

Autoantibodies in disorders of the brain

Citation for published version (APA):

Zong, S. (2019). *Autoantibodies in disorders of the brain: expanding the spectrum*. [Doctoral Thesis, Maastricht University]. Maastricht University. <https://doi.org/10.26481/dis.20191106sz>

Document status and date:

Published: 06/11/2019

DOI:

[10.26481/dis.20191106sz](https://doi.org/10.26481/dis.20191106sz)

Document Version:

Publisher's PDF, also known as Version of record

Please check the document version of this publication:

- A submitted manuscript is the version of the article upon submission and before peer-review. There can be important differences between the submitted version and the official published version of record. People interested in the research are advised to contact the author for the final version of the publication, or visit the DOI to the publisher's website.
- The final author version and the galley proof are versions of the publication after peer review.
- The final published version features the final layout of the paper including the volume, issue and page numbers.

[Link to publication](#)

General rights

Copyright and moral rights for the publications made accessible in the public portal are retained by the authors and/or other copyright owners and it is a condition of accessing publications that users recognise and abide by the legal requirements associated with these rights.

- Users may download and print one copy of any publication from the public portal for the purpose of private study or research.
- You may not further distribute the material or use it for any profit-making activity or commercial gain
- You may freely distribute the URL identifying the publication in the public portal.

If the publication is distributed under the terms of Article 25fa of the Dutch Copyright Act, indicated by the "Taverne" license above, please follow below link for the End User Agreement:

www.umlib.nl/taverne-license

Take down policy

If you believe that this document breaches copyright please contact us at:

repository@maastrichtuniversity.nl

providing details and we will investigate your claim.

Appendix I Valorization

Note: Different from a formal discussion on the academic way, I would like to write the valorization part into a story.

5 years ago in 2014, when the first time I had a skype with Prof. Pilar Martinez, who was an assistant professor then, and she said there was an ongoing project about detecting neuronal autoantibodies in psychiatric disorders in her group, I should have never imagined that one day, we would search those autoantibodies from neurological patients with psychiatric symptoms to psychosis, schizophrenia, bipolar, depression and anxiety! Nevertheless, now we found that a subgroup of patients with anxiety potentially had a relation to novel neuronal surface autoantibodies! At the beginning, I was fascinated immediately when Dr. Carolin Hoffmann, who was still a PhD student, and later became my colleague and one of my closest friend in Maastricht, explained me the hypothesis that there might be a subgroup of psychiatric patients who were actually caused by neuronal autoantibodies and thus could be treated by immunotherapy. As a resident in neurosurgery department, I personally faced the question that why some patients developed psychiatric symptoms after traumatic injury or head surgeries and never gave my patients a really satisfying answer. I knew the etiology of psychiatric disorders were so diverse and hard to clearly mapped. I thought, wow, this would be a chance to approve a direct causative of those sticky disorders, at least in some of them. Then I decided to come and gave a shot. In the second year in 2015, our first-round test of anti-NMDAR autoantibodies in psychosis turned out to be negative [1]. Still, I brought our idea to a famous hospital in Beijing to seek collaboration, I met Prof. Guan. He welcomed me well and gave me a lot of positive feedbacks including sharing their autoantibodies positive samples to us, which in the end did not work out because of the ethical rules. He also asked me a question which I remembered for a while: “Do you really think those autoantibodies could be found in purely psychiatric patients? You may go too far”. At that time, I said I was not sure.

If we had a chance to go back to 12 years ago, when 2007, Prof. Dalmau described the first cases-series of NMDAR autoantibodies encephalitis [2], I guess at that moment, no one could imagine that his research was actually lightening the whole field of autoantibodies mediated central nervous disorders and followed by the discovery of more than 13 novel neuronal surface autoantibodies [3]! Autoimmune encephalitis is a rare disease. When we apply the concept of neuronal autoantibodies mediated disorder to psychiatric disorders, it is still a rare condition as indicated in **Chapter 4** and **Chapter 5** as well as previous studies [4, 5]. Be as it may, it is already a solid fact of their existence. There is no second condition that ties psychology and neurology so close that specialists from both fields are working together to gain the knowledge about it and thus helping the patients. Some patients have already benefited from it and the trend of enclosing more input in research is going on. Another aspect is using those functional autoantibodies as a tool to study the basic biological changes in psychiatric disorders [6]. Those basic mechanism might be common in psychiatric patients without autoantibodies. In this way, not only the patients who have autoantibodies but all would benefit from it.

Over the years, there are contradictories exist in this field as we described in **Chapter 1**, **Chapter 2** and **Chapter 3**. One of the main possible reasons is the methodology problems. I worked years in developing and comparing the autoantibody detecting methods as showed in **Chapter 4**, **Chapter 5** and **Chapter 6** and deeply understand the method gaps exist between researchers, labs and countries. The development of new techniques is coming up but still needs

to be optimized, the communication between researchers is ongoing but still needs to boost and the barriers between countries have never been broken. Even though, I personally have seen lot of improvements already. Last November on a lancet summit, I asked Prof. Dalmau of the diagnostic value of Immunohistochemistry method he developed, which led the findings of all the novel neuronal autoantibodies in his group. He mentioned he always used this method in his lab for diagnosis and believed that it still was a very useful method in finding unknown autoantibodies [7]. I also met Prof. Guan there. Different from the opinion he gave years ago, his group brought 3 posters all of which gave a large space for psychiatric symptoms. He also wrote a letter to explain the pictographs of encephalitis in Chinese characters in Lancet Neurology this year [8]. He showed his social medium later that they actually worked very closely with psychiatrists from Beijing and tried to diagnose some anti-NMDAR encephalitis at the early stage. Another thing has to mention, their lab has already put anti-GAD65 tests in the routine neuronal autoantibody test panel which they did not perform before. What will be the future of other even rare neuronal autoantibodies that have not been covered by commercial kits? As we showed in current dissertation, before better methods developed, the tissue-based assay is still a good choice.

All in all, as a novel field, our current research takes out the first step. It emphasizes the importance of continuously studying autoimmunity in neuropsychiatric disorders, the comparing between methods and developing new techniques as well as the communicating between researchers, researchers to clinicians and societies. Still there are many confusions that need to be clarified in the future. It would be of great that we look back 10 years later and I am looking forward to it!

Reference

1. de Witte, L.D., et al., *Absence of N-Methyl-D-Aspartate Receptor IgG Autoantibodies in Schizophrenia: The Importance of Cross-Validation Studies* *N-Methyl-D-Aspartate Receptor IgG Autoantibodies in Schizophrenia* Letters. JAMA Psychiatry, 2015. **72**(7): p. 731-733.
2. Dalmau, J., et al., *Paraneoplastic anti-N-methyl-D-aspartate receptor encephalitis associated with ovarian teratoma*. Annals of neurology, 2007. **61**(1): p. 25-36.
3. Dalmau, J., C. Geis, and F. Graus, *Autoantibodies to Synaptic Receptors and Neuronal Cell Surface Proteins in Autoimmune Diseases of the Central Nervous System*. Physiological reviews, 2017. **97**(2): p. 839-887.
4. Schou, M., et al., *Prevalence of serum anti-neuronal autoantibodies in patients admitted to acute psychiatric care*. Psychological Medicine, 2016. **46**(16): p. 3303-3313.
5. Lennox, B.R., et al., *Prevalence and clinical characteristics of serum neuronal cell surface antibodies in first-episode psychosis: a case-control study*. The Lancet Psychiatry, 2017. **4**(1): p. 42-48.
6. Pollak, T.A., et al., *Autoantibodies to central nervous system neuronal surface antigens: psychiatric symptoms and psychopharmacological implications*. Psychopharmacology, 2016. **233**(9): p. 1605-1621.
7. Dalmau, J., *NMDA receptor encephalitis and other antibody-mediated disorders of the synapse*. Neurology, 2016. **87**(23): p. 2471.
8. Guan, H., *Pictographs of encephalitis in Chinese characters*. The Lancet Neurology, 2019. **18**(4): p. 331.

Appendix II Acknowledgements

Throughout the writing of this dissertation, I have received a great deal of support and assistance from my supervisors, colleagues, friends, and families. Firstly, I would like to thank my supervisor, Prof. Dr. **Pilar Martinez**, whose expertise was invaluable in the formulating of the research topic, the guidelines to conduct most of my experiments and methodology in particular. I still remember the first day when we met, your passionate smile and practical suggestions calmed me so much, especially at that moment I was still shaking about my decision of coming to the Netherlands to start my real research life. During the past five years, you advised me on multiple projects, you discussed with me on all the results step by step, you carefully corrected on each of my manuscripts. I can't imagine how could I complete these tough work without your guidance.

Equally grateful in my heart to my co-supervisor Dr. **Mario Losen**. Your rich knowledge in fundament research really brings every detail to my brain, and your strict and serious work style do keep improving all my manuscripts to the next level! Together with Pilar, both of your help to me is not only shown in my academic study but also in daily life! It is a huge change in my life when I moved from China to the Netherlands. You may not remember that day I shared with Pilar the news about Yumi's born. After joyful congratulations, the first question she asked me was how would I take the baby home. Before my answer, Pilar already said I would call Mario to drive you home. To you it would be only a simple kind offer, but to me, there was an emotion wave at my heart. When my families were still thousands of miles away, you and Pilar are the people who always stand behind me.

The same thanks go to my co-supervisor Dr. **Rob Rhoul**. As you may know, I was a clinical doctor and proud of what I was doing before. Also, my study topic here actually is so close to neurology although we always claim we are working on psychiatry disorders. For these reasons, I always want a supervisor who is working in the hospital, best from neurology department. Then when we have the GAD project and increasingly meetings between us make this possible. You may not notice I was so happy that you can be one of my co-supervisors. Definitely a lot of important and valuable advises from you. Thank you!

I would also like to thank other senior researchers that helped a lot with my main projects in Maastricht. **Peter**, you have checked almost all my manuscripts and gave tremendous suggestions and feedbacks to make them to the next level. Can't say more words for the work! **Jan**, most of the time, you responded to me immediately after I asked or requested something from you. We got many samples directly from your hands and even once you sent them to the lab when I was just at the step of using samples! **Marc**, thanks for sharing your research experiences and your comments on my projects, especially we discussed on why researchers in this field did not use human brain slices directly to do the IgG staining, you told me actually you had tried that before and stated the problem with it. Such valuable information that made me know clearer about which direction should go. **Wim**, thank you for every time you passed by my office, always came in and say hello to me and shared your knowledge about the culture difference between China and Europe, which definitely helped me to adapt myself easier in the new environments. **Bart**, thanks for your welcome comments on my daily work in the

department and also feedbacks directly on the PTSD project. In the last 2 years when I got some results from our cross-sectional studies, I started to realize the importance of longitudinal study and it happened that we had the chance to explore this with the PTSD cohort. Also, the same thanks for **Laurence**'s help during this study, your patient communication and actively attitudes on the collaboration! **Anita**, when I am writing this part, I just received your card about Julia's born, congratulations! Now you are 2 babies' mother, proud of you! So great to have discussions with you on the clinical things, the reality in the current research field and the potential future directions. I really see the ambitions in your talk and hope we can continue these works in the future.

I would like also to acknowledge other senior researchers and friends in Maastricht who helped a lot during my stay in Maastricht. **Ping**, how could I describe my gratitude for your help during my study in Maastricht! You are the one who introduced me to Pilar and made this happen! Together with Qing, we consulted you so many times about work and daily life kinds of stuff. I still remember in the first few weeks when we came here, you helped us to call the energy company to make the contract for our apartment. When Qing was pregnant, you came to help her with the future plan, and when Yumi was born, you and **Kaimei** (mooi) visited us and played with Yumi. You must not notice but I have to say, although most of the time, we mainly discussed daily life or Qing's project when we gathered, still I learned from you the professional attitude, strict and efficient to work! Thanks a lot! **Harry**, thanks for the help during my scholarship application procedure and your care of my daily life in Maastricht. Your work definitely helped a lot of Chinese students who want to get a better study opportunity. You also gave me a lot of chances to communicate with you directly about the university policies on Chinese students. **Sunny**, thanks for the trust to let me participate in the event about startups in Maastricht meeting investors from China. Indeed, you may not know I visited one of the investors in the summer this year who was very interested in my proposal! Also, thanks for providing me other similar opportunities and your comments on my projects! Hope we can communicate more in the future!

I would also like to thank many scientists from outside of Maastricht for their support and trust in my work that directly related to my thesis. **Prof. Penninx** from Amsterdam provided the most directly help in analyzing the results of NSAbs tests in the NESDA cohort. You are the scientist I have met who really has a full view of the field of depression and anxiety and runs the project in such a successful way. **Gerard**, we met several times during the NESDA research days, visited your office and communicated continuously by email! Thanks a lot for your practical suggestions! **Prof. Dalmau** from Barcelona, thanks for the technique helps, direct communication during conferences and daily emails. Your suggestions do make me be surer about where I have reached and where would be the next step. Thanks, **Dr. Titulaer** from Rotterdam and **Dr. Waters** from Oxford for providing us the materials (plasmids, blood samples), the communicating and the feedback on my results as well. These three scientists also shared their materials like plasmids, antibodies or blood samples with us, the details were described in the acknowledgment part of the according chapters. Besides, I would also like to thank for the scientists that provided their help during my study, even some of them were not turned into a publication, including **Prof. Maarten Reith** from New York, **Prof. Randy Blakely** from FAU and **Dr. Amy Eshleman** from OHSU who provided the SERT, DAT and

NET cell line, **Prof. Masaki Fukata** from NIPS, Japan who sent me the plasmids containing GABAAR subunits, **Prof. Yoshinori Moriyama** from Okayama University who sent me the VGLUT antibodies and proteins, **Prof. Salah El Mestikawy** from Sorbonne Université, Paris (UPMC) and **Dr. Etienne Herzog** from Université de Bordeaux who shared the VGLUT plasmids, **Dr. Romana Höftberger** and **Dr. Inga Koneczny** from Medical University of Vienna who shared the anti-NF155 antibodies and positive controls and **Dr. Luis Querol** from Barcelona who shared the NF-155 plasmid with us.

Here I want to thank greatly to my dear colleagues. As mentioned above, **Inga** also worked in Maastricht for a period of time, during which I learned ELISA from you. Your passion for academic impressed me a lot! Thank you also for the daily suggestions and helps. **Abhishek, Ece,** and **Murat**, thank you for the guidance in the lab in my early PhD study, your suggestions helped me a lot to improve my experimental skills and made me know better in this the field we were working with. **Jo**, you were the most experienced PhD when I came here, both in the lab and in life. Impressed me a lot when I heard your things about making ice cream using liquid nitrogen, repairing your phones by yourself, and your collection of all the postcards on the wall. You made me the travel to Aachen for Christmas Market and for the first time trying glue wine and knowing the concept of Limburg carnival. Thank you! The same to **Yara**, jajaja, Jo and you are together now in Munich! For some reason, you are like my big sister although, in real life, I do not have a sister at all in my family, all brothers! **Carolin Hoffman**, no matter how many words should I put here would be just not enough. From my interview, you already started to plan for my stay. Also, the first day I came to the lab, you arranged almost everything for me. What a great introduction tour! I later realized that you had such a good relationship with most of the collages in our department, and actually for this reason, to date I always feel that I am one of the Chinese students who mingled so well among colleagues. Not to mention that you taught me all the lab techniques hand by hand and introduced me to the most influential labs and scientists in this field one by one. If a PhD student can be another PhD student's supervisor, you will definitely on my supervision list. Also, it is hard to not mention in someday of our lab day out, we chatted about the basic concept different between Europe and China and that lovely day you mentioned you wanted to be a mother someday after you PhD and own your farm with your boyfriend **Joao**. And now it already comes true!!! Congrats for that, and the best words here also to **Joaquim and Joao**. I also enjoyed when we visited each other many times to make Chinese food or European food together, which made me understand the culture much better and deeper than I expected. It is a pity that Qing and I were not there for your wedding but for sure hope someday, we, together with Yumi may have the chance to visit you and your families in Portugal.

Marina! What a lovely smart girl! When I, the one fresh off the boat, came to Maastricht, 2 months later, you were here too. Immediately, I felt like that when I enter a class, you were my classmate, while Jo and Carolin were all a bit like teachers. We worked and studied together which made the tense study and training things much easier and more interesting. Also, your Kala Ok and group events that made the life in Maastricht not boring at all. Besides these charming parts in your characters, I also want to say the strict and efficient way you do have when we work together, which leaned me a lot actually. Good luck with your writing and animal experiments! **Simone**, similar to Marina, we all started our PhD studies in the same

year and experienced similar stages in the lab. Because both of us living with families and had a daughter, your suggestions were always very practical and tailored for my situation. Thanks a lot! Now, we almost will defend our dissertation on the same day (in the end, I will be 1-week earlier!), good luck with everything and hope to see you soon in Maastricht! **Caterina, Qian, Daan, Nikita** and **Tanya**, so glad that we worked together in the same group and had the chance to discuss our results, share our opinions and helped each other in the lab! Here I also would like to thank **Koen** as well when so many times we went to the same events and made jokes. Keep on your boxing and Cheers bro! **Artemis**, for many times, small talks with you really made me feel relax and deepen my understanding of Europe cultures. Also, I would like to thank other colleagues, **Roy, Fred, Maarten, Joao, Ehsan, Gusta, Marion, Shannen**, for many times, we went to the same events and shared stories, a lot of pleasant memories build my life in Maastricht! For many other colleagues, **Britt, Marion, Sandra, Sarah, Anne, Wouter, Christian, Roel, Glenn, Pim, Nick, Milaine, Dean, Elentina Sylvana, Alix, Bethany, Perla, Rose, Ellis, Kyonghwan, Gowoon, Majed, Mohammed, Faris, Faisal** and **many** may not mentioned here who belong to our big lunch group, I really enjoyed the talks, greetings and jokes you have made! There were also my students doing their intership here such as **Nils, Nienke**, and **Sofia** who did part of the work and I really enjoyed the time that we worked together. Thanks!

I would also like to thank my colleagues that we worked together as PhD representatives of the MHeNS, including **Nynke, Lotta, Simone, Lotter, Angelique**. The way we arranged things as organizers really deepen my knowledge of how things went on and the true school structure or strategies that I did not reach before. We gathered together many times in the school restaurant, the coffee shops and tried snacks at Nynke's place. A lot of fun! The same thank my colleagues that we worked together as lab day-out committee members including **Govert, Roy, Danny**, and **Thoe**. Really a good chance to work on the same event and let it happen!

I would also like to thank other senior scientists from the department, including **Jos, Yasin, Daniel, Gunter, Ali**, you are the ones who enriched my knowledge of neuroscience. Also, I would thank the support I have received from our tech-team, especially **Hellen, Marjan, Denise, Marcella**, and **Rachelle**. I would thank **Rachelle** and **Ankie** for their help during my study here in Maastricht.

I would also like to thank my best friends **Junfang** and **Jieyi**. We had the chance to spend our 7 years in the same class in China during our medical study and 3 years Maastricht. Now both of you already successfully start your career as medical doctors in one of the most famous hospitals in China. Good job! I always remember the huge help you made to me in my first year here and later the joyful of so many times we visited each other and traveled together. And even at almost the same month, we two families had babies and became parents! **Zhiqi**, what a handsome baby! How wonderful a sence when you and **Yumi** seated in **Kaimei's** swing chair!Thoes lovely day! Cheers for friendship!

I would like also thank my other friends from China, including **Shuo, Yuan** and **Quan, Xiaoqing, Yilin, Qi, Jianqin** and **Guilin, Ning, Shujin, Aomin, Huajie, Ming, Xinying, Tianyu, Lianci, Han, Ying, Shijie, Longping, Wenjie, Wenting, Letong, Yi, Shunxin, Xinwei, Jianhua** ..., I may not write my words separately for each of you but deeply in my

heart, I always feel lucky that we have the chance to study in the same university far away from home. The life in Maastricht is great but sometimes also could be hard, nothing could compare the help from you guys that we do share the same values and backgrounds.

Ma and Pa, Ma in law and Pa in law, my **brothers** and my **sisters** in law, you are the people who always stand behind me and I always know, no matter what happens here, there is a warmly place called home!

And also thank my daughter **Yumi**, you never know how much the motivation I got from you. Actually, you may never know you are always so strong, so determined and always try so hard when you want to get something, which turns out to protect my dream much better than you may ever think.

In the end, I would thank the most my wife Qing. Words will never express enough about what I feel but still, I will try gathering sentences here to describe it. People always say life is like a box of chocolates. When I look back that many amazing things happened during my PhD study, there were 2 things always jumped out immediately, one was that you married me in the summer of 2015 and another was later you gave birth to Yumi in the spring of 2017! You just ignored the uncertainty of life and bravely moved forward to me which really swept away all my hesitation and worries! I would say my PhD study is largely part of your work because, the really truth is, your courage inspires my courage and your determination builds my determination.....Looking forward to tasting the next box of chocolates with you!