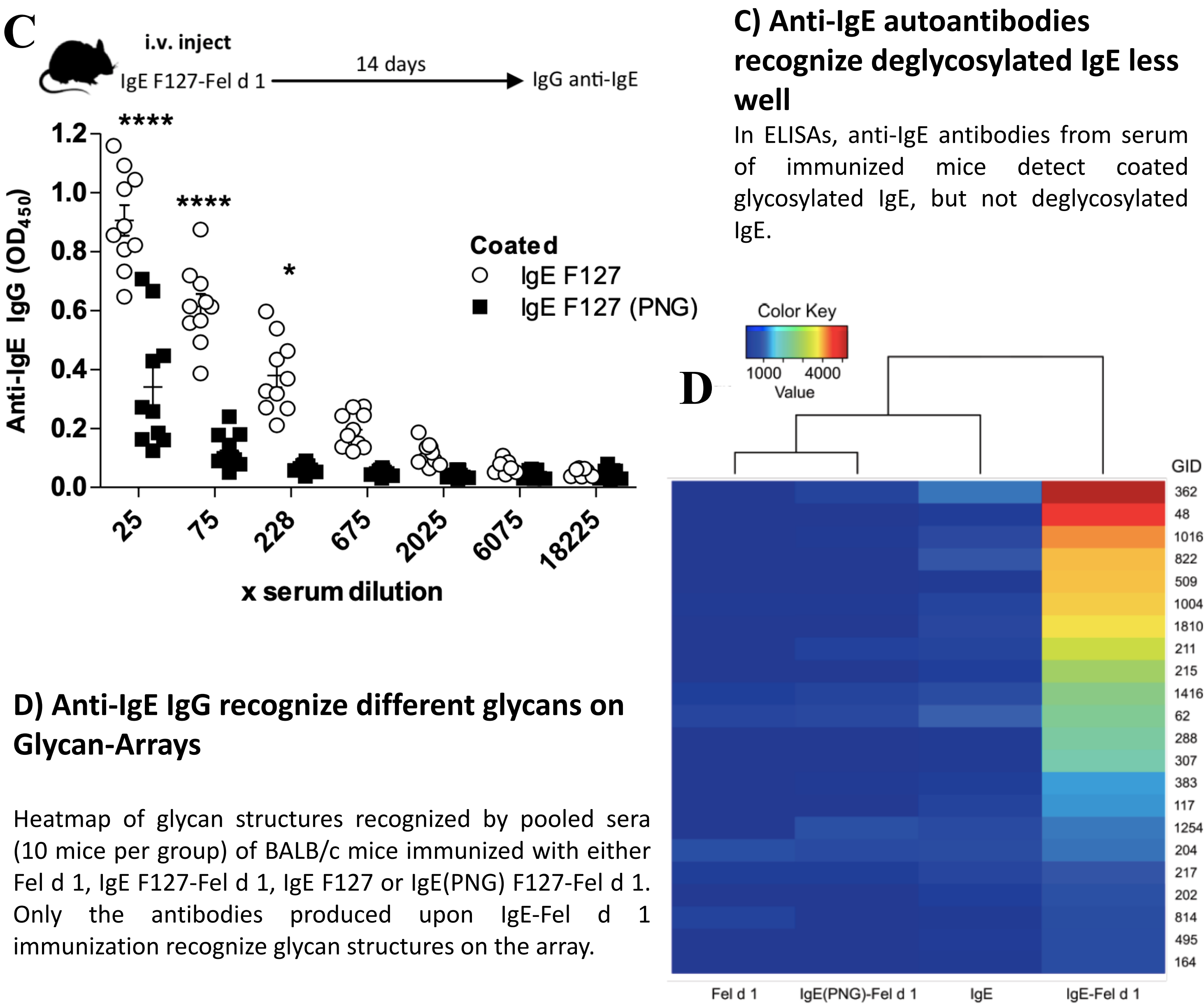
A) IgE in complex with allergen induce anti-IgE IgG

Mice were immunized with the monoclonal IgE F127 alone or in complex with the major cat allergen Fel d 1. IgE in complex with an allergen induces anti-IgE IgG antibodies, but IgE alone does not induce an autoantibody response.

B) IgG in complex with allergen does not induce anti-IgE IgG

No anti-IgE autoantibodies are induced by immunizing the mice with IgG antibodies in complex with Fel d 1.

ANTI-IgE ANTIBODIES ARE GLYCAN SPECIFICITY



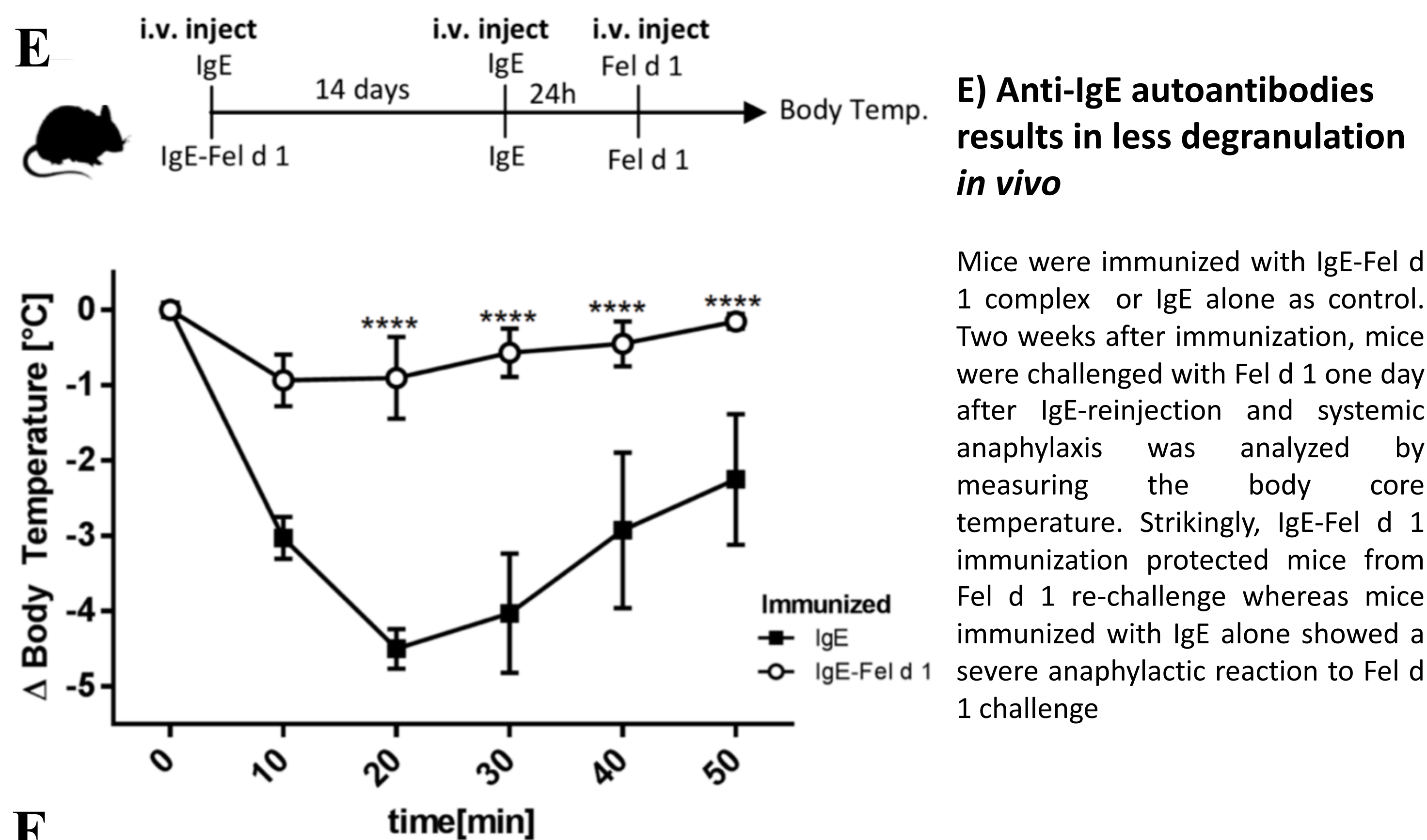
C) Anti-IgE autoantibodies recognize deglycosylated IgE less well

In ELISAs, anti-IgE antibodies from serum of immunized mice detect coated glycosylated IgE, but not deglycosylated IgE.

D) Anti-IgE IgG recognize different glycans on Glycan-Arrays

Heatmap of glycan structures recognized by pooled sera (10 mice per group) of BALB/c mice immunized with either Fel d 1, IgE F127-Fel d 1, IgE F127 or IgE (PNG) F127-Fel d 1. Only the antibodies produced upon IgE-Fel d 1 immunization recognize glycan structures on the array.

PROTECTIVE ANTI-IgE ANTIBODIES



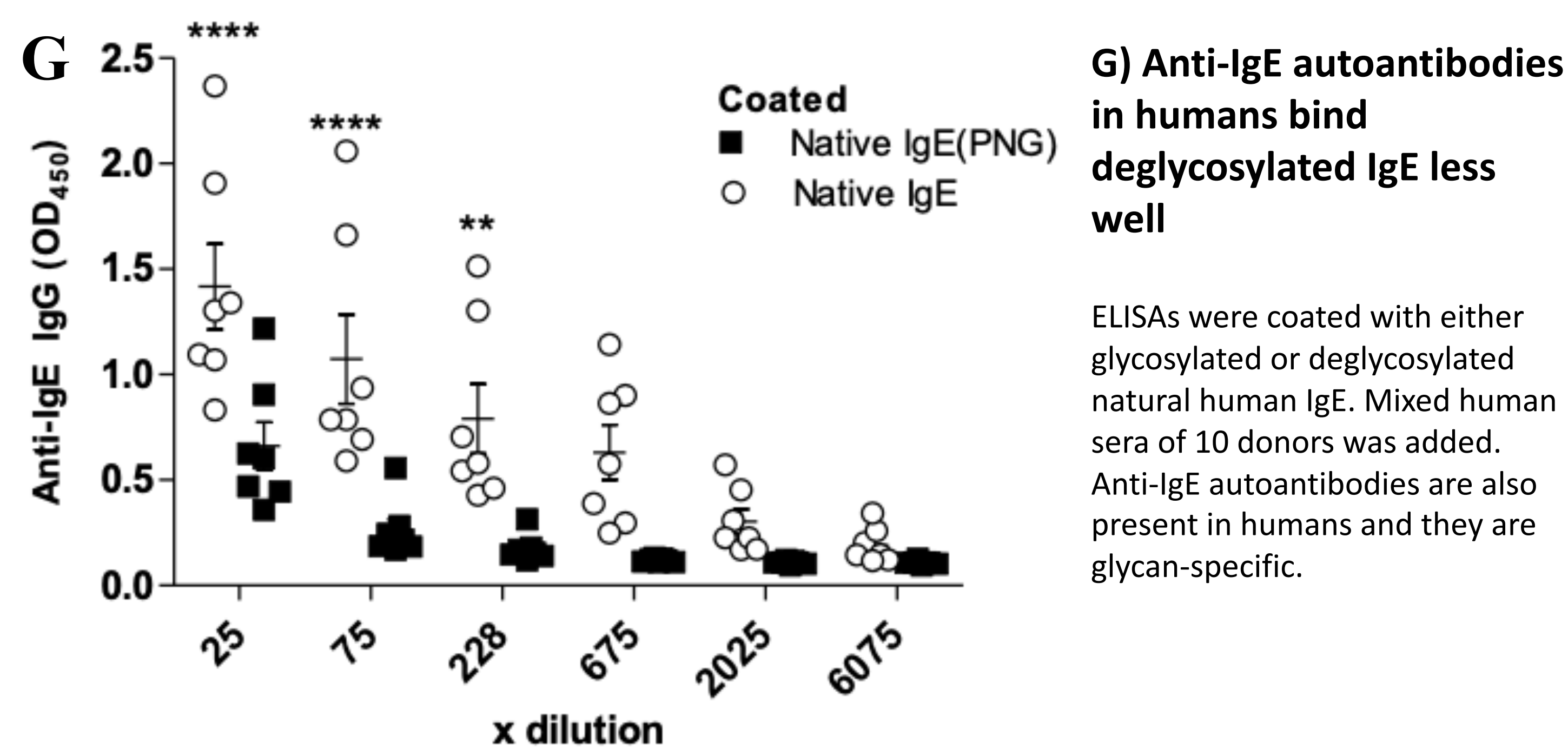
E) Anti-IgE autoantibodies results in less degranulation *in vivo*

Mice were immunized with IgE-Fel d 1 complex or IgE alone as control. Two weeks after immunization, mice were challenged with Fel d 1 one day after IgE-reinjection and systemic anaphylaxis was analyzed by measuring the body core temperature. Strikingly, IgE-Fel d 1 immunization protected mice from Fel d 1 re-challenge whereas mice immunized with IgE alone showed a severe anaphylactic reaction to Fel d 1 challenge.

F) Anti-IgE autoantibodies results in less degranulation of bone marrow derived mouse mast cells *in vitro*

Upon Fel d 1 challenge, BMMCs incubated with IgE and anti-IgE serum displayed reduced degranulation as measured by CD63 up-regulation compared to BMMCs incubated with IgE and sera of naive mice (control serum).

GLYCAN SPECIFIC ANTI-IgE ANTIBODIES ARE PRESENT IN HUMANS



G) Anti-IgE autoantibodies in humans bind deglycosylated IgE less well

ELISAs were coated with either glycosylated or deglycosylated natural human IgE. Mixed human sera of 10 donors was added. Anti-IgE autoantibodies are also present in humans and they are glycan-specific.

CONCLUSION

- Glycan-specific anti-IgE autoantibodies are inducible by IgE-immune complex immunization.
- These anti-IgE antibodies protect from anaphylaxis.
- Glycan specific anti-IgE antibodies, which are also present in humans, might provide first evidence on their role of IgE regulation.

Contact: kevin.plattner@dbmr.unibe.ch