Microstructural and metabolic alterations in the zebrafish brain induced by toll-like receptor 2 deficiency: insights from ultra-high field magnetic resonance imaging and spectroscopy

Singer, R.

LIST OF PUBLICATIONS
CURRICULUM VITAE
ACKNOWLEDGEMENTS

FULL PUBLICATIONS

1. **Metabolic Profiling of Suprachiasmatic Nucleus Reveals Multifaceted Effects in an Alzheimer's Disease Mouse Model.** Eeza MNH, **Singer R**, Höfling C, Matysik J, de Groot HJM, Roβner S, Alia A. J Alzheimers Dis. **2021**;81(2):797-808. doi: 10.3233/JAD-201575. PMID: 33843677; PMCID: PMC8203226.

2. **Evaluation of suprachiasmatic nucleus in Alzheimer's disease with non-invasive magnetic resonance methods. Singer R**, Alia A. Neural Regen Res. **2022**;17(8):1753-1754. doi: 10.4103/1673-5374.332136. PMID: 35017432; PMCID: PMC8820719.

3. **Probing microstructural changes in muscles of leptin-deficient zebrafish by non-invasive ex-vivo magnetic resonance microimaging.** Eeza MNH, **Singer R**, Ding Y, He J, Zuberi Z, Baelde HJ, de Groot HJM, Matysik J, Spaink HP, Alia A. **2023** PLOS ONE 18(4): e0284215. Doi: 10.1371/journal.pone.0284215

4. **Ultrahigh field diffusion magnetic resonance imaging uncovers intriguing microstructural changes in the adult zebrafish brain caused by Toll-like receptor 2 genomic deletion. Singer R**, Oganezova I, Hu W, Liu L, Ding Y, de Groot HJM, Spaink HP, Alia A. NMR Biomed. **2024** May 14:e5170. doi: 10.1002/nbm.5170. PMID: 38742727.

5. **Unveiling the Exquisite Microstructural Details in Zebrafish Brain Non-Invasively Using Magnetic Resonance Imaging at 28.2 T. Singer R,** Oganezova I, Hu W, Ding Y, Papaioannou A, de Groot HJM, Spaink HP, Alia A. Molecules **2024**, 29, 4637. https://doi.org/10.3390/molecules29194637

CONFERENCE PROCEEDINGS

1. **Magnetic Resonance Imaging of zebrafish (Danio rerio) at ultra-high magnetic field (1.2 GHz). Singer R**, Krug JR, Hu W, Spaink HP, de Groot HJM, Alia A. Proceedings Euromar (2022)

2. **Micro-imaging of zebrafish (*Danio rerio*) at 1.2 GHz (28.2 Tesla). Singer R**, Prompers JJ. Proceedings Euromar (2022). Presented during the satellite meeting of Euromar (2022)

3. **Diffusion-based MRI at 28.2 T to resolve microstructural details and white matter tracts in young adult zebrafish brain. Singer R**, Hu W, Spaink HP, de Groot HJM, Alia A. Proceedings Euromar (2023)

4. **Exploring the potential of ultrahigh magnetic field (1.2GHz) in Diffusion MRI to resolve white matter tracts in zebrafish brain. Singer R**, Alia A. Presented during the annual Dutch NMR Discussion group (NMR DG) meeting (2024).

5. **Unbeatable super-resolution of voxel-based MR spectroscopy of zebrafish brain achieved at 28.2 T. Singer R**, Hu W, Liu L Spaink HP, de Groot HJM, Alia A. Presented during the MRI parallel session during Euromar (2024)

6. **Diffusion MRI and localised spectroscopy of organoids on 1.2 GHz.** Tax C.M.W., Nikolaeva T, **Singer R**, Jakobs C, Alia A, Velders A, Pasterkamp J, Krug J.R. Proceedings Euromar (2024)

7. **The Precision of Ultrahigh Field MRI: Detecting intriguing Microstructural Changes in the Brain of Leptin-Deficient Zebrafish. Singer R**, Velthuyzen A, Ding Y, de Groot HJM, Spaink HP, Alia A. Proceedings Euromar (2024)

CURRICULUM VITAE

Rico Singer was born on the first of July 1993 in 's-Gravenhave (The Hague / Den Haag) in the Netherlands. He obtained his high school diploma in 2011 at the Baken Park Lyceum. For his bachelor's degree, Rico moved to Delft and studied Chemistry at the Hogeschool Rotterdam (Rotterdam University of Applied Sciences). During his studies, Rico participated in the Analytical Science Talent Program (ASTP), an honors program offered by the Top Institute for Comprehensive Analytical Science and Technology (TI COAST). Additionally, Rico completed internships focusing on analytical chemistry. At Royal Dutch Shell in Rijswijk, concentrating on size exclusion chromatography; at BiosparQ B.V. in Leiden, focusing on matrix-assisted laser desorption/ionization (MALDI) mass spectrometry, and for his bachelor's thesis project at the DSM Biotechnology Center in Delft, focusing on method development for semi-preparative liquid chromatography. In 2017, Rico attended the pre-master's program in Chemistry at the University of Amsterdam (UvA) and the Vrije Universiteit (VU), after which he commenced his Master Analytical sciences at the same institutes. During his master, Rico performed another internship at the DSM Biotechnology Center in Delft, focussing on integrating various analytical techniques to isolate and identify bioactive compounds from microorganisms. This involved semi-preparative liquid chromatography, mass-spectrometry, and various nuclear magnetic resonance methods. Alongside his Bachelor and Master degree, Rico was a member of the "Delftse Studenten Roeivereniging Proteus-Eretes", where he was actively involved in the organisation of various events, including rowing competitions, business days, and field trips. Furthermore, he was significantly involved in the organisation of the lustrum year, which spanned an entire year and encompassed numerous activities. Additionally, he managed self-operating sub-committees responsible for organizing specific events during this year.

After his Master degree, Rico started his PhD project in 2020 at the university of Leiden in the Biophysical Organic Chemistry and solid-state NMR (BPOC SSNMR). During his PhD, Rico was supervised by Dr. A. Alia and Prof. Dr. H.J.M. de Groot. Rico's PhD project focused on optimizing and applying a wide range of magnetic resonance imaging (MRI) and magnetic resonance spectroscopy (MRS) techniques for examining the tiny zebrafish (*Danio rerio*) brain at ultra-high fields. This included utilizing the state-of-the-art 28.2 T MR system. During his PhD, Rico presented his work at several scientific conferences, including Euromar 2022 in Utrecht, the Netherlands; Euromar 2023 in Glasgow, United Kingdom; and Euromar 2024 in Bilbao, Spain. Furthermore, he presented his work at the 2023 Dutch NMR Discussion group meeting in Oss and during the 2024 national ultra-high field NMR (uNMR-NL) annual meeting in Utrecht. Rico participated in scientific courses and summer schools, including the course "Scientific Conduct for PhDs" provided by the graduate school of Leiden University and the International Society of Magnetic Resonance (ISMAR) summer school "Theory of NMR" at Schloss Windischleuba in the field of General NMR (2022) and Relaxation (2023).

ACKNOWLEDGEMENTS

Groundbreaking solitary research efforts are rare. Every scientist, whether they're a graduate student or a tenured professor, needs to acknowledge that their accomplishments would not have been able without the help of others. I am no exception to this rule. Although it is often difficult to find the right words to express my gratitude, I will make every effort to do so.

First and foremost, I would like to express my sincere gratitude to **A. Alia** and **Huub de Groot**, for their guidance and support throughout my PhD project. You consistently challenged me to think critically about my work, while also recognizing and celebrating my progress. Thanks to your mentorship, I have developed numerous skills that will undoubtedly be invaluable as I continue my scientific career.

Of course, I would like to thank my paranymphs, **Titus de Haas** and **Bas Kreupeling**, for their invaluable support in helping me bring my PhD journey to a successful conclusion. Your encouragement, assistance, and presence throughout this journey meant a lot to me.

I would also like to acknowledge the help from the master and bachelor students that were involved in various parts of this work, **Ina Oganezova**, **Alexandra Velthuijzen**, and **Camiel Hildenberg**. It was a pleasure to work with you.

In many ways, the Biophysical Organic Chemistry and Solid-State NMR (BPOC/SSNMR) group and its staff deserve my gratitude. Although I was the sole person involved in MRI, there was no lack in scientific discussions which elevated the quality of my research. These past few years would not have been the same without our lunches, drinks, pizzas, coffees, cakes, buitendagen, baby showers, Christmas parties, and unplanned corridor talks. For this, I would like to thank the remaining part of the BPOC/SSNMR group during my time there: **Francesco**, **Anjali**, **Agur, Dario, Suzanne**, **Dario**, **Lolita**, **Serkan**, **Tirong**, **Matthijs**, **Art**, **Padmaja**, **Xinmeng**, **Willem**, **Jan Paul**, **Laura, Rianne, Anouska**, **Chuang**, **Margareth**, **Willemijn**, **Tjeerd**, **Emiel**, **Rick**, **Jonas**, **Jos**, **Sam**, **Roxane**, **Simon**, **Pien**, and **Aegir**. Of course, there is a single person that deserves special thanks. Thank you, **Liesbeth,** for everything these years. You are truly a key member of the group.

For their endless technical support, I would like to thank **Karthick Sai Sankar Gupta**, **Fons Lefeber**, and **Maria Nardella**. You made my long days spend behind the MRI a whole lot better.

For their many hours spend on helping me handle the zebrafish, I would like to thank **Yi Ding**, **Li Liu**, **Herman Spaink**, and most of all **Wanbin Hu**. Without your help, there would have been very little for me to measure in the MRI.

From the Universität Leipzig, **Narmin Bashirova** and **Muhamed Nour Hashem Eeza** deserve my gratitude for the many personal, scientific and philosophical discussions we shared, as well as for their assistance in the field of MRI and HR-MAS.

The micro MRI at UHF community within the Netherlands is rather small. As a result, the support from members of this community proved invaluable throughout my PhD project. I am especially grateful to **Julia Krug** and **Tatiana Nikolaeva** from Wageningen university & Research for always being available. Additionally, I would like to extend my sincere thanks to **Chantal Tax** from UMC Utrecht for generously sharing her expertise in diffusion MRI, involving me in her projects, and providing me the opportunity to present my work to her group.

My thanks to the ultra-high field Nuclear Magnetic Resonance facility in for the Netherlands (uNMR-NL), supported by the Nederlandse Organisatie voor Wetenschappelijk Onderzoek (NWO), for enabling me to perform countless hours of MRI experiments at various ultra-high field MRI systems all over the Netherlands.

I thank **Volker Lehmann**, **Thomas Oerther**, **Antonios Papaioannou**, **Oliver Gruschke**, and **Sarah Mailhiot** from Bruker BioSpin, who were always available to answer my many questions. My sincere gratitude to the **Swaantje Mondt Fonds** and the **Stichting Praesidium Libertatis I** for their generous financial support during my PhD, which enabled me to undertake a research visit to the Institute for Medical Physics and Biophysics at Universität Leipzig, Germany.

For their many hours spend on making this PhD thesis look as good as it does, I would like to thank **Susanne Ottenheym** and **Ward van Zeijl**. You turned my ideas into reality, and for that I am grateful.

It is easy for one to start feeling isolated during a PhD, especially if it coincides with a global pandemic. There are many people, to many to start listing here, which helped me through these years. Not by providing me with any scientific input, but simply by being there for me when I needed some distraction. Without these dear friends, I would not have made it this far. Thank you for everything.

None of what I accomplished would have been possible without the support of my family; **Richard**, **Anoesjka**, **Axl** , **Sanne** and **Diëgo**. Thank you for keeping me sane and continuously reminding me of the things that are truly important in life.

Above all, **Mart**, you helped me through all these years. You told me that you already know how grateful I am for your tireless support and that there is no need to put those words on paper. Nonetheless, I thank you for everything and I truly believe I wouldn't have come this far without you by my side.

Finally, I dedicate my closing words to **Maria Spaans**, **Oma Riet**, whose absence I continue to feel every day. On the fifth of May 2023, in the middle of my PhD, I had to say goodbye to you. The fact that you are no longer here breaks my heart, but I know you would have been extremely proud of me.