Sensing transport: label-free in vitro assays as an atTRACTive alternative for solute carrier transporter drug discovery

Sijben, H.J.

LIST OF PUBLICATIONS

Part of this thesis

Gorostiola Gonzalez, M.†, **Sijben, H.J.**†, Dall' Acqua, L., Liu, R., IJzerman, A.P., van Westen, G.J.P., Heitman, L.H. (2022) Molecular insights into disease-associated glutamate transporter variants using *in silico* and *in vitro* approaches. (manuscript in preparation)

Bongers, B.J.†, **Sijben, H.J.**†, Hartog, P.B.R., IJzerman, A.P., Heitman, L.H., van Westen, G.J.P. (2022). Proteochemometric modelling identifies novel norepinephrine transporter inhibitors. *Journal of Chemical Information and Modelling* (manuscript submitted)

Sijben, H.J., Dall' Acqua, L., Liu, R., Jarret, A., Christodoulaki, E., Onstein, S., Wolf, G., Verburgt, S.J., Le Dévédec, S.E., Wiedmer, T., Superti-Furga, G., IJzerman, A.P., Heitman, L.H. (2022). Impedance-based phenotypic readout of transport function: a case for glutamate transporters. *Frontiers in Pharmacology*, **13**, 872335

Sijben, H.J., Superti-Furga, G., IJzerman, A.P., Heitman, L.H. (2022). Targeting solute carriers to modulate receptor–ligand interactions. *Trends in Pharmacological Sciences*, **43**(5), 358–361

Sijben, H.J., van Oostveen, W.M., Hartog, P.B.R., Stucchi, L., Rossignoli, A., Maresca, G., Scarabottolo, L., IJzerman, A.P., Heitman, L.H. (2021). Label-free high-throughput screening assay for the identification of norepinephrine transporter (NET/SLC6A2) inhibitors. *Scientific Reports*, **11**, 12290

Sijben, H.J., van den Berg, J.J.E., Broekhuis, J.D., IJzerman, A.P., Heitman, L.H. (2021). A study of the dopamine transporter using the TRACT assay, a novel in vitro tool for solute carrier drug discovery. *Scientific Reports*, **11**, 1312.

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Other publications

Mocking, T.A.M., **Sijben, H.J.**, Vermeulen, Y.W., IJzerman, A.P., Heitman, L.H. (2022). MPP+-induced changes in cellular impedance as a measure for organic cation transporter (SLC22A1-3) activity and inhibition. *International Journal of Molecular Sciences*, **23**(3), 1203

Dvorak, V., Wiedmer, T., Ingles-Prieto, A., Altermatt, P., Batoulis, H., Bärenz, F., Bender, E., Digles, D., Dürrenberger, F., Heitman, L.H., IJzerman, A.P., Kell, D.B., Kickinger, S., Körzö, D., Leippe, P., Licher, T., Manolova, V., Rizzetto, R., Sassone, F., Scarabottolo, L., Schlessinger, A., Schneider, V., **Sijben, H.J.**, Steck, A., Sundström, H., Tremolada, S., Wilhelm, M., Wright Muelas, M., Zindel, D., Steppan, C.M., Superti-Furga, G. (2021) An overview of cell-based assay platforms for the solute carriers family of transporters. *Frontiers in Pharmacology*, **12**, 722889

Superti-Furga, G., Lackner, D., Wiedmer, T., Ingles-Prieto, A., Barbosa, B., Girardi, E., Goldmann, U., Gürtl, B., Klavins, K., Klimek, C., Lindinger, S., Liñeiro-Retes, E., Müller, A.C., Onstein, S., Redinger, G., Reil, D., Sedlyarov, V., Wolf, G., Crawford, M., Everley, R., Hepworth, D., Liu, S., Noell, S., Piotrowski, M., Stanton, R., Zhang, H., Corallino, S., Faedo, A., Insidioso, M., Maresca, G., Redaelli, L., Sassone, F., Scarabottolo, L., Stucchi, M., Tarroni, P., Tremolada, S., Batoulis, H., Becker, A., Bender, E., Chang, Y., Ehrmann, A., Müller-Fahrnow, A., Pütter, V., Zinder, D., Hamilton, B., Lenter, M., Santacruz, D., Viollet, C., Whitehurst, C., Johnsson, K., Leippe, P., Baumgarten, B., Chang, L., Ibig, Y., Pfeifer, M., Reinhardt, J., Schönbett, J., Selzer, P., Seuwen, K., Bettembourg, C., Biton, B., Czech, J., de Foucauld, H., Didier, M., Licher, T., Mikol, V., Pommereau, A., Puech, F., Yaligara, V., Edwards, A., Bongers, B.J., Heitman, L.H., IJzerman, A.P., **Sijben, H.J.**, van Westen, G.J.P., Grixti, J., Kell, D.B., Mughal, F., Swainston, N., Wright-Muelas, M., Bohstedt, T., Burgess-Brown, N., Carpenter, L., Dürr, K., Hansen, J., Scacioc, A., Banci, G., Colas, C., Digles, D., Ecker, G., Füzi, B., Gamsjäger, V., Grandits, M., Martini, R., Troger, F., Altermatt, P., Doucerain, C., Dürrenberger, F., Manolova, V., Steck, A., Sundström, H., Wilhelm, M., Steppan, C. M. (2020). The RESOLUTE consortium: unlocking SLC transporters for drug discovery. *Nature Reviews Drug Discovery*, **19**(7), 429–430.

Martella, A., **Sijben, H.**, Rufer, A.C., Grether, U., Fingerle, J., Ullmer, C., Hartung, T., IJzerman, A.P., van der Stelt, M., Heitman, L.H. (2017). A novel selective inverse agonist of the CB2 receptor as a radiolabeled tool compound for kinetic binding studies. *Molecular Pharmacology*, **92**(4), 389–400.

Poster and oral communications

3-5-2018	LACDR Spring Symposium (*poster*)
16-2-2019	LACDR Spring Symposium (*poster*)
5-8-2019	BioParadigms BioMedical Transporters Conference 2019 (Lucerne, Switzerland) (*poster*)
23-9-2019	FIGON Dutch Medicines Days (*poster*) **Awarded with the Poster Competition Prize**
2-7-2020	LACDR Spring Symposium (*poster*)
31-5-2021	LACDR Spring Symposium (*oral*)
27-9-2021	FIGON Dutch Medicines Days (*poster*)
7-10-2021	6th RESOLUTE Consortium Meeting (Basel, Switzerland) (*oral*)
14-4-2022	LACDR Spring Symposium (*poster*) **Awarded with the Poster Competition Prize**

CURRICULUM VITAE

Huub Sijben was born in Woerden, The Netherlands, on August 8th 1993. He graduated from pre-university education at the Katholieke Scholengemeenschap Etten-Leur in 2011. That same year, he started the Bachelor's program Bio-Pharmaceutical Sciences at Leiden University. In his second year, he became a board member of the study association L.P.S.V. „Aesculapius" for one year, for which he organized a study trip to Vienna and Budapest amongst other activities. At the end of his Bachelor's program, Huub performed an internship at the division of Medicinal Chemistry of the Leiden Academic Centre for Drug Research (LACDR) under the supervision of dr. Maarten Doornbos and prof. dr. Laura Heitman. This project focused on the effect of ions on activation of the human metabotropic glutamate receptor 2 (mGluR2).

In September 2015, Huub started the Master's program Bio-Pharmaceutical Sciences at Leiden University with an emphasis on molecular pharmacology. He performed a nine-month internship at the division of Drug Discovery and Safety (formerly Medicinal Chemistry) of the LACDR under the supervision of dr. Andrea Martella and prof. dr. Laura Heitman. This project was in collaboration with Hoffmann-La Roche and focused on the characterization of a novel radiolabeled inverse agonist for the human cannabinoid receptor 2, which resulted in a co-authorship on a publication in Molecular Pharmacology. After finishing this project, Huub wrote a literature review on the use of nanoparticles in allergy vaccines under the supervision of dr. Romain Leboux. For his second research project, he performed a six-month internship at the biopharmaceutical company Heptares (currently Sosei Heptares) in Welwyn Garden City, United Kingdom, under the supervision of dr. Elena Segala. This project was in collaboration with prof. dr. Ad IJzerman from the division of Drug Discovery and Safety of the LACDR and focused on the ligand binding mechanisms of partial and inverse agonists of the adenosine A2A receptor using X-ray crystallography. Huub successfully obtained his Master's degree with a cum laude distinction in 2017.

After his studies, Huub coordinated a second-year Bachelor's practical course on Cellular Biochemistry at Leiden University. He then pursued his interest in molecular pharmacology and started as a PhD candidate at the division of Drug Discovery and Safety of the LACDR in 2018 under the supervision of prof. dr. Laura Heitman and prof. dr. Ad IJzerman. The project was part of the European

RESOLUTE consortium, which is funded by the Innovative Medicines Initiative (IMI), European Union and major pharmaceutical companies (EFPIA). The topic of his research was the development of cell-based label-free assays for solute carrier transporters, with the aim to use such assays in drug discovery. During his PhD, Huub frequently joined consortium meetings and work package videoconferences, where he presented his work via oral and poster presentations. Moreover, he presented posters at national and international conferences, being awarded with a poster prize at the FIGON Dutch Medicines Days 2019 in Leiden. Huub initiated several collaborations with RESOLUTE partners that resulted in two joint publications, both of which are included in this thesis.

Huub continued his career as a Research Scientist at Artica Therapeutics, taking on new challenges in drug discovery and development.

ACKNOWLEDGEMENTS

Prior to the start of my PhD trajectory, I looked up to the immense and daring task of compiling and writing this thesis. While four years may seem a long time at first, the pace at which time flies by during this academic journey can only be comprehended by living the experience firsthand. This might seem frightening if you are in the understanding that you are at it on your own. Although I am indeed the single person whose name is on the title page of this book, luckily I have never been alone during this four year adventure (which half of the time coincided with the COVID-19 pandemic). In fact, I have had the pleasure to work with the most experienced, fun, social and helpful people I could have ever wished for. I have reserved this section to express my gratitude towards those who have significantly contributed to the scientific content, substantially increased my motivation and considerably decreased my worries.

First and foremost, I owe my PhD position at the DDS division as well as most of my knowledge on the topics of this thesis to my promotors Ad IJzerman and Laura Heitman. It is under their supervision that I gained the confidence and perseverance to set up several interesting projects and bring them to completion. Their approach to conduct research by creating a socially open and kind environment has been a source of inspiration – one that certainly keeps me motivated for the years to come.

Moreover, I am indebted to the partners of the RESOLUTE consortium, which has been an integral part of my PhD. Without the many (virtual) work package meetings and near endless supply of cell lines and reagents my projects would not have advanced at the rate that they have. Thanks to Giulio, Daniel, Alvaro, Tabea and all the others for keeping the SLC spirit afloat, even over great distances and during lockdowns. Also, a big thanks to the CeMM and Axxam teams, which have greatly contributed to several chapters of this thesis.

Although an interesting topic is important to keep a PhD going, having a nice work environment is even more essential to get things done. Therefore, I want to thank all of my colleagues who I have had the pleasure to work with over the past years. Special thanks to Anna – who taught me how to study transporters using xCELLigence – Cas, Chenlin, Jara, Jeremy, Henk, Lisa, Maarten, Natalia, Rongfang, Tamara, Xue, Xuesong and Yao for being an amazing Bio Group.

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