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## **Chitin in the fungal cell wall: Towards valorization of spent biomass of *Aspergillus niger***

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## Curriculum Vitae

Tim Marijn van Leeuwe was born on the 2nd of February, 1990 in Haarlem, The Netherlands. After finishing high school with the minimum required grades in 2008, Tim took a break from any form of education to figure out what to do next. In 2009, after failing the tests to become an F-16 fighter pilot, he enrolled for the BSc Industrial Design at Delft University only to figure out that this was not his calling. Spurred-on by his unwillingness to work yet another year as a full-time HEMA employee, he enrolled for the bachelor program of Biology at Leiden university after some convincing words of Tonny Regensburg. Despite the initial interest in ecology and evolution, Tim soon developed his interest in molecular biology and microbiology. After studying abroad in Sweden during the final year of his bachelor degree, Tim enjoyed performing microbial research in the group of Dr. Bert van der Zaal under Dr. Martijn Rolloos' supervision. Following graduation in 2012, Tim assisted Tonny Regensburg during lab practicals for first year BSc Biology students at Leiden University. Next, a job at a probiotics company kept Tim busy for the next months before deciding to enroll for the MSc program Microbial Biotechnology and Health at Leiden University in 2013. During his first research internship, Tim combined his interest of evolution with microbiology in the group of Dr. Daniel Rozen. Tim's second internship was at the University of California, Berkeley in the group of Prof. N. Louise Glass, under supervision of Dr. Trevor Starr. This is where Tim discovered his fascination for filamentous fungi. It developed his determination and passion for research and he graduated *cum laude* in 2015. Six months later, at the start of 2016, Tim was offered a position as PhD student by Dr. Arthur Ram to work on the cell walls of the filamentous fungus *Aspergillus niger*. The work performed as a PhD student is described in this Thesis. After four successful publications during his contractual time and after having enjoyed the (research) environment during his PhD, Tim decided not to pursue an academic career. Instead, he has decided to explore other talents and skills in a different environment. As such, Tim is now enrolled in the military program to become a Fighter Controller for the Royal Netherlands Airforce.

## Publications

**van Leeuwe, T.M.**, Wattjes, J., Niehues, A., Forn-Cuni, G., Arentshorst, M., Geoffrion, N., Melida, H., Molina, A., Tsang, A., Meijer, A.H., Moerschbacher, B.M., Punt, P.J., Ram A.F.J., A seven-membered cell wall related transglycosylase gene family in *Aspergillus niger* is relevant for cell wall integrity in cell wall mutants with reduced  $\alpha$ -glucan or galactomannan. *The Cell Surface* (2020). *In press*. doi: 10.1016/j.tcs.2020.100039

**van Leeuwe, T.M.**, Arentshorst, M., Punt, P.J., Ram A.F.J., Interrogation of the cell wall integrity pathway in *Aspergillus niger* identifies a putative negative regulator in transcription involved in chitin deposition. (2020) *Gene X* 5, 100028. doi: 10.1016/j.gene.2020.100028

**van Leeuwe, T.M.**, Gerritsen A., Arentshorst, M., Punt, P.J., Ram A.F.J., Rab GDP-dissociation inhibitor *gdiA* is an essential gene required for chitin deposition in *Aspergillus niger*. (2020) *Fungal Genet. Biol.* 136, 103319. doi:10.1016/j.fgb.2019.103319

**van Leeuwe, T.M.**, Arentshorst, M., Ernst, T. Alazi, E., Punt, P.J., Ram, A.F.J., Efficient marker free CRISPR/Cas9 genome editing for functional analysis of gene families in filamentous fungi. *Fungal Biol Biotechnol* 6, 13 (2019) doi: 10.1186/s40694-019-0076-7

Westhoff S\*, **van Leeuwe T.M.\***, Qachach O, Zhang Z, van Wezel GP, Rozen DE. The evolution of no-cost resistance at sub-MIC concentrations of streptomycin in *Streptomyces coelicolor*. *ISME J.* 2017;11(5):1168–1178. doi: 10.1038/ismej.2016.194

\*Authors contributed equally

# Acknowledgements

It is hard to believe that my time is up as a PhD student. Those four years sounded like an eternity back when I started in early 2016. Just like my research skills, my persona has developed quite a bit over the past years. For the better I must say. I've seen two relationships end, I have bought a house and sold it again within the same year, I have moved six times and in the mean time I have managed to perform some actual research, published four papers and made some amazing friendships that I still cherish to this day. I usually don't do this, but first of all I would like to thank myself for staying sane and managing life while under pressure of finishing my PhD track. In extension of that, I would like to thank the people that were there for me during these trying times, whom have made it possible for me to focus on the work that has been put into this thesis. Mom and dad, without you I would not be here and if it weren't for your support—be it emotionally, financially or temporary housing—I think this journey would have been a lot more stressful. I do not think I can thank you enough for all that you have done and continue to do. I am grateful.

Next, I want to thank my friends outside of Leiden. I have known you for a long time; Jelmer, Ivar, Ale, Wouter, Peter, Nick, Tije and Bub, you have been my home base for the longest time and you have been there for me when I needed it. Ale, I want to thank you for opening up your home during personal struggles. Thanks a lot. Guys, despite the fact that you (most likely) have no clue whatsoever about the work that I have described in this thesis, your friendship has made it possible for me to enjoy life outside of my work. Someone who might have a clue about this work is Ser. Ser, it has always been fun to talk to you about science. I am happy that we have been sharing that passion for the past 10 years. Even though we went our separate paths, I have really enjoyed reciprocal visits to our respective hometowns, often enjoying some fried food and blissful beverages. I am happy to know that you may already call yourself a doctor. Congrats!

To the friends and fellow PhD students that have made the final years of my PhD a lot of fun, I want to thank you. Doris and Anne, I am so happy that we hit it off so well and it's great that we continue to see each other even though I moved to Utrecht. I do miss seeing each other on an almost daily basis to talk about science, PhD struggles, our personal lives or how sore we were from working out with Elvis. Doris, I want to thank you in particular for introducing me to Tjana. Even though this resulted in me moving away from Leiden, know that we are very happy together. Mandy, I have really enjoyed the winter sport holidays of the past two years that we organized together for our research group. Tjana, Eveline, Sander, Bob, Eelke, Isa, Noortje, Renée, Alex, Thomas, I want to thank you all for making sure it was such a good vacation.

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Lastly, I want to thank the people that have worked close with me. First of all, Arthur. I want to thank you for asking me to fill this PhD position. Even though I had just started another PhD position, this felt like the right calling and I am very happy to have switched to your group. You have been the best mentor I could have asked for. At the start of my PhD I experienced both guidance and freedom within the FunChi project's set goals. This gave me a great amount of independence while not getting lost. I also appreciate you challenging my ideas from time to time in order to keep me grounded. Peter, it has been great to be working with you. With less frequent interaction than I have had with Arthur, you have always been very helpful during discussions, coming up with new ideas or literature and in the later stages of the PhD, you were very helpful (and incredibly speedy) in revision of the manuscripts. Importantly, you have helped learn to cut my losses and make some tough decisions, project-wise, in order to move forward. Arthur and Peter, I will never forget your kindness, enthusiasm, intellect and supervision. Thank you so much.

Finally, students, staff, people from all across the IBL, and to all other people who I have not mentioned personally, know that I am grateful for the time we worked together or just for the simple chats. I wish you all the best.

Tim M. van Leeuwe