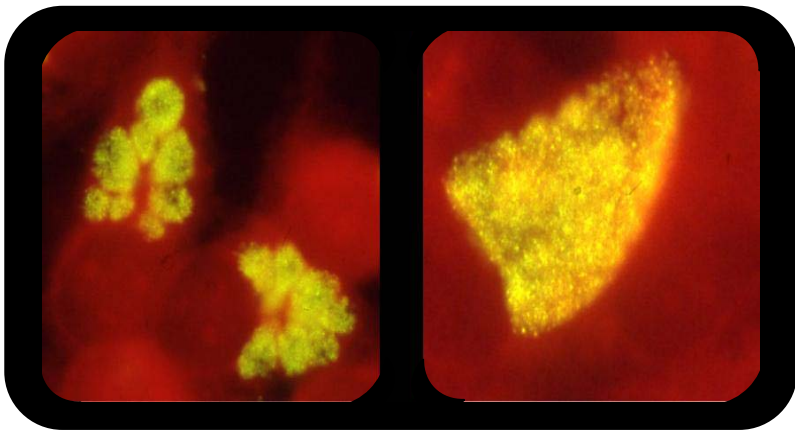


8th AACM

Eighth Annual Amsterdam

***Chlamydia* Meeting**



Hotel Mercure Amsterdam aan de Amstel

9 December 2011
9.00 – 17.45

Preface

Welcome: this year we organize our Annual Amsterdam *Chlamydia* Meeting (AACM) for the 8th time, and we included, like the last two years, all *Chlamydiae* species.

The symposium will be opened by Dr. Nicole Dukers informing us on CT clearance after antibiotic treatment. In total we have 14 senior and junior speakers. We are confident that the speakers will spark the minds of both young as well as established Chlamydiologists and trigger valuable discussions this day!

The Laboratory of Immunogenetics: it was established by Emeritus Prof. A. Salvador Peña in 1992, and has become part of the Department of Pathology in 2005 (Prof. Chris J.L.M. Meijer). The Laboratory links fundamental scientific research and clinical applications (translational research). Research is divided into two interactive and productive lines: chronic inflammatory diseases (J.B.A. Crusius, PhD) and infectious diseases (S.A. Morré, PhD; from Jan 1st, 2008, Head of the Laboratory of Immunogenetics).

Studies in twins and adopted children have shown that host genetic factors form an important element in the susceptibility to and the severity of infectious diseases such as *Chlamydia trachomatis*, *C. pneumonia*, and *C. psittaci* infections in humans. Bacterial, environmental, and host genetic factors determine the clinical course of *Chlamydiae* infections and an integrated multi-disciplinary approach is used to study these factors.

Acknowledgements: We would like to thank our main sponsor, Roche Diagnostics, without their support this meeting would not be possible in current format. We would also like to thank the other sponsors and those involved in the organization of this meeting. We like to express our gratitude to Thomson-Reuters and Prous Science (Barcelona, Spain) for the continuous support and for their contribution to make the publication of the 1st and 2nd proceedings possible in a special supplement of the journal "Drugs of Today" in 2006 and 2009.



Dr. Servaas A. Morré
Head of the Laboratory of
Immunogenetics

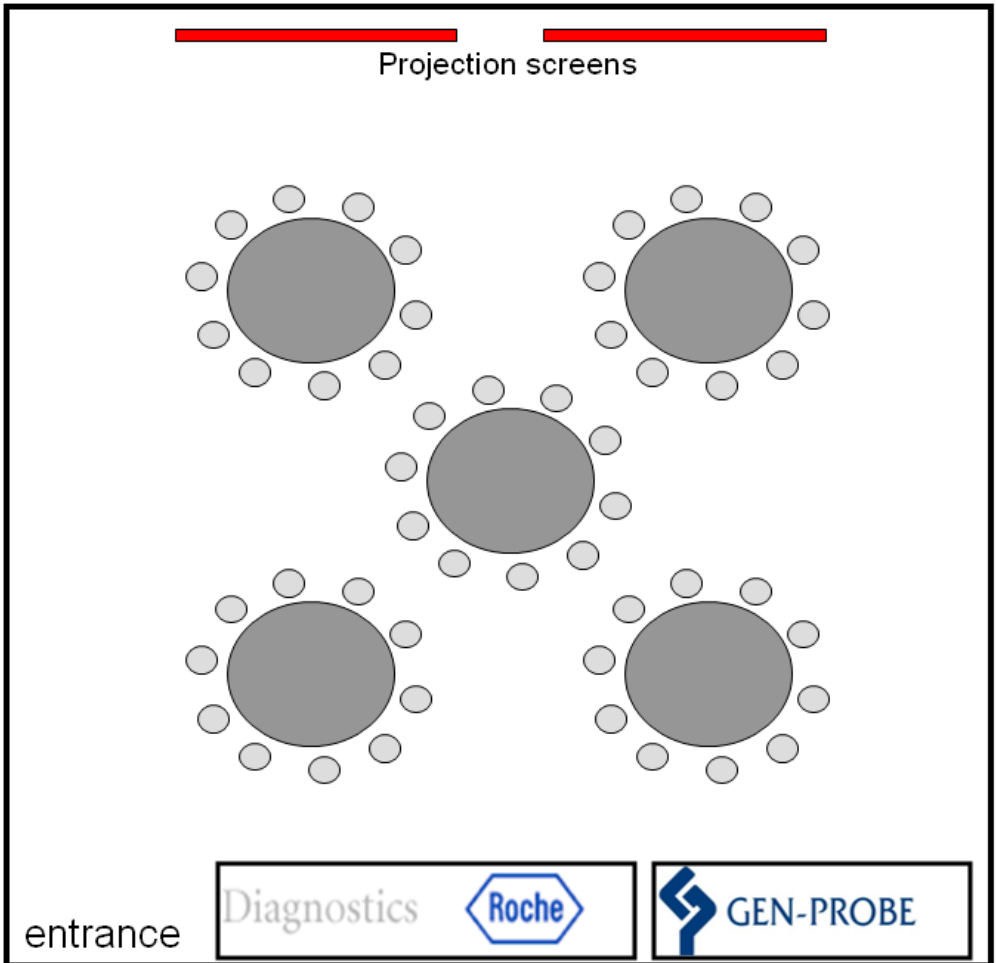


Dr. Sander Ouburg
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Cover photographs: Immunofluorescence staining of *Chlamydia trachomatis* within epithelial cells. HeLa cells were infected with a clinical isolate and stained with a monoclonal antibody specific for the major outer membrane protein (OmpA) of *C. trachomatis*. The left panel shows a nonfusogenic phenotype, while the right panel shows a fusogenic phenotype. Images courtesy of Yvonne Pannekoek, Department of Medical Microbiology, Academic Medical Center, Amsterdam, The Netherlands.

Floor plan (1st Floor, rooms 1+2)



Programme

- 9.00 – 9.30 **Registration to the symposium
(1st floor, Foyer outside Rooms 1-2)**
- 9.30 – 9.40 Opening: **Dr. Servaas Morré**
- 9.40 **Dr. Nicole Dukers (NL)**
Time to Chlamydia trachomatis clearance after treatment and implications for test of cure practices; First results of the Incure trial
- 10.10 **Dr. Nicole Dukers (NL), on behalf of Laura van Dommelen, MD**
Influence of temperature, medium and storage duration on Chlamydia trachomatis DNA detection by polymerase chain reaction
- 10.25 **Drs. Geneviève van Liere (NL)**
Alarming amount of missed non-urogenital infection with Chlamydia trachomatis and Neisseria gonorrhoeae when testing by standard operating procedures in men who have sex with men and swingers
- 10.40 **Drs. Stephan Verweij (NL)**
Chlamydia trachomatis vaginal swab based local immune response (proof of principle for IgG & IgA detection)
- 10.55 – 11.30 **Coffee Break (In front of the meeting room)**
- 11.30 **Dr. Sylvia Bruisten (NL)**
Using MLST typing on Chlamydia trachomatis for epidemiological purposes
- 12.00 **Drs. Monique Pereboom (NL)**
Chlamydia screening by a midwife
- 12.15 – 13.00 **Lunch (In font of the meeting room)**
- 13.00 **Dr. Marc Vandenbruaene (BE)**
Questioning azitromycin for Chlamydia infection

Programme

- 13.30 Dr. Servaas Morré (NL)
Introduction to CT antibiotic resistance: Dutch Reference tasks
- 13.45 Dr. Servaas Morré (NL)
Chlamydia trachomatis and subfertility: possibilities for improved triage for women
- 14.15 Dr. Yvonne Pannekoek (NL)
Chlamydia psittaci in MALT lymphomas: The Dutch experience
- 14.45 – 15.15 **Coffee break (In front of the meeting room)**
- 15.15 Drs. Stefanie Lagae (BE)
Role for the Chlamydia psittaci type III secretion apparatus in innate immunity
- 15.30 Dr. Eline op de Coul, on behalf of CSI (NL)
Dutch Chlamydia screening: CSI conclusions and future perspective
- 16.00 Dr. Ingrid Rours (NL)
Cost-effectiveness of Chlamydia trachomatis screening in Dutch pregnant women
- 16.30 Prof.dr. Henry de Vries (NL)
Point-of-care test in Surinam for detection of urogenital Chlamydia in women shows low sensitivity
- 17.00 – 17.10 **Closing remarks**
- 17.10 – 17.45 **Drinks (Foyer, lobby level)**



Nicole Dukers-Muijers, PhD

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Curriculum Vitae

Dr. Nicole HTM Dukers-Muijers (1972) is infectious diseases epidemiologist and coordinator research at the department of infectious diseases, public health service South Limburg, and also works at Caphri, University Medical Centre, Maastricht. With a masters degree in Biological Health Sciences & Epidemiology, University of Maastricht, she obtained her doctorate (2002) at the University of Amsterdam (promotors: Prof. Dr. R. Coutinho and Prof. Dr. J. Goudsmit). In total she has 15 years of relevant international work experience as a researcher and project leader in The Netherlands (PHS Amsterdam, University of Amsterdam, RIVM), San Francisco US (UCSF), China and Ethiopia. She published over 50 papers in international peer-reviewed journals including high impact journals as Journal of Infectious Diseases, New England Journal of Medicine, AIDS, American Journal of Epidemiology, European Journal of Epidemiology, PNAS USA, Vaccine, and Sexually Transmitted Infections.

Her research interest is in the field of public health, with a major focus on prevention of sexually transmitted infections, and on research methodology, including prospective cohort studies and sexual networks. In her projects, she combines biological, psycho-behavioural, and network assessments to gain a deeper understanding of the processes underlying acquisition, spread, and control of infections. Her research is mainly practice based, *i.e.* answering infectious diseases control driven questions and using information for societal application, with focus on networks. She was applicant of several funded grant proposals. She is an ad-hoc reviewer for journals 'AIDS', 'Journal of AIDS', 'Sexually Transmitted Infections', 'Journal of infectious diseases', and for research project funds, like ZonMW/NWO. She is a member of the editorial board of the journal BMC Public Health.

Abstract

Background: When a person, diagnosed with Chlamydia trachomatis (Ct), is being treated for Ct, STI Centres generally advise a short period of sexual abstinence; in most cases no test of cure is performed. These are considered best practices to cure Ct and prevent further spread, assuming high antibiotic



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Curriculum vitae

Laura van Dommelen graduated in Medicine in 2005 at the University of Maastricht. She started her training as consultant in Medical Microbiology at the Maastricht University Medical Centre and finished in July 2010. Currently, she is working as medical-microbiologist at the PAMM Laboratory for Medical Microbiology. Her interest in sexually transmitted infections started by a research project on syphilis serology. She subsequently started a thesis project on STI testing and will focus on *Chlamydia trachomatis* detection in different sample types in this presentation.

The abstract will be presented by Dr. Nicole Dukers

Abstract

Chlamydia trachomatis (Ct) is the most prevalent bacterial sexually transmitted microorganism worldwide and therefore continuous efforts are made to improve diagnostic assays. Many researchers conveniently use stored samples for their Ct research, since prospective studies are more laborious. We have studied the impact of temperature, type of medium, and duration of storage on Ct DNA detection.

For this purpose, phosphate buffered saline, 2-sucrose-phosphate (2-SP) medium, COBAS Amplicor medium and urine samples were spiked with Ct serovar D elementary bodies and were stored at room temperature (RT), 4°C, -20°C, and -80°C. Furthermore, clinical Ct positive urine samples, as well as Ct positive swabs in COBAS Amplicor medium were collected, pooled, and stored in triplicate, at the same temperatures. Samples were tested in triplicate on day 0 and subsequently after 1, 7, 14, and 30 days of storage for the presence of Ct DNA.

Ct could be detected in all media at all time-points, irrespective of the incubation temperature. For spiked PBS and 2-SP and pooled Ct positive swabs in COBAS Amplicor medium, the number of cycles needed was independent of storage duration and temperature. For Ct DNA detection in spiked COBAS Amplicor medium, the number of cycles increased over time at -20°C and -80°C (both



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Curriculum Vitae

Geneviève van Liere graduated in Health Sciences - Infectious Diseases in 2010 at the VU University in Amsterdam. She completed her Master thesis in Valencia, Venezuela where she worked in the field, in the lab, and studied prevalence and risk factors of the intestinal parasite *Schistosoma Mansoni*. Currently, she is working as a PhD student at the Public Health Service (GGD) South Limburg in collaboration with Medical Microbiology, Maastricht University. Her PhD project is about epidemiology of *Chlamydia trachomatis* with focus on testing policy.

Abstract

Background: Currently, persons at risk for sexually transmitted diseases (STDs) are only tested anorectally/oropharyngeally on indication, mostly guided by self-reported symptoms or self-reported anal sex. However, the suitability of these indicators is being debated. In this study we assessed the burden of undetected STD in risk groups, based on systematic screening at three anatomical sites.

Methods: MSM and high risk heterosexuals *i.e.* swingers, self-identified heterosexuals who as a couple have sex with other couples, attending a sexual health center, were systematically screened for urogenital, anorectal, and oropharyngeal STD yielding 2092 consultations. Prevalences of *Chlamydia trachomatis* (CT) and *Neisseria gonorrhoeae* (NG) were assessed, as well as anatomic site distribution of CT and the rate of missed anorectal diagnoses if tested by behavioral-indication only.

Results: Prevalences of anorectal STD were 12%, 10%, and 7% in homosexual MSM, bisexual MSM, and female swingers, respectively. Report of anal sex was not associated with anorectal STD. In the risk groups, between 50%-60% of the anorectal STD infections were diagnosed without self-reported anal sex. Symptoms were reported by 5% and associated with anorectal STD, but only when anal sex was reported. In MSM, prevalence of oropharyngeal STD was 4%, which was comparable to prevalence of urogenital STD.

Conclusions: Self-reported symptoms and anal sex should not be used as indicators for anorectal STD testing in MSM and female swingers. In these groups, systematic anorectal screening is warranted, especially considering



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Curriculum vitae

Stephan Verweij, born on June 22nd 1988, is a master student biomedical sciences at the VU University Amsterdam. During his master internship at the department of pathology, laboratory immunogenetics, he worked with *Chlamydia trachomatis* (CT). He developed an L2b specific primer/probe set, this manuscript is submitted to Clinical Microbiology and Infections. He also continued his research to relations between serovars and serological responses. In June 2010 he presented these findings at the International Symposium for Human *Chlamydial* Infections in Austria during a poster presentation. In October 2009, he participated in the EpiGen*Chlamydia* Training Challenge at the Oxford University. Currently, he is finishing his master's degree and is working at the laboratory of immunogenetics of the VU medical center on serological response studies in patients having CT.

Abstract

It is still unclear which women will progress to symptoms and late complications after a *Chlamydia trachomatis* (CT) infection. The use of an easy obtainable non invasive marker would be of great value to identify women at enhanced risk and potentially intensified treatment and/or follow-up. In the current study we investigated whether local vaginal immunological responses (IgG and IgA) can be measured (proof of principle) by comparing the local results to systemic serological results of these markers and to the presence of CT DNA in the vaginal swab. Concordance within and between local and systemic determination for IgG and IgA are compared to the DNA status. In the current presentation obtained results will be presented and discussed and the potential future applications outlined.



Sylvia Bruisten, PhD

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Curriculum vitae

Sylvia Bruisten is a medical molecular microbiologist. She is currently employed as the head of the department of molecular diagnostics at the Public Health laboratory of the Municipal Health Service (GGD) Amsterdam, the Netherlands. She is also supervisor of several PhD students who perform molecular epidemiological studies on hepatitis viruses (HAV, HBV, and HCV), and also on several sexually transmitted bacteria such as *Chlamydia trachomatis*.

Sylvia studied Biology/Biochemistry in Nijmegen after which she started her PhD project on 'the regulation of Complement genes in the mouse' at the Netherlands Cancer Institute in Amsterdam at the Department of Immuno-Genetics supervised by Prof dr. P. Borst. After completing her thesis in 1989, she worked as a molecular biologist at the CLB (now called 'Sanquin') She participated in coöperative studies with the Academic medical center and the GGD Amsterdam, on the Amsterdam Cohort studies, that all involved the early detection of HIV-1 sequences in blood and blood products.

She is currently a member of several boards of committees (BBC-MMO, CK, CTB) and working groups (NWKV, WMDI) of the Dutch Microbiology organisation, NVMM. She was the chair organiser of a European Virology congress in 2009 at the VU Medical center.

Abstract

A multilocus sequence typing (MLST) method of *Chlamydia trachomatis* (CT) was developed and evaluated at the Public Health Laboratory of the GGD Amsterdam using a well defined set of heterosexual couples. This MLST method proved to be stable for all 6 polymorphic regions including the genovar determining *ompA* region. We applied this MLST typing method to characterize transmission patterns of *Chlamydia* infection among different groups at risk.

We collected samples from different high risk populations: men having sex with men (MSM) (n=222 CT positive) and heterosexuals (n=256 positive), both visiting the Amsterdam STI clinic in 2008 – 2009, women visiting a birth control clinic during 2009-2010 in Paramaribo, Suriname, and males and females visiting an STI clinic in Paramaribo, Suriname. (total n= 183 CT positive). Epidemiological

data were collected using structured questionnaires. The samples (urethral, anal, vaginal) were all tested for *chlamydia* positivity with TMA (GenProbe) and in MSM with an in house LGV/non-LGV PCR. Positive samples with a sufficiently high load were typed using MLST. Large numbers of samples (n=32) could be sequenced in a single run using adapted M13 primers. Minimum spanning trees were generated in BioNumerics and *Chlamydia* clusters (defined as strains with zero or one single locus variance) and combined with the epidemiological data. In accordance with previous studies the *ompA* genovar distribution differed between heterosexual patients and MSM. MLST typing demonstrated a stronger separation between homosexual and heterosexual patients. In MSM CT strains were grouped in several large clusters coinciding with predominantly genovars G, D, and J. Among heterosexuals CT clusters were much more diverse and smaller, with the genovars E, F, and D as the most prevalent types. This evokes the question whether these differences are due to biological factors (such as differences in tissue tropism for specific CT strains), or due to behavioural differences (such as partner selection). For the Surinamese study only samples from heterosexuals, mostly from women, were included. Some particular CT clusters circulated mainly among people from Surinamese descent, both in Paramaribo and in Amsterdam, and were largely absent from non-Surinamese patients. The Surinamese population is characterized by a multi-ethnic background; CT strains from all clusters were spread homogeneously among the various ethnic groups, suggesting that disassortative ethnic sexual mixing is common. In conclusion, MLST proved to be useful to discriminate various CT transmission clusters among populations at risk for infection.



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Curriculum vitae

Monique Pereboom was born on March 20th 1982 in Vinkega (The Netherlands). After finishing her Bachelor in Nursing in 2006 at the Hanzehogeschool Groningen, she studied Health Sciences - Infectious Diseases at the VU University in Amsterdam. She accomplished her master thesis in Guinea Bissau - West Africa (Bandim Health Project) where she investigated the risk factors for measles hospitality en mortality. In April 2009 she started her work as a junior researcher at the Department of Midwifery Science.

Her PhD project is about the perspectives of primary care midwives and pregnant women regarding *Chlamydia trachomatis* and other infectious diseases in pregnancy.

A part of her PhD project is about the prevalence of *Chlamydia trachomatis* in pregnant women and about the knowledge and screening methods of primary care midwives. This PhD project is part of the national study "DELIVER; Data Primary Care Midwifery", (www.deliver-studie.nl).

Abstract

The prevalence of *Chlamydia trachomatis* (CT) is 4,4% in all Dutch women, and was estimated to be between 3,6% and 6,9% in pregnant women in Rotterdam. CT in pregnancy can result in adverse pregnancy outcomes, as miscarriage, preterm rupture of membranes, and preterm labour. However, CT in pregnancy can also be vertically transmitted. Up to 75% of the infants delivered vaginally from a CT infected mother acquire CT at some anatomic site.

Currently, screening pregnant women for CT infection is obligatory in Germany. The USA, Australia, and Canada highly recommend screening all pregnant women aged 24 or younger, as well as older pregnant women who are at increased risk for CT infection. European countries such as Czech Republic, Estonia, Germany, Portugal, Finland, Slovak Republic, and Sweden also recommend screening for *Chlamydia* in the antenatal care.

Although screening for some sexually transmitted diseases is already included in standard antenatal care in the Netherlands, pregnant women are not routinely screened for CT infection and at this moment reconsideration about including



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Curriculum Vitae

Marc Vandendruaene trained at the University of Antwerp and graduated as a medical doctor in 1985. He is an HIV-STI physician at the Institute of Tropical Medicine in Antwerp, Belgium (1988-2011). He has a background in medical journalism, in science communication, and in organizing post-graduate academic courses, symposia, and seminars on HIV/AIDS/ STI in Belgium for medical and paramedical professionals. From 1985-1987 he worked in Free Clinic-Antwerp, a youth health facility. He has been involved in *chlamydia*-screening programs in school health care (1995-2002). From 1995-2007 he coordinated activities of the Flemish AIDS-Reference Centers (KAR: Klinische Aids-Researchgroep, Vlaanderen). From 1988-2005 he worked closely with HIV/AIDS/STI-prevention organizations in Flanders.

Abstract

My presentation will be based on a symposium held during the 2011-ISSTD-Conference in Québec, entitled '*Questioning Azithromycin for Uncomplicated Genital Chlamydial Infection*' with presentations of Patrick Horner (Bristol, UK), Hunter Handsfield (Seattle, US), and William Geisler (Birmingham, US). I will bring the central ideas and data of these presentations, more specifically the clinical trial data. I will also add data on syphilis resistance. ISSTD: International Society for STD Research, www.isstdr.org.

Chlamydia trachomatis has longtime been considered as a micro-organism with low tendency for selection of resistance mutations. Online quote of a *chlamydia*-expert, answering *chlamydia*-patients questions: "You can be 100% certain the antibiotics you were given will cure *chlamydia*. Fortunately, *chlamydia* virtually never becomes resistant to antibiotics. That is a problem with many infections, but not this one" (<http://www.medhelp.org> 31 October 2005).

This statement was e.g. challenged by the findings in a Clinical Infectious Diseases article by JR Schwelke and colleagues. They write that organizing trials on non-gonococcal urethritis (NGU) is not really 'a sexy' subject anymore, so it has not been done in the last 15 years. They organized a clinical trial with 305 male NGU-patients in the US and found that doxycycline had significantly better efficacy against *Chlamydia trachomatis*, then azithromycin, with cure rates of 95% in the doxycycline and 77% in the azithromycin arm of the study. They conclude that ruling out re-infection is always very difficult in high-risk populations, but that their data could possibly be an indication of a real decrease in response of *Chlamydia trachomatis* over the previous 2 decades. For a clear understanding: their group did not perform resistance testing for *chlamydia*. [CID 2011:52(2):163-170].

Scrutinizing literature yielded in a 1973-Nature-article from the hand of H.Keshisyan. They performed *Chlamydia trachomatis* resistance testing for rifampin with yolk sack cell lines. They conclude that the selection of rifampin-resistant mutants in *chlamydial* populations is not fundamentally different from that in populations of gram negative bacteria [Nature 1973;244: July 20:173-174].

Chlamydia resistance testing is not routinely performed due to the absence of a standardized procedure to perform assays. Only 3 laboratories worldwide do resistance testing: 2 in the US and 1 in the UK. Cell lines are necessary. Comparison between different cell line experiments is very difficult if not impossible [personal communication, Servaas Morr ]. *Chlamydia trachomatis* antimicrobial resistance assays are difficult to understand and interpret, writes S. Wang and colleagues in the Journal of Infectious Diseases in 2005. There are more questions than answers: Which cell lines to be taken? How to deal with technical complexity? What about standardization, timing and duration of antibiotic exposure? What to take as endpoint: inclusions or DNA/RNA? [JID 2005;191:917]. Is the situation comparable with the history of resistance development in syphilis. During a long period syphilis experts tended to repeat that *Treponema pallidum* resistance selection was rare. Resistance testing was however not routinely done because of technical challenges. *Treponemata* only grow in vivo, in a rabbit model. Without testing it is hard/impossible to make statements about resistance development. Sheila Lukehart described in 2004 in the New England Journal of Medicine a case of syphilis patient treated with azithromycin, who returned to the clinic due to persistence of symptoms. A mutation in the *T. pallidum* 23S rRNA-genes was subsequently found (A2058G), in a strain of this patient. In-vivo rabbit-experiments, indeed confirmed functional azithromycin resistance of this strain. Lukeharts group sequenced 132 other *T. pallidum* strains from historically stored samples (1912-2003) from different geographical regions and found prevalences of this mutation ranging from 4% in San Francisco up to 88% in Dublin [N Engl J Med 2004;351(2):154-8]. The group of K. Van Damme sequenced syphilis strains of patients in Madagascar and found no evidence of mutants: zero mutants in 141 strains [Sex Transm Dis 2009;36(12):777-8]. Lukeharts data changed medical practice: the use of azithromycin for treating syphilis in geographical regions with macrolide resistance should be avoided.

Conclusion: Doubt has risen about efficacy of azithromycin for *Chlamydia trachomatis* therapy. Without standardized resistance testing it is hard to objectify possible resistance development. Ruling out re-infection is difficult. Data on possible *chlamydia*-resistance need confirmation. *Chlamydia*-experts propose a research agenda with trials comparing azithromycin versus doxycycline and with research on antimicrobial susceptibility testing of *Chlamydia trachomatis*. Interim recommendations for clinical practice contain more question tags, then clear guidelines. Should doxycycline be the therapy of choice for laboratory confirmed *chlamydia*-patients, who are willing to comply a 7-day regimen? Should we propose a test of cure after azithromycin therapy (> 3 weeks after therapy) ? Should we reemphasize retesting after 3-6 months?

Acknowledgements: I am grateful to Patrick Horner, Hunter Handsfield, and William Geisler who shared their 2011-Qu bec-ISSTD-slides with me, what made preparing this presentation much easier.

Further reading

Commentary article: H. Hunter Handsfield, MD. Questioning Azitromycin for Chlamydial Infection. Sexually Transmitted Diseases [2011;38(11):1028-9].



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Curriculum vitae

Servaas A. Morré, PhD, who is working on *Chlamydia trachomatis* for almost 17 years, graduated at the VU University, the Netherlands, in Biochemistry and Molecular Biology in 1994. From November 1st 2001, he joined the Laboratory of Immunogenetics, VUmc. His research is focused on the immunogenetics of infectious diseases with still special attention to *Chlamydia trachomatis*, bacterial meningitis (Prof Marceline van Furth, VUmc), and Human Papilloma Virus (Prof. C.J.L.M. Meijer). Together with Prof. Salvador Peña, he organised the "First Mini-symposium *Chlamydia trachomatis* Infections" in December 2004 and in December 2010 we organized our Seventh "Annual Amsterdam *Chlamydia* Meeting". In July 2005 at the 16th Biennial meeting of the International Society for Sexually Transmitted Diseases Research (ISSTD) he was a member of the Scientific Committee and organized amongst others the workshop "Immunogenetics of *Chlamydia trachomatis* Infections", with Prof. David Mabey (London, UK, Trachoma research). He was organizing Committee member of 6th Meeting of the European Society for *Chlamydia* Research, University of Aarhus, Aarhus, Denmark, July 1-4, 2008 and at this meeting also session organizer: "Immunogenetics of *Chlamydia trachomatis* infections". Since 2007 he is Scientific Consortium Director, of the European Framework Programme 6 (FP6) grant (LIFESCIHEALTH FP6, Co-ordination Actions (CA)) on functional genomics research entitled: "Contribution of molecular epidemiology and host-pathogen genomics to understand *Chlamydia trachomatis* disease (Acronym: EpiGenChlamydia)" with 20 European, African, and US groups. He will be the organizer of the 7th Meeting of the European Society for *Chlamydia* Research 1-6 July 2012 in Amsterdam. Together with Prof. Paul Savelkoul (Medical Microbiology and Infection Control, VUmc), he is co-founder and co-director of a VUmc spin-off company called Microbiome Ltd (Sept 2005), a company specializing in Medical and Microbiological diagnostics, typing, and laboratory consultancy. From the first of January 2008 he is head of the Laboratory of Immunogenetics. Since 1st of Sept 2009 several RIVM reference tasks concerning *Chlamydia trachomatis* were allocated to the VU University Medical Center, Laboratory of Immunogenetics. Finally from the 1st of June he is Director



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Curriculum vitae

Servaas A. Morré, PhD, who is working on *Chlamydia trachomatis* for almost 17 years, graduated at the VU University, the Netherlands, in Biochemistry and Molecular Biology in 1994. From November 1st 2001, he joined the Laboratory of Immunogenetics, VUmc. His research is focused on the immunogenetics of infectious diseases with still special attention to *Chlamydia trachomatis*, bacterial meningitis (Prof Marceline van Furth, VUmc), and Human Papilloma Virus (Prof. C.J.L.M. Meijer). Together with Prof. Salvador Peña, he organised the "First Minisymposium *Chlamydia trachomatis* Infections" in December 2004 and in December 2010 we organized our Seventh "Annual Amsterdam *Chlamydia* Meeting". In July 2005 at the 16th Biennial meeting of the International Society for Sexually Transmitted Diseases Research (ISSTD) he was a member of the Scientific Committee and organized amongst others the workshop "Immunogenetics of *Chlamydia trachomatis* Infections", with Prof. David Mabey (London, UK, Trachoma research). He was organizing Committee member of 6th Meeting of the European Society for *Chlamydia* Research, University of Aarhus, Aarhus, Denmark, July 1-4, 2008 and at this meeting also session organizer: "Immunogenetics of *Chlamydia trachomatis* infections". Since 2007 he is Scientific Consortium Director, of the European Framework Programme 6 (FP6) grant (LIFESCIHEALTH FP6, Co-ordination Actions (CA)) on functional genomics research entitled: "Contribution of molecular epidemiology and host-pathogen genomics to understand *Chlamydia trachomatis* disease (Acronym: EpiGenChlamydia)" with 20 European, African, and US groups. He will be the organizer of the 7th Meeting of the European Society for *Chlamydia* Research 1-6 July 2012 in Amsterdam. Together with Prof. Paul Savelkoul (Medical Microbiology and Infection Control, VUmc), he is co-founder and co-director of a VUmc spin-off company called Microbiome Ltd (Sept 2005), a company specializing in Medical and Microbiological diagnostics, typing, and laboratory consultancy. From the first of January 2008 he is head of the Laboratory of Immunogenetics. Since 1st of Sept 2009 several RIVM reference tasks concerning *Chlamydia trachomatis* were allocated to the VU University Medical Center, Laboratory of Immunogenetics. Finally from the 1st of June he is Director



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Curriculum vitae

Yvonne Pannekoek received her M.Sc. in 1987 at the University of Amsterdam where she studied Biology with emphasis on Molecular Microbiology. She graduated in 1993, at the University of Amsterdam, Dept. of Medical Microbiology of the Academic Medical Center, thesis entitled "Identification of Neisserial stress proteins: Molecular and immunological properties of Neisserial Hsp60". A part of her PhD work was carried out at the Max-Planck-Institut für Biologie, Abt. Infektionsbiologie, Tübingen, Germany, former laboratory of Prof. dr. Thomas F. Meyer where she worked as a visiting research fellow. After her graduation she joined the laboratory of Prof. dr. Patrik M. Bavoil, at that time situated in the University of Rochester, Dept. of Microbiology and Immunology, NY, US, where she worked as a postdoctoral fellow on the pathogenesis of *Chlamydia* infections. During that period she discovered the type III secretion system of *Chlamydia*. For this work she, together with other members of the Bavoil lab, received the best poster award during the Third European *Chlamydia* meeting that was held in 1996 in Vienna, Austria. In 1995 she returned to the Dept. of Medical Microbiology at the AMC in Amsterdam where she currently is appointed as Assistant Professor. Her main interests are the pathogenesis and molecular epidemiology of infections caused by *N. meningitidis*, *S. pneumoniae*, and *Chlamydiae*.

Abstract

Since the first publication in 2004, a large number of reports in peer reviewed journals have raised the question of an association between ocular adnexal lymphoma and a *Chlamydia psittaci* infection. Reports from several groups from various countries have investigated the prevalence of *C. psittaci* in their own series of ocular adnexal lymphomas by detection of *C. psittaci* DNA by PCR and displayed prevalence rates between 0 and 80%, with wide variability among countries and even within different regions of same country. These discrepancies could reflect a variable association between *C. psittaci* and ocular adnexal lymphoma in different geographical regions or methodological biases and will be discussed.



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Curriculum vitae

Stefanie Lagae, born on April 21st 1986, started her studies at Ghent University where she obtained her diploma of master in Biomedical Sciences in 2009. Currently, she is working as an assistant of Prof Daisy Vanrompay in the lab of Immunology and Biotechnology of the Animal Cell at Ghent University. Her work mainly focuses on the bacterium *Chlamydia psittaci* and more specifically bacterium-host cell interactions, immune responses, and vaccine development.

Abstract

The Type III secretion system (T3SS) of *Chlamydiaceae* plays an important role at different stages of their biphasic developmental cycle like for instance i) during entry, when inducing actin recruitment to the entry site following translocation of the T3SS effector protein tarp, ii) during resistance to phagolysosomal fusion through modification of the inclusion membrane, and iii) at the end of the developmental cycle when reticulate bodies detach from the inclusion membrane and differentiate to elementary bodies. The T3SS is highly conserved among several G- bacteria and plays also a role in regulating the innate immune response of the host cell following infection with pathogens such as *Shigella* spp., *Pseudomonas* spp., and *Burkholderia* spp. as well as *Chlamydia trachomatis*. *Chlamydia psittaci* also possesses a functional T3SS. Primary replication takes place in epithelial cells in upper respiratory tract. Later on, the bacteria can be found in epithelial cells and macrophages of the lower respiratory tract. Subsequently, *C. psittaci* can be found in plasma and blood monocytes, resulting in a systemic infection. Unfortunately, less is known about the underlying host innate immune response of *C. psittaci* infected macrophages and monocytes. As monocytes/macrophages play such an important role in the innate immune system, it is rather unique that *C. psittaci* as well as other *Chlamydiaceae* are able to survive and even replicate within those cells. In this way, the hypothesis arose that the T3SS might play a role in this process.



Eline Op de Coul, PhD

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Curriculum vitae

Eline Op de Coul graduated at the Agriculture University in Wageningen. She finished her PhD on the molecular epidemiology of HIV-1 at the Amsterdam Public Health Service (GGD) in 2001. From 2001, she started working as a senior epidemiologist in the field of HIV & STI surveillance at the Centre for Infectious Disease Control at the National Institute for Public Health and the Environment (RIVM) in the Netherlands. Currently, she is involved in the evaluation of screening programs (Chlamydia screening (CSI), infectious diseases screening among pregnant women), and projects on partner notification.

Abstract

Objective To evaluate the effectiveness of register-based yearly *chlamydia* screening.

Design Cluster randomised trial with stepped wedge implementation in three risk-stratified blocks.

Setting Three regions of the Netherlands; Amsterdam, Rotterdam, South Limburg.

Participants 317,304 16-29 year old women and men listed on municipal registers at start.

Intervention From March 2008 to February 2011, the *Chlamydia* Screening Implementation programme offered yearly *chlamydia* screening tests. Postal invitations asked people to use an internet site to request a kit for obtaining self-collected samples, which were sent for testing to regional laboratories. Treatment and partner notification were done by the general practitioner or at a sexually transmitted infection clinic.

Main outcome measures Percentage of *chlamydia* tests positive (positivity), percentage returning a specimen (uptake), estimated *chlamydia* prevalence. Secondary outcomes were: positivity according to sex, age, region, and sociodemographic factors, adherence to screening invitations, and incidence of self-reported pelvic inflammatory disease.

Results The participation rate was 16.1% (43,358/269,273) after the first invitation, 10.8% after the second, and 9.5% after the third invitation, compared



Ingrid Rours, MD, PhD

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Curriculum vitae

G. Ingrid J.G. Rours obtained her medical degree at the University of Amsterdam in 1990. She studied the relation between breastfeeding and malnutrition in North-East Brazil for the Royal Institute for Tropical Medicine, Amsterdam, and worked at the Institute of Childhealth in Dhaka, Bangladesh where she also participated in Shigellosis research at the International Centre of Diarrhoeal Disease Research, Dacca, Bangladesh. She temporarily worked at the Burgerziekenhuis, Amsterdam, and as a physician for the Royal Dutch Airlines in Dar-Es-Salaam, Tanzania. In 1991 she moved to Johannesburg, South Africa, where she worked as a medical officer and subsequently specialised in paediatrics working on rotation in the Chris Hani Baragwanath Hospital, Johannesburg General Hospital & Coronation Hospital of the Witwatersrand University, and the Child Abuse Clinic, Transvaal Memorial Institute. Meanwhile she started her research regarding *Chlamydia trachomatis* infections in pregnant women and infants in collaboration with the National Reference Centre for Sexually transmitted diseases and South African Institute for medical research. She returned to the Netherlands in 1997, worked as a paediatrician at the Maasstad Hospital, Sittard, and did a fellowship in neonatology until 2001 at the UMC St Radboud, Nijmegen. Subsequently she worked as a paediatrician in the POPD and child abuse clinic at the Erasmus MC-Sophia, where she also started a fellowship Paediatric Infectious Diseases and Immunology and research regarding *C. trachomatis* infections in pregnant women and infants in the Netherlands. From 2009 she worked as a paediatrician at the Maasstad Hospital, Rotterdam, and started a Master of Science Clinical Epidemiology at the Netherlands Institute of Health Sciences (NIHES). Since 2010 she has a paediatric practice, continues the chlamydial research at the Erasmus University and she has just finished her Master's degree in Clinical Epidemiology.

Abstract

Background: *Chlamydia trachomatis* infections during pregnancy may have serious consequences for women and their offspring. *Chlamydial* infections are largely asymptomatic. Hence, prevention is based on screening.



Prof.dr. Henry J.C. de Vries, MD, PhD

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Curriculum vitae

Henry de Vries is a dermatologist-venereologist with expertise in infectious skin diseases especially sexually transmitted infections and tropical skin diseases. His PhD thesis in 1994 was focussed on cutaneous wound healing and was rewarded with the Leiden Hippocrates Study prize 1995, and the Sandoz Research Prize 1997. Recent research topics involve; *lymphogranuloma venereum* proctitis, an emerging STI in mostly HIV positive gay men in industrialised countries, cutaneous leishmaniasis, an emerging infectious ulcerative tropical skin disease, and the viral pathogenesis of lichen ruber planus. He works as professor of skin infections at the University of Amsterdam, department of Dermatology and as consultant dermatologist at the Amsterdam municipal health service STI outpatient clinic, with 30.000 patients/year the largest STI setting in the country. He is expert advisor to the National Centre for Infectious diseases control, member of the national Health Council (Gezondheidsraad) which advises the minister on the mass HPV vaccination against cervical cancer and was chair of the commission to organise a quality and audit system for STI outpatient clinics appointed by the National Centre for Infectious diseases control.

Abstract

Background

Affordable and reliable point of care (POC) tests to diagnose urogenital *chlamydia* infections (POC-Ct) are needed, especially in resource limited settings. WHO has formulated standards that POC tests have to meet. One of those is that the test should be sensitive. Three POC-Ct tests currently on the market all showed poor sensitivity between 12% and 17% in a non-manufacturer sponsored clinical study (van Dommelen 2010). One POC-Ct test evaluated in a manufacturer-sponsored study claims over 80% sensitivity (Mahilum-Tapay 2007). We evaluated the performance of this POC-Ct in two outpatient clinics in Suriname, S.A.

Methods

Between July 2009 and February 2010 963 women were included in a high risk STI clinic (n1/4181) and a low risk birth control clinic (n1/4782) in Paramaribo, Suriname. Nurse collected vaginal swabs were obtained for the POC-Ct

(Diagnostics for the Real World, LTD, Cambridge, UK) and control NAAT (APTIMA Combo 2, Gen-Probe, San Diego, USA) in a cross-over model. Swabs were processed according to the manufacturers instructions. POC-Ct was compared to NAAT and sensitivity, specificity, positive- and negative predictive value (PPV, NPV) were calculated. Quantitative Ct load was determined with a real-time PCR targeting the cryptic plasmid. Ct load was expressed as inclusion forming units (IFU) based on defined serial dilutions. An independent t-test was used to compare log-transformed Ct loads between true positive and false negative POC-Ct results.

Results

Ct prevalence, determined by NAAT, was 23% at the high risk STI clinic and 9% at the low risk birth control clinic. Four samples were excluded due to discrepancy in POC-Ct result between two lab technicians (n1/43) and failure of POC-Ct (n1/41). Performance results of POC-Ct compared to NAAT are shown in the table. Quantitative Ct bacterial load was 65 times higher when POC-Ct detected Ct infection (geometric mean 115 IFU) compared to loads that POC-Ct did not detect (geometric mean 1.8 IFU, $p < 0.001$). Human DNA concentration did not differ between the true positive and false negative POC-Ct results ($p = 1/40.904$). Sensitivity of POC-Ct in samples with low Ct load was 16%.

Conclusion

The sensitivity and to a lesser extent the PPV of the POC-Ct did not meet the expectations as described previously (83.5%). The POC-Ct missed samples with a low Ct load. With a sensitivity of 41.7% the Diagnostics of the Real World POC-Ct test does not meet the ASSURED criteria of a sensitive test formulated by the WHO.

Table 1 Performance results of POC-Ct compared to NAAT

	NAAT +	NAAT -	Sensitivity	Specificity	PPV	NPV
POC-Ct +	48	30	41.7%	97.6%	61.5%	92.4%
POC-Ct -	67	814				



Symposium Organizer Servaas A. Morré

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Curriculum vitae

Servaas A. Morré, PhD, who is working on *Chlamydia trachomatis* for over 17 years, graduated at the VU University, the Netherlands, in Biochemistry and Molecular Biology in 1994. He worked at The Zaadunie, Department of Cell biology on plant genetics: polyploidization of *Brassica oleracea* (Cauliflower) during cell culture (M. Tan, PhD) and at the Department of Biochemistry and Molecular Biology VU on the genetic aspects of processing of ribosomal RNAs in *Saccharomyces cerevisiae* (Prof. H. Raué, PhD, R. van Nues PhD).

As an Erasmus Fellow he studied at the Universidade Do Porto, Laboratório de Genética Molecular, Portugal, on POLO: an essential kinase for mitosis in *Drosophila melanogaster* (Prof. C. Sunkel, PhD). His PhD thesis performed in Department of Pathology (VU University) was on the epidemiology, diagnostics, and immunopathogenesis of human urogenital *Chlamydia trachomatis* infections. As a postdoc, the Van Coeverden Adriani Foundation made it possible to extend his *Chlamydia* research in the Department of Infectious Diseases, The City of Hope Medical Center, California, USA, in collaboration with Dr. Jim Ito and Dr. Joseph Lyons, specialists in murine modeling. From November 1st 2001, he joined the Laboratory of Immunogenetics, VUmc. His research is focused on the immunogenetics of infectious diseases with still special attention to *Chlamydia trachomatis*, bacterial meningitis (Prof Marceline van Furth, VUmc), and Human Papilloma Virus (Prof. C.J.L.M. Meijer). Together with Prof. Salvador Peña, he organised the "First Mini-symposium *Chlamydia trachomatis* Infections" in December 2004 and in December 2010 we organize already our Seventh "Annual Amsterdam *Chlamydia Meeting*". In July 2005 at the 16th Biennial meeting of the International Society for Sexually Transmitted Diseases Research (ISSTD) he was a member of the Scientific Committee and organized amongst others the workshop "Immunogenetics of *Chlamydia trachomatis* Infections", with Prof. David Mabey (London, UK, Trachoma research). He was organizing Committee member of 6th Meeting of the European Society for *Chlamydia* Research, University of Aarhus, Aarhus, Denmark, July 1-4, 2008 and at this meeting also session organizer: "Immunogenetics of *Chlamydia trachomatis* infections". He is coordinator of the International *Chlamydia* consortium ICTI (Integrated approach on *Chlamydia trachomatis* Infections) and since 2007 he is Scientific Consortium Director, of the European Framework Programme 6 (FP6) grant (LIFESCIHEALTH FP6, Co-ordination Actions (CA)) on functional genomics research entitled: "Contribution of molecular epidemiology and host-pathogen genomics to understand *Chlamydia trachomatis* disease (Acronym: EpiGen*Chlamydia*)" with 20 European, African, and US groups. This consortium has recently submitted the FP7 EpiGen*Chlamydia*-II Consortium grant to the EU. As a partner he is participating in two other European FP6 programmes.

He will be the organizer of the 7th Meeting of the European Society for *Chlamydia* Research 1-6 July 2012 in Amsterdam. Together with Prof. Paul Savelkoul (Medical Microbiology and Infection Control, VUmc), he is co-founder and co-director of a VUmc spin-off company called Microbiome Ltd (Sept 2005), a company specializing in Medical and Microbiological diagnostics, typing, and laboratory consultancy. From the first of January 2008 he is head of the Laboratory of Immunogenetics. Since 1st of Sept 2009 several RIVM reference tasks concerning *Chlamydia trachomatis* were allocated to the VU University Medical Center, Laboratory of Immunogenetics. Finally from the 1st of June he is Director of the Institute of Public Health Genomics, Dept of Genetics and Cell Biology, Maastricht University where we works Thursdays and Fridays

Future STI / *Chlamydia* Meetings

- 10th German *Chlamydia* Workshop
April 4 - 6 2012, Erfurt, Germany
<http://131.130.66.201/dcw/>
- 6th *Chlamydia* Basic Research Society (CBRS)
2013, San Antonio (TX), USA
- 22nd ECCMID
March 31 – April 3 2012, London, United Kingdom
<http://www.congrex.ch/eccmid2012>
- 13th IUSTI World Meeting & 20th ISSTD
July 14 – 17 2014, Vienna, Austria
www.iusti.org / <http://www.stivienna2013.com/>
- 7th European *Chlamydia trachomatis* meeting
July 1 – 6 2012, Amsterdam, The Netherlands
- 9th Annual Amsterdam *Chlamydia* Meeting
December 2012, Amsterdam, The Netherlands

An overview of PhD work in The Netherlands on *Chlamydia trachomatis*

Table I: PhD theses in the Netherlands

2011 Koen D. Quint*	VU University Amsterdam
2010 Caroline J. Bax*	University of Leiden / Medical Center Haaglanden
2010 Janneke E. den Hartog*	Maastricht University
2010 Ingrid Rours	Erasmus University Rotterdam
2008 Liesbeth Duijts*	Erasmus University Rotterdam
2007 Denise A.M. Perquin	University of Leiden / Medical Center Haaglanden
2006 Sander Ouburg	VU University Amsterdam
2006 Joke Spaargaren*	University of Amsterdam and VU University Amsterdam
2006 Tanja P. Gijssen*	Maastricht University
2006 Hannelore M. Götz*	Erasmus University Rotterdam
2005 Jan E.A.M. van Bergen*	University of Amsterdam
2004 Joseph M. Lyons*	City of Hope Medical Center, CA, USA, and VU University Amsterdam
2003 Laura S. Murillo	VU University Amsterdam
2002 Monica Molano Luque	VU University Amsterdam
2001 Irene G.M. van Valkengoed*	VU University Amsterdam
1999 Servaas A. Morré*	VU University Amsterdam
1999 Johannes W. Trum	University of Amsterdam
1999 Bernardus W.J. Mol	University of Amsterdam
1998 Yvonne T.H.P. van Duijnhoven	University of Amsterdam
1997 Marita J.W. van de Laar	University of Amsterdam
1995 Jar Lan*	VU University Amsterdam
1994 Josina van Ulsen	Erasmus University Rotterdam
1994 Jacobus M. Ossewaarde*	University of Utrecht
1993 Hans J.H. Theunissen*	Erasmus University Rotterdam
1992 Johannes T.M. van der Schoot*	University of Amsterdam
1992 Arent J.P. Boeke and Janny H. Dekker	VU University Amsterdam
1992 André H. van der Willigen	Erasmus University Rotterdam
1991 Eric C.J. Claas	VU University Amsterdam
1990 Gijsbertus J.H.M. Ruijs*	Rijksuniversiteit Groningen
1989 Henk J. Vonsée	Rijksuniversiteit Limburg
1987 Kie H. Tjiam*	Erasmus University Rotterdam

**Chlamydia trachomatis* is the major focus in the thesis.

Table II: Current PhD fellows working (partially) on *Chlamydia trachomatis*.

Arnold Catsburg	VU University Amsterdam
Vitaly Smelov	St. Petersburg State Medical University, Russia and VU University Amsterdam
Laura van Dommelen	Maastricht University
Esmée Lanjouw	Erasmus University Rotterdam
Ouafae Karimi	VU University Amsterdam
Reinier Bom	University of Amsterdam
Stephan P. Verweij	VU University Amsterdam
Claire Geluk	Medical Center Haaglanden
Monique Pereboom	VU University Amsterdam
Jannie van der Helm	Public health services / University of Amsterdam

An overview of PhD work on *Chlamydiae*

Table II continued

Jonathan Lal	Maastricht University
Jelena Malogajski	Maastricht University
Ivan Brankovic	Maastricht University

Table III: PhD theses on *Chlamydiae*

2011 Veerle Dickx*	Ghent University, Belgium	CPs
2010 Katelijn Schautteet*	Ghent University, Belgium	C / CT
2010 Caroline van Droogenbroeck*	Ghent University, Belgium	CPs
2009 J.J.M. Bouwman	Utrecht University, The Netherlands	CP
2009 Delphine Beeckman*	Ghent University, Belgium	CPs
2008 Kristel Verminnen*	Ghent University, Belgium	CPs
2008 Taher Harkinezhad*	Ghent University, Belgium	CPs
2008 M.D. de Kruif	University of Amsterdam, The Netherlands	CP
2007 Edou R. Heddema*	University of Amsterdam, The Netherlands	CPs
2007 Ellen Boelen*	Maastricht University, The Netherlands	CP
2006 Arnaud Daniël Hauer	Leiden University, The Netherlands	CP
2005 Tom Geens*	Ghent University, Belgium	CPs
2005 Marnix Van Loock*	Catholic University Leuven, Belgium	CPs
2005 Manuela Voorend*	Maastricht University, The Netherlands	CP
2005 Tryphon Vainas	Maastricht University, The Netherlands	CP
2004 H.F. Berg	University of Amsterdam, The Netherlands	CP
2004 Boulos Maraha*	VU Universtiy, Amsterdam, The Netherlands	CP
1997 Roel P.A.J. Verkooyen*	Erasmus University Rotterdam, The Netherlands	CP
1994 Daisy Vanrompay*	Belgium	CPs

Table IV: Current PhD fellows working (partially) on *Chlamydiae*.

Stefanie Lagae	Ghent University, Belgium	CPs
Lizi Yin	Ghent University, Belgium	CPs / C
Evelien de Clercq	Ghent University, Belgium	CT
Kristien de Puyssseleyr	Ghent University, Belgium	C
Leentje de Puyssseleyr	Ghent University, Belgium	C
Sarah van Lent	Ghent University, Belgium	CPs

**Chlamydiae* are the major focus in the thesis.

C: *Chlamydiae*

CT: *C. trachomatis*

CP: *C. pneumoniae*

CPs: *C. psittaci*

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Announcement



8th
**Annual Amsterdam
Chlamydia Meeting**

December 2012

*Organiser: Servaas Morré
Laboratory of Immunogenetics,
Dept. of Pathology, VUmc, Amsterdam*

We hope to welcome you all in 2012

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Ing. Jolein Pleijster
Laboratory of Immunogenetics, Dept. of Pathology
VUmc, Amsterdam



Assistant symposium coordinator

Lay out & design, odd jobs:

Sander Ouburg, PhD
Laboratory of Immunogenetics, Dept. of Pathology
VUmc, Amsterdam

This symposium is accredited (5 points) by:

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