Visceral leishmaniasis – malaria co-infections

van den Bogaart, E.

Citation for published version (APA):
Authors and Affiliations

Samira H. Abdelrahman
Department of Medicine, Gedarif Teaching Hospital, Gedarif, Sudan

Emily R. Adams
Department of Biomedical Research, Royal Tropical Institute, Amsterdam, the Netherlands
*Present address:* Department of Parasitology, Liverpool School of Tropical Medicine, Liverpool, United Kingdom

Hashim B. M. Ahmed
Department of Medicine, University of Gedarif, Gedarif, Sudan

Payal P. S. Balraadjsing
Department of Biomedical Research, Royal Tropical Institute, Amsterdam, the Netherlands
*Present address:* Center for Infection and Immunology, Academic Medical Center, University of Amsterdam, Amsterdam, the Netherlands

Marieke M. Z. Berkhout
Department of Biomedical Research, Royal Tropical Institute, Amsterdam, the Netherlands

Helena M. de Bes
Department of Biomedical Research, Royal Tropical Institute, Amsterdam, the Netherlands
*Present address:* Crucell Holland B.V., Leiden, the Netherlands

Francois Chappuis
Médecins Sans Frontières/Geneva University Hospitals, Geneva, Switzerland

Irma van Die
Department of Molecular Cell Biology, VU University Medical Centre, Amsterdam, the Netherlands

Jean-Claude Dujardin
Department of Biomedical Sciences, Institute of Tropical Medicine & University of Antwerp, Antwerp, Belgium

Paul England
IOTA Pharmaceuticals Ltd, Cambridge, UK

Dorien Faber
Department of Biomedical Research, Royal Tropical Institute, Amsterdam, the Netherlands
*Present address:* Solidaridad Network, León, Nicaragua

Martin P. Grobusch
Center of Tropical Medicine and Travel Medicine, Academic Medical Center, University of Amsterdam, Amsterdam, the Netherlands

Dawson B. Mbulamberi
Ministry of Health, Kampala, Uganda

Pètra F. Mens
Department of Biomedical Research, Royal Tropical Institute, Amsterdam, the Netherlands
*Present address:* Department of Medical Microbiology & Center of Tropical Medicine and Travel Medicine, Academic Medical Center, University of Amsterdam, Amsterdam, the Netherlands
Ayman B.Y.M. Nour
Blue Nile National Institute for Communicable Diseases, University of Gezira, Wad Medani, Sudan

Bakri Y M Nour
Blue Nile National Institute for Communicable Diseases, University of Gezira, Wad Medani & Department of Medicine, Gedarif Teaching Hospital, Gedarif, Sudan

Kristina M. Orrling
Department of Chemistry and Pharmaceutical Sciences, VU University Medical Centre, Amsterdam, the Netherlands
Present address: Lygature, Leiden, the Netherlands

Koert Ritmeijer
Public Health Department, Médecins Sans Frontières, Amsterdam, the Netherlands

Henk D. F. H. Schallig
Department of Biomedical Research, Royal Tropical Institute, Amsterdam, the Netherlands
Present address: Department of Medical Microbiology, Academic Medical Center, University of Amsterdam, Amsterdam, the Netherlands

Gerard J. Schoone
Department of Biomedical Research, Royal Tropical Institute, Amsterdam, the Netherlands

Elizabeth Sentongo
Department of Medical Microbiology, Makerere University College of Health Sciences, Kampala, Uganda

Masja Straetemans
Department of Biomedical Research, Royal Tropical Institute, Amsterdam, the Netherlands
Present address: Department of Health, Royal Tropical Institute, Amsterdam, the Netherlands

Shyam Sundar
Department of Medicine, Institute of Medical Sciences, Varanasi, India

Al-Badawi A. Talha
Faculty of Medical Laboratory Sciences, University of Gezira, Wad Medani, Sudan
Acknowledgments

This thesis reports part of the research I have been conducting at KIT Biomedical Research in Amsterdam. Based on what was initially conceived as the subject of a short research fellowship, this research is the result of intensive efforts aimed at exploring the complexity of the VL-malaria co-infection from multiple perspectives and translating it into a coherent set of studies. Many people have taken part to these efforts, some formally, many others in a less official way. To all of them I owe my most heartfelt gratitude. James Allen once said: ‘No duty is more urgent than that of returning thanks’. I believe nothing could be more true, and I will try to fulfill my duty to the best I can.

First and foremost, I wish to thank my promotor, Martin Grobusch, for providing me with the opportunity to pursue my PhD degree at the University of Amsterdam. Martin, from the moment you accepted to supervise me during my PhD study, you have been for me an example to draw inspiration from. With your enormous positivity, your kindness and willingness to help and your exemplary efficiency — I still wonder how you do it —, you have guided me wisely throughout all these years, advising me and encouraging me whenever the end appeared as unreachable. I am very grateful to you for all this.

My enormous gratitude also goes to my co-promotor, Henk Schallig, for encouraging my research in the field of co-infections and for allowing me to grow as a research scientist. Henk, eight years ago you have welcomed the request of a newly graduate from Italy asking to join your group as a visiting scholar. You have introduced her to the fascinating field of leishmaniasis and guided her all the way to this thesis. It may have not always been easy (also due to the shortage of funding), and surely not as short as we hoped, but in the end it was certainly worth the effort. Thanks for giving me this incredible opportunity, providing me the tools and wisdom to embark on my research endeavors and supporting me during this entire journey.

I also would like to thank the committee members. I am honored that you have accepted to be part of my defense committee and dedicated your time and expertise to assess my thesis.

A special thanks goes to all the parasitologists that had passed through KIT Biomedical Research and who I had the good fortune to work with. Starting from Emily Adams and Pètra Mens, who have supported me as (unofficial) co-promotors. Emily, with your enthusiasm, friendliness, and deep knowledge of kinetoplastids, you have valuably contributed to shaping this project into its final fit. Thanks for your advice, your support (scientific, moral as well as practical) and the long and interesting chats. Thanks Pètra for discussing and updating the progress of my work on a regular basis, guiding me in the preparation of the field studies and assisting me with all administrative procedures. With your pragmatism and supervision, we have succeeded in performing two interesting field studies with limited (financial) resources. My most heartfelt gratitude goes to Laura de Bes, my right-hand person in the lab during my entire PhD! Laura, I will never forget the amazing time we had working together in the lab, day and night, week and weekends, culturing parasites, isolating Dss and FACSing!!! You had
always been there for me, sharing my successes and my failures, and supporting me until the very end of this long way. I will be eternally grateful to you for all this. Special thanks goes to Gerard Schoone, for his enormous support and his outstanding patience in counting the *Leishmania* parasites at the microscope (*I would still be counting if it wasn’t for you!*). Thanks for all your help, the nice chats, your creativity in the lab and outside the lab and your good sensor of humor. Thanks also to the rest of the Parasitology group, in particular Inge Versteeg and my ex-roommate Eline Kattenberg, for your valuable suggestions and the nice time we had together.

At KIT-BR I would like to acknowledge all my (unfortunately ex)-colleagues, for their incredible support, their expert advice and their words of encouragement. Thanks for the great time we have spent together during coffee breaks, borrels, presentations, etc. and for sharing together the good and recently, less good, moments. I wish you all the luck for a bright future to come. A special acknowledgement goes to Paul Klatser for giving me the opportunity to work at KIT-BR and to the Rapid Diagnostics group for making my last PhD years such a pleasant experience. Thanks to Henk Smits for granting me time officially (and unofficially) to finish writing my thesis and to Rob Pastoor for adjusting my Sudan map. A special thanks also goes to Theresia Abdoel for her support, advise and close friendship during all these years. Thanks Trees for your invaluable help in and outside the lab, your critical approach that has often been so fulfilling and of course, for all the afternoon coffees at the AMC. Thanks to Masja Straetemans for her guidance during data analysis and for her kindness and willing to help with all my statistical questions and to Ente Rood, my roommate, for the nice chats, the expert advice on statistics and for preparing a totally new map on the co-infection distribution! Thanks to Sandra Menting for giving me useful tips on molecular biology, showing interest in my PhD progress and urging me to wrap up this thesis by ‘extorting’ me free cappuccinos (*which I probably still owe her a dozen or so*). Thanks to Marga Goris for sharing the difficulties and frustration that come along while doing your PhD study and for advising me on the last steps of it. A big thank you to Sylvia Talahatu for her efficiency in all the administration issues and for helping me constantly (*also when the end-of-year deadline for making orders was long passed*) and to all the management and technical staff.

A special recognition goes to my students, Payal Balraadisjng, Marieke Berkhout, Dorien Faber and Aurélien Saliki for their valuable contributions to this work and for the enthusiasm they shared on this research topic. It was great fun and pleasure working together with you. You have proven to me that teaching can be an amazing learning experience; I will always be grateful to you for this.

Some of the chapters in this thesis would not have been possible without the support of *Médecines sans Frontières*, in particular of François Chappuis and Koert Ritmeijer, who made available to us the extensive datasets that they have gathered in Uganda and Sudan. François, Koert, it has been an honor and a great pleasure to work and write with you. Thanks to Dawson Mbulamberi (*Ministry of Health in Uganda*), for providing support to part of this research following my persistent requests and to Elizabeth Sentongo (*Makerere University*) for her expert advice on the epidemiology of parasitic diseases in Uganda.
My heartfelt acknowledgements go to the team in Sudan, namely Bakri Nour and Ayman Nour from the Blue Nile National Institute for Communicable Diseases, Al-Badawi Tahla from the University of Gezira, Samira Abdelrahman from the Gedarif Teaching Hospital and Hashim Ahmed from the University of Gedarif, for organizing, coordinating and performing the field work, in collaboration with the Tabarakallah Hospital staff. Bakri, your enthusiasm for our work and your great support have made possible our studies in Sudan, contributing to an important piece of this thesis. Thanks to Al-Badawi for his practical and logistic contribution and to all patients at Tabarakallah Hospital for participating to our study.

A special thanks goes to Ti Pharma for providing financial support to part of this thesis, through the project T4-302 ‘Phosphodiesterase inhibitors for neglected tropical diseases’ and to all the staff involved in it. In particular, I am very grateful to Paul England for his expert advice on compound screening and for having patiently taught me how to analyze a drug response curve properly.

Thanks to Kristina Orrling for providing the compounds used for assay validation, and to the rest of the PDE team, Rob, Chimed, Geert-Jan, David, Thomas, Andrew and co-workers for the pleasant collaboration, great progress meetings and fruitful discussions.

At the VU University in Amsterdam, I would like to thank Irma van Die for her support and guidance during this study, particularly with respect to the immunological part. Thanks Irma for your time and patience in helping me interpreting the data, your constructive responses to my e-mails and for welcoming in your lab. Thanks also to Noortje and Loes for teaching me some basic immunological techniques.

I would like to acknowledge Asrat Hailu (Addis Ababa University), Zewdu Hurissa (University of Gondar) and Sayda El-Safi (University of Khartoum) for their initial contribution in compiling the explorative questionnaires.

Thanks to Jean-Claude Dujardin (Institute of Tropical Medicine) and to Shyam Sundar (Banaras Hindu University) for providing this research with a critical reagent, such as a recently isolated Leishmania strain.

I cannot forget to express my appreciation to my MSc thesis mentors, Donatella Taramelli and Nicoletta Basilico (University of Milan), who first suggested the idea of Leishmania-Plasmodium co-infection on which this thesis was subsequently built on and who provided me with a generous amount of β-hematin. Thanks Prof, thanks Nico for opening me the doors to such an interesting field; I could not have wished a better subject for my PhD thesis!
Addendum

I am thankful to Wendy van der Meide (U-CyTech) for teaching me how to perform ELISPOT assays and for helping promptly with all my last-minute questions, without any self-interest. Thanks Wendy! My appreciation also goes to Berend Hooibrink (AMC), for introducing me to and advising me on all FACS-related issues and to Tassili Weehuizen for the very nice chats in the BSL-3 lab and for helping me with the mouse IFN-γ. I greatly appreciate it, Tassili.

A special recognition goes to Elena Pinelli and Carmen Aranzamendi (RIVM), for their enthusiasm in collaborating with us on this project and for introducing me to the world of dendritic cells. Unfortunately, the lack of funding prevented things to work out as we hoped – I am deeply sorry for this missed opportunity of collaboration – but I highly appreciate the time and input you gave me on this research.

Thanks to all my family and friends for genuinely showing interest and support to my work. In particular, a big, big thank you to my parents for their contribution to this thesis (yes, they really did!), their support and encouragement during this exciting, but at times also difficult journey and for teaching me how to strive for perfection in everything I do. Thanks papà for drawing all but one the maps that are published in this thesis without getting any official acknowledgement, and thanks mamma for carefully checking all the print proofs of my articles and of this thesis, too. This thesis owes you very much and I want to dedicate to you. A special thanks to my sister for addressing my clinical questions, performing last-minute literature reviews (while on holiday!), puzzling over my lab results in the search of a plausible explanation/theory and sharing my frustration and enthusiasm. Thanks Lory! I look forward to the day we will work together on an interesting bug and have a publication signed by the van den Bogaart sisters!

Dear Issa, my right-hand man in life, words cannot express my enormous gratitude and appreciation for the endless support and encouragement you have granted me during these years. Thanks for your love and for your trust, for your patience while waiting me after long days and weekends spent at work, for sharing my ups but also many downs and for encouraging me to reach the end of this long journey.

Dear Rayan, my curly prince, pride and joy of my life, you have been and always will be a true inspiration in my life. I dedicate this thesis also to you and Issa, as a recognition for the time and attention I have taken from you to finish off this work. From now on, I promise you, no more excuses for not enjoying an outing every weekend!

Thank you | Bedankt | Grazie!

Amsterdam, September 2016

Erika
**PhD Portfolio**

---

**Poster and Presentations**

*Oral presentation: ‘Epidemiology of visceral leishmaniasis-malaria co-infections in East Africa’.*


*Oral presentation: ‘Malaria-visceral leishmaniasis co-infections in East Africa’.*


*International Health Dutch Society for Parasitology (NVP) Spring Meeting, Utrecht, the Netherlands, 22 May 2014. 8th European Congress on Tropical Medicine, Copenhagen, Denmark, 10-13 Sep 2013.*

*Oral presentation: ‘Simple colorimetric assay for high-throughput screening of drugs against Leishmania intracellular amastigotes’.*


*Poster presentation: ‘Design, synthesis and pharmacological evaluation of potent inhibitors of trypanosomal phosphodiesterase B1’.*


*Poster presentation: ‘A novel colorimetric assay for screening anti-leishmanial drugs’.*

**van den Bogaart E**, Faber D, Schoone G, Schallig H, Adams E.


*Oral presentation: ‘Co-infections with visceral leishmaniasis and malaria: clinico-epidemiological setting and biological implications’.*

**van den Bogaart E**.

*TropenCentrum, Academic Medical Centrum (AMC), Amsterdam, the Netherlands, 25 Mar 2012.*
Courses

Laboratory Animal Science *(article 9)*, *University of Utrecht, the Netherlands*, 27 Jun – 8 Jul 2016.

Short Course on Teaching of Teachers, *KIT, Amsterdam, the Netherlands*, 8 & 10 Apr 2014.


Developing a Cochrane Diagnostic Test Accuracy Review, *Cochrane Collaboration, Amsterdam, the Netherlands*, 2010.

Teaching

Rapid Diagnostics, practical, Summer Course to Students from Jazan University (Saudi Arabia), *KIT, Amsterdam, the Netherlands*, 2015.

Supervision of MSc. students’ scientific internship (PB, MB, DF, AS), *KIT, Amsterdam, the Netherlands*, 2011–2012.

(Inter)national Conferences, Symposia and Workshops

Panel debate on the theme ‘From Innovation to Impact’, *KIT, Amsterdam, the Netherlands*, 26 Jan 2016.


Mini-symposium ‘The nature of the bug: do certain *Mycobacterium tuberculosis* genotypes have an intrinsic ability to cause multidrug-resistant clusters?’, *KIT, Amsterdam, the Netherlands*, 2014.


50th Spring Meeting of the British Society for Parasitology, Glasgow, United Kingdom, 2 – 5 Apr 2012.

TiPharma Spring Meeting, Utrecht, the Netherlands, 17 Apr 2012.

Mini-symposium on Human African Trypanosomiasis, KIT, Amsterdam, the Netherlands, 10 Dec 2012.

Phagocytosis of hemozoin by RAW 264.7 cells but not THP-1 cells promotes intracellular survival of *Leishmania donovani* amastigotes with a nitric oxide-independent mechanism.

**van den Bogaart E**, Mens PF, Adams ER, Grobusch MP, Schallig HD.
*Parasitol Int*, in press.

**In vitro** evaluation of traditionally used Surinamese medicinal plants for their potential anti-leishmanial efficacy.

Mans DR, Beerens T, Soekhoe RC, Schoone GJ, Oedairadjsingh K, Hasrat JA, **van den Bogaart E**, Schallig HD.

*Leishmania donovani* infection drives the priming of human monocyte-derived dendritic cells during *Plasmodium falciparum* co-infections.

**van den Bogaart E**, de Bes HM, Balraadjsing PP, Mens PF, Adams ER, Grobusch MP, van Die I, Schallig HD.

Cytokine profiles amongst Sudanese patients with visceral leishmaniasis and malaria co-infections.

**van den Bogaart E**, Talha AB, Straetemans M, Mens PF, Adams ER, Grobusch MP, Nour BY, Schallig HD.

Simple colorimetric trypanothione reductase-based assay for high-throughput screening of drugs against *Leishmania* intracellular amastigotes.


Duplex quantitative Reverse-Transcriptase PCR for simultaneous assessment of drug activity against *Leishmania* intracellular amastigotes and their host cells.


Concomitant malaria among visceral leishmaniasis in-patients from Gedarif and Sennar States, Sudan: a retrospective case-control study.

**van den Bogaart E**, Berkhout MM, Nour AB, Mens PF, Talha AB, Adams ER, Ahmed HB, Abdelrahman SH, Ritmeijer K, Nour BY, Schallig HD.
Catechol pyrazolinones as trypanocidals: fragment-based design, synthesis, and pharmacological evaluation of nanomolar inhibitors of trypanosomal phosphodiesterase B1.

Prevalence, features and risk factors for malaria co-infections amongst visceral leishmaniasis patients from Amudat Hospital, Uganda.
van den Bogaart E, Berkhout MM, Adams ER, Mens PF, Sentongo E, Mbulamberi DB, Straetemans M, Schallig HD, Chappuis F.

Old and new targets for innovative antimalarial compounds: the different strategies of the Italian Malaria Network.
About the author

Erika van den Bogaart was born on August 3rd 1979 in Bergamo, Italy, and grew up in a small village on the Italian Prealps. After graduating from pre-university education in 1998, she continued her studies in Medicinal Chemistry and Technology at the University of Milan, for which she received her MSc in 2007. During this study, driven by her interest for tropical diseases, she served a 2-year internship at the Parasitology group of the University of Milan, where she gained extensive experience in the field of antimalarial drug discovery and pharmacodynamics. She continued working on the characterization of the mechanism of action of 4-aminoquinolines as a postgraduate fellow.

In November 2008, after having been entitled with her license to practice Pharmacy, she was awarded with a one-year personal fellowship from the University of Milan, by which she joined the Parasitology group at KIT Biomedical Research, Amsterdam. Here, she was introduced to the fascinating field of leishmaniasis, and developed the initial concept of what later has become the subject of her PhD study. In 2009, she began working on a project for the development of new phosphodiesterase inhibitors as novel therapeutic agents against leishmaniasis and trypanosomiasis, supported by the Top Institute Phar. Meanwhile, on a part-time basis, she pursued her studies on the visceral leishmaniasis–malaria co-infection, under supervision of Dr. Henk Schallig and Prof. dr. Martin Grobusch (Academic Medical Center). She collaborated with multiple international (Médecins sans Frontières, Blue Nile National Institute for Communicable Diseases in Sudan, IOTA Pharmaceuticals Ltd in the UK) and national (VU University in Amsterdam) partners and supervised several MSc students during their internship at KIT Biomedical Research. She presented her research at various national and international academic meetings, and prepared several publications for peer-review journals which have resulted in this thesis.

In 2012, she obtained a position as research fellow at the Rapid Diagnostics group of KIT Biomedical Research, where she has been responsible for the development and evaluation of simple diagnostic tests for poverty-related infectious diseases, and the application of novel technology platforms for diagnostic assays. In July 2016, she joined the Mondial Diagnostics Foundation as senior scientist, continuing her research activities on the development of simplified diagnostic tools for poverty-related diseases.

She lives together with Issa and their little son Rayan.