Department of Pediatric Pneumology & Immunology
Charité–Universitätsmedizin Berlin

DISSERTATION

Evolution of serum IgE and IgG antibodies to 35 molecules in childhood

Zur Erlangung des akademischen Grades
Doctor medicine (Dr. med.)

vorgelegt der Medizinischen Fakultät
Charité – Universitätsmedizin Berlin

von

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Datum der Promotion: 01.03.2019

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Abstrakt

HINTERGRUND: Die häufig beschriebene Schutzfunktion allergenspezifischer IgG-Antikörper wurde vor kurzem durch Daten ergänzt, die ein Differentialbild für atopische und nicht-atopische Patienten sowie eine "(vor-) atopische" IgG-Antwort beschreiben. Abgesehen von der Route und der Expositionsdosis können die physikalisch-chemischen Eigenschaften von Proteinen für die Unterscheidung zwischen Antigenen und Allergenen entscheidend sein.

ZIEL: Wir haben versucht, die Längsentwicklung von Allergen-spezifischen IgG- und IgE-Profilen von einer breiten Gruppe von allergenen Molekülen in der Kindheit zu untersuchen.

METHODEN: Die deutsche Multicenter-Allergie-Studie untersuchte eine Geburtskohorte, die 1990 geboren wurde. Blutproben wurden im Alter von 1, 2, 3, 5, 6, 7, 10, 13 und 20 Jahren gesammelt. Die Teilnehmer wurden in die vorliegende Analyse aufgenommen, wenn sie (1) ≥1 Serumprobe im Alter von 1-3 Jahren hatten; (2) ≥2 Serumproben im Alter von 5-7 Jahren und (3) eine Serumprobe im Alter von 10 Jahren. Kinder wurden als "atopisch" betrachtet, wenn sie eine positive IgE-Antwort (Cut-off ≥ 0,30 ISU) auf mindestens ein Molekül zeigten, während diejenigen, die IgE niemals zu irgendeinem untersuchten Molekül produzierten, als "Nicht-Atopics" gruppiert wurden. IgE- und IgG-Antikörper gegen native und rekombinante allergene Moleküle wurden mit einem Multiplex-Mikroarray-Ansatz (ImmunoCAP ISAC TM -103 und ISAC TM -112, TFS, Schweden) getestet. Fünfunddreißig Moleküle wurden für die Analyse nach Ausschlusskriterien ausgewählt, die zuvor validiert wurden.

ERGEBNISSE: Wir fanden eine differentielle Evolution der IgG- und IgE-Reaktionen auf einen breiten Satz von Allergenmolekülen: 1) IgE-Reaktionen waren meist gegen eine eingeschränkte Gruppe von luftgetragenen Molekülen gerichtet, mit einer Sequenz- und Prävalenzhierarchie, die - auf Populationsniveau - weitgehend während des ganzen ersten Lebensjahres beibehalten: Phl p 1> Bet v 1> Feld d 1> Phl p 5> Der p 2> Der p 1; 2) Im Gegensatz dazu war das IgG-Repertoire viel breiter und verbreitete sich bei den meisten Kindern zuerst zu tierischen lebensmittelbasierten Molekülen, dann zu pflanzlichen Lebensmitteln und schließlich zu luftgetragenen Molekülen; 3) Eine ungewöhnlich starke und anhaltende IgG-Antwort geht einer IgE-Antwort auf das gleiche luftgetragene Molekül vor oder begleitet sie.

SCHLUSSFOLGERUNGEN: Obwohl die Evolution von allergenspezifischen IgG- und IgE-Reaktionen im Laufe der Kindheit stark abweicht, geht eine starke IgG-Antwort voran oder begleitet das Auftreten von IgE bei atopischen Probanden. Die Ergebnisse deuten darauf hin, dass die Erzeugung von hochaffinitätsallergenspezifischem IgE in den meisten Fällen eine intermediäre IgG_1-Phase und sequentielle Klassenumschaltung ($\mu \rightarrow \gamma_1 \rightarrow \varepsilon$) erfordert.

Abstract

BACKGROUND: The often described protective role of allergen-specific IgG antibodies has been recently complemented by data describing a differential picture for atopic vs. non-atopic patients, as well as a „(pre-)atopic“ IgG response. Apart from the route and dose of exposure, the physico-chemical properties of proteins may be decisive for the distinction between antigens and allergens.

OBJECTIVE: We sought to investigate the longitudinal evolution of allergen-specific IgG and IgE profiles to a broad panel of allergenic molecules in childhood.

METHODS: The German Multicentre Allergy Study examined a birth cohort born in 1990. Blood samples were collected at the ages of 1, 2, 3, 5, 6, 7, 10, 13 and 20 years. Participants were included in the present analysis if they had (1) ≥1 serum sample at age 1-3ys; (2) ≥2 serum samples at age 5-7ys and (3) a serum sample at age 10ys. Children were considered „atopic“ if they exhibited a positive IgE response (cut-off ≥ 0.30 ISU) to at least one molecule, while those never producing IgE to any oft he examined molecules were grouped as „non-atopics“. IgE and IgG antibodies to native and recombinant allergenic molecules were tested with a multiplex microarray approach (ImmunoCAP ISAC™-103 and ISAC™-112, TFS, Sweden). Thirty five molecules were selected for the analysis according to exclusion criteria previously validated.

RESULTS: We found a differential evolution of the IgG and IgE responses to a broad set of allergen molecules: 1) IgE responses were mostly directed against a restricted group of airborne molecules, with a sequence and prevalence hierarchy that - at population level - is largely maintained throughout the whole first decade of life: Phl p 1 > Bet v 1 > Feld d 1 > Phl p 5 > Der p 2 > Der p 1; 2) In contrast, the IgG repertoire was much broader, spreading in most children first to animal foodborne molecules, then to vegetable foodborne and finally to airborne molecules; 3) An abnormally strong and persistent IgG response precedes or accompanies an IgE response to the same airborne molecule.

CONCLUSIONS: Although the evolution of allergen-specific IgG and IgE responses throughout childhood differs widely, a strong IgG response precedes or accompanies the appearance of IgE in atopic subjects. The results suggest that the generation of high affinity allergen-specific IgE needs, in most cases, an intermediary IgG_1 phase and sequential class switching ($\mu \rightarrow \gamma_1 \rightarrow \varepsilon$).

Affidavit

"I, Xinyuan Huang, certify under penalty of perjury by my own signature that I have submitted the thesis on the topic '**Evolution of serum IgE and IgG antibodies to 35 molecules in childhood**' I wrote this thesis independently and without assistance from third parties, I used no other aids than the listed sources and resources."

All points based literally or in spirit on publications or presentations of other authors are, as such, in proper citations (see "uniform requirements for manuscripts (URM)" the ICMJE www.icmje.org) indicated. The sections on methodology (in particular practical work, laboratory requirements, statistical processing) and results (in particular images, graphics and tables) correspond to the URM (s.o) and are answered by me. My interest in any publications to this dissertation correspond to those that are specified in the following joint declaration with the responsible person and supervisor. All publications resulting from this thesis and which I am author correspond to the URM (see above) and I am solely responsible.

The importance of this affidavit and the criminal consequences of a false affidavit (section 156,161 of the Criminal Code) are known to me and I understand the rights and responsibilities stated therein.

Date Signature

<u>Detailed Declaration of Contribution</u>

Xinyuan Huang had the following share in the following publication:

He planned the study design, organized and supervised the conduction of this study and performed the analysis and interpretation of the results. He additionally collected and interpreted the eQTL data. He wrote the manuscript.

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Zepp[6], MD, Antje Schuster[7], MD, Raffaele D'Amelio[3], MD, Ulrich Wahn[1], MD, Thomas Keil[8,9], MD, Susanne Lau[1], MD, and Paolo Maria Matricardi[1*], MD; Evolution of the IgE and IgG repertoire to a comprehensive array of allergen molecules in the first decade of life; Allergy. 2018;73:421–430. https://doi.org/10.1111/all.13269

Signature, date and stamp of the supervising University teacher

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Signature of the doctoral candidate

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EXCERPT OF THE JOURNAL SUMMARY LIST
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Rank	Full Journal Title	Total Cites	Journal Impact Factor	Eigenfactor Score
1	JOURNAL OF ALLERGY AND CLINICAL IMMUNOLOGY	46,218	13.081	0.083210
2	**ALLERGY**	**16,206**	**7.361**	**0.025050**
3	Journal of Allergy and Clinical Immunology-In Practice	1,653	5.317	0.006190
4	CLINICAL AND EXPERIMENTAL ALLERGY	10,959	5.264	0.015390
5	CLINICAL REVIEWS IN ALLERGY & IMMUNOLOGY	2,403	5.263	0.005400
6	CONTACT DERMATITIS	5,712	4.335	0.004280
7	PEDIATRIC ALLERGY AND IMMUNOLOGY	3,787	3.775	0.006840
8	CURRENT ALLERGY AND ASTHMA REPORTS	2,071	3.735	0.005170
9	ANNALS OF ALLERGY ASTHMA & IMMUNOLOGY	6,970	3.728	0.008690
10	IMMUNOLOGY AND ALLERGY CLINICS OF NORTH AMERICA	1,463	3.610	0.002560
11	Current Opinion in Allergy and Clinical Immunology	2,861	3.463	0.006330
12	Clinical and Translational Allergy	636	3.239	0.002340
13	ALLERGOLOGY INTERNATIONAL	1,487	3.194	0.003270
14	JOURNAL OF INVESTIGATIONAL ALLERGOLOGY AND CLINICAL IMMUNOLOGY	2,073	3.094	0.002550
15	Allergy Asthma & Immunology Research	1,094	2.957	0.003140
16	Allergy Asthma and Clinical Immunology	850	2.869	0.002140
17	INTERNATIONAL ARCHIVES OF ALLERGY AND IMMUNOLOGY	5,175	2.720	0.006600
18	ALLERGY AND ASTHMA PROCEEDINGS	1,937	2.614	0.003140
19	JOURNAL OF ASTHMA	3,201	1.746	0.005060
20	Postepy Dermatologii i Alergologii	418	1.683	0.000910
21	ALLERGOLOGIA ET IMMUNOPATHOLOGIA	954	1.439	0.001550
22	ASIAN PACIFIC JOURNAL OF ALLERGY AND IMMUNOLOGY	698	1.011	0.001040
23	Pediatric Allergy Immunology and Pulmonology	171	0.958	0.000640
24	Iranian Journal of Allergy Asthma and Immunology	457	0.812	0.000850
25	Revue Francaise d Allergologie	276	0.363	0.000130
26	ALLERGOLOGIE	187	0.311	0.000080

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THE SELECTED PUBLICATION

Xinyuan Huang, Olympia Tsilochristou, MD, Serena Perna, M.Sc., Stephanie Hofmaier, Antonio Cappella, MD, Carl-Peter Bauer, MD, Ute Hoffman, MD, Johannes Forster, MD, Fred Zepp, MD, Antje Schuster, MD, Raffaele D'Amelio, MD, Ulrich Wahn, MD, Thomas Keil, MD, Susanne Lau, MD, and Paolo Maria Matricardi, MD;

Evolution of the IgE and IgG repertoire to a comprehensive array of allergen molecules in the first decade of life;

Allergy. 2018;73:421–430. https://doi.org/10.1111/all.13269

Curriculum Vitae

My curriculum vitae does not appear in the electronic version of my paper for reasons of data protection.

Complete list of publications

Xinyuan Huang, Olympia Tsilochristou, MD, Serena Perna, M.Sc., Stephanie Hofmaier, Antonio Cappella, MD, Carl-Peter Bauer, MD, Ute Hoffman, MD, Johannes Forster, MD, Fred Zepp, MD, Antje Schuster, MD, Raffaele D'Amelio, MD, Ulrich Wahn, MD, Thomas Keil, MD, Susanne Lau, MD, and Paolo Maria Matricardi, MD;

Evolution of the IgE and IgG repertoire to a comprehensive array of allergen molecules in the first decade of life;

Allergy. 2018;73:421–430. https://doi.org/10.1111/all.13269

Acknowledgements

I would like to sincerely thank my adviser Dr. Paolo M. Matricardi, for giving this great chance to work with him and his team on allergen molecules and mobile health technology, for his enthusiasm and professionalism in the supervision on my work. I have really learned so much from him and his team during the two years in Berlin, not only science, but also culture and language and so on. I have spent an excellent time in the two years and all of these experience will be the best memory in my life.

Then, I would like to thank all my colleges of our team. I acknowledge Olympia Tsilochristou, Serena Perna, Stephanie Hofmaier, Daniela Posa, Arasi Stefania, Eissa Hakimeh, Sveva Castelli, Marco Di-fraia, Carla Mastrorilli, Alina Schwarz, Azahara Rodriguez Dominguez, Rohrbach Alexander, for keeping their kindness with me all the time. They give me encouragement and helpful advices. I cannot forget all of these forever.

In addition, I would like to thank Zhonghua Helmke, Pamela Glowacki and Franziska Grimm, the officers of Charité International Cooperation (ChIC) and Charité Welcome Center. Thank you for solving the life problems and public affairs, making me live easily in Berlin.

Moreover, I would like to thank my Chinese friends Ning Zhou, Hong Chen, Liming Sheng, Yong Liu, thank you for your kind and friendship.

Of course, I must thank the leaders of my hospital (Shengzhou People’s Hospital) who made the decision and supported me to study in Berlin. They gave me this great opportunity and fund to study in Charité Universitätsmedizin Berlin three years ago. Thank you for your trust.

Finally, I would like to thank my parents, my wife and my son, my brother, and my cousin who works in Leipzig, for their love and support throughout my stay in Berlin. I love you all.