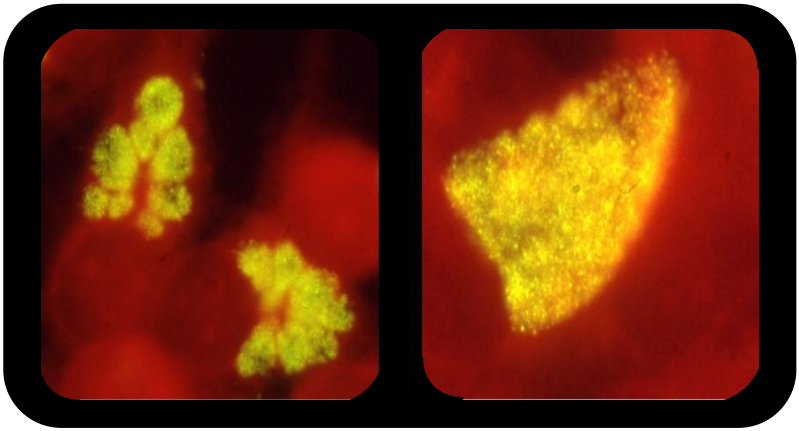
6th AACM

Sixth Annual Amsterdam

Chlamydia Meeting



Hotel Mercure Amsterdam aan de Amstel

17 November 2009
9.30 – 17.00

Preface

Welcome: this year we organize our Annual Anmsterdam *Chlamydia* Meeting for the sixth time, and we included, like last year all *Chlamydiae* species.

We are confident that our foreign key-note speakers: Prof. Robin Bailey (UK), Prof. Joseph Igietseme (CDC, US), Dr. Björn Herrmann (SE) and Dr. Delphine Beeckman (BE), together with the Dutch speakers will spark the minds of both young as well as established Chlamydiologists and trigger valuable discussions this day and enrich the Proceedings of this Symposium!

The Laboratory of Immunogenetics: it was established by Emeritus Prof. A. Salvador Peña in 1992, and has become part of the Department of Pathology (Head Prof. Chris J.L.M. Meijer) in 2005. 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. pneumoniae* and *C. psittaci* infections in humans. Bacterial, environmental and host genetic factors determine the clinical course of the *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 profound gratitude to Thomson-Reuters and Prous Science (Barcelona, Spain) for the continuous support and for their contribution to make the publication of the 2nd proceeding possible in a special supplement of the journal “Drugs of Today” in November 2009.

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

Dr. Sander Ouburg
Senior Postdoc
Infectious Diseases

Laboratory of Immunogenetics, Dept. Pathology, VUmc, Amsterdam, The Netherlands

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.

Amsterdam, 17 november 2009

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 – 11.10 Clinical *Chlamydia* infection

9.40 Prof. Robin Bailey (UK)
Ocular CT
10.05 Dr. Henry de Vries (NL)
LGV infections
10.30 Prof. Joseph Igietseme (US)
Chlamydia *vaccines*
10.55 Drs. Koen Quint (NL)
Role of C. trachomatis *in cervical pre-cancer and cancer among HPV-positive women*

11.10 – 11.40 **Coffee Break (In the meeting room)**

11.40 – 12.35 Chlamydiae and *Chlamydophila psittaci*

11.40 Dr. Tjaco Ossewaarde (NL)
Chlamydia – what else?
12.05 Ir. Delphine Beeckman (BE)
C. psittaci *host cell interactions*

12.30 – 13.30 **Lunch (In the meeting room)**

13.30 – 14.00 *Chlamydia* typing

13.30 Drs. Vitaly Smelov (RU)
C. trachomatis *serovar distribution in Russia*
13.50 Drs. Stephan Verweij (NL)
Serology and CT *serovars*

14.10 – 15.10 *Chlamydia* diagnostics

14.10 Prof. Paul Savelkoul (NL)
Roche COBAS 4800 vs. *Abbott RealTime*
14.35 Dr. Björn Herrmann (SE)
CT MLST typing
15.00 Drs. Laura van Dommelen (NL)
Point of Care testing

15.20 – 15.45 **Coffee break (In the meeting room)**

15.45 – 16.35 *Chlamydia trachomatis* in the Netherlands

15.45 Dr. Servaas Morré (NL)
Chlamydia trachomatis *research in the Netherlands*
16.10 Dr. Hannelore Götz (NL)
Participation and positivity rates in a large scale population-based Chlamydia *Screening Implementation in the Netherlands*

16.35 – 16.45 **Closing remarks**

16.45 – 18.00 **Drinks (Foyer, lobby level)**

Robin Bailey, MD, PhD, MRCP

London School of Hygiene and Tropical Medicine, London, England

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Curriculum Vitae

Robin Bailey is Professor of Tropical Medicine at the London School of Hygiene and Tropical Medicine in the UK, and Director of the Masters Programme in Tropical Medicine and International Health. He has been studying ocular *Chlamydia trachomatis* infection (trachoma) since 1984 when he was a medical student. Along the way he has qualified as an infectious disease physician, and has managed to spend a total of 8 years based in The Gambia, West Africa. He wrote his PhD on the epidemiology and immunology of trachoma in 1996. He is fortunate to be part of the largest group of researchers working on this disease worldwide. He has, somehow, published nearly 100 papers on all aspects of trachoma, its pathogenesis, immunology, epidemiology, microbial diversity, host susceptibility and control. He has active research projects in The Gambia, Senegal, Guinea-Bissau, Tanzania and Zambia. He is grateful to the UK Medical Research Council, the Wellcome Trust, the Edna McConnell Clark Foundation, and the Bill and Melinda Gates Foundation for supporting his work.

Abstract

If the cellular immune response to *Chlamydia trachomatis* (Ct) is subject to genetic influences the degree and mechanisms of such genetic control may have important implications for vaccine development. We estimated the relative contribution of host genetics to the total variation in lymphoproliferative responses to Ct antigen by analysing these responses in 64 Gambian twin-pairs from trachoma endemic areas. Zygosity was determined by RFLP analysis of minisatellite probes and microsatellite typing. Proliferative responses to serovar A EB antigen were estimated in monozygotic(MZ) and dizygotic(DZ) twin pairs. We found stronger correlation and lower within-pair variability in these responses in MZ than in DZ twin pairs. The heritability estimate was 0.39 (p=0.07) suggesting that host genetic factors contributed 39% of the variation. A better understanding of these genetic influences will contribute to the elucidation of preventive therapies for ocular Ct infection and may identify important mechanisms in protection for rational vaccine construction. In the current presentation we will summarize our (genetic) work on ocular *Chlamydia trachomatis* infections.

Amsterdam, 17 november 2009

Henry 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, leprosy in particular its immunological aspects, and the viral pathogenesis of lichen ruber planus. He works as consultant dermatologist at the Amsterdam municipal health service STI outpatient clinic, with approximately 29.000 patients/year the largest STI setting in the country, and at the University of Amsterdam, department of Dermatology. He is member of the HPV vaccine committee of the Dutch Ministerial Health Counsel and expert on STI of the Centre for Infectious Diseases control (CIb/RIVM) .

Abstract

Lymphogranuloma venereum (LGV) was formerly known as a sexually transmissible infection confined to equatorial regions but also as an “imported” sexually transmissible infection in the Western world. However since 2003, with the first cases of LGV proctitis among men who have sex with men reported in the Netherlands, an ongoing epidemic has been revealed in Western society dating back to at least 1981. In this presentation the state of the art diagnostics and treatment and common complications concerning LGV are discussed. Moreover, risk factors and the background of the recent epidemic of LGV in the Western world among men who have sex with men are summarized. There is a need for new diagnostic assays, to prevent complications and to protect the community of more expansive transmission. Shorter antibiotic treatment courses for LGV are necessary but require large controlled clinical trials. The microbial and immunological background of LGV infection in relation to HIV should be studied in detail and could help to explain the considerable number of asymptomatic LGV cases.

Amsterdam, 17 november 2009

Prof. Joseph U. Igietseme, PhD

Centers for Disease Control and Prevention (CDC), & Morehouse School of Medicine, Atlanta, Georgia, USA

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Curriculum vitae

Joseph Igietseme is the Chief of Molecular Pathogenesis Laboratory at the Centers for Disease Control and Prevention (CDC), Atlanta, Georgia USA, and Professor at both Morehouse School of Medicine and Emory University Medical School in Atlanta, Georgia, USA. Dr. Joseph U. Igietseme received his PhD in Immunology and Microbiology from Georgetown University, Washington, DC, USA in 1987. He subsequently trained in infection and immunity at the University of Miami School of Medicine, Miami, Florida, and the University of Arkansas for Medical Sciences, Little Rock, Arkansas, USA, focusing on T cell immunity, immunoregulation and immunopathogenesis.

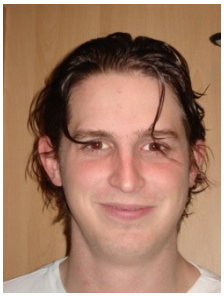
Dr. Igietseme was appointed as Assistant Professor in the Department of Microbiology & Immunology at the University of Arkansas for Medical sciences, Little Rock, Arkansas from 1993 -1996, as Associate Professor of Microbiology & Immunology at Morehouse School of Medicine, Atlanta Georgia from 1998-2002, and as Professor of Microbiology & Immunology at Morehouse School of Medicine, Atlanta Georgia in 2002. He was appointed as Chief of Molecular Pathogenesis Laboratory at the National Center for Infectious Disease, Centers for Disease Control and Prevention (CDC), Atlanta, Georgia USA in 2002. Dr. Igietseme maintains Professorial position at Morehouse School of Medicine and Adjunct Professor at Emory Medical School, Atlanta Georgia USA.

Dr. Igietseme's research focus is in Basic and applied immunology and microbiology, infection and immunity and Vaccinology. Identification of correlates of protective immunity, elucidating the cellular, molecular and biochemical mechanisms of immunity and immunopathogenesis, and designing effective delivery systems for vaccines against *Chlamydia*, *Herpes simplex viruses*, *Neisseria gonorrheae*, and other microbial agents of STDs are among his areas of active research. Dr. Igietseme's Research is supported by the CDC and the National Institutes of Health (NIH) and he has over 200 peer-reviewed research publications, reviews articles and presentations.
Dr. Igietseme is a member of NIH and CDC expert panels, grant review committees, and study sections, American Association of Immunologists,

American Society for Microbiology, American Association for the Advancement of Science, Georgia Academy of Science and a reviewer or Editorial Board of several biomedical journals.
Finally, Dr Igietseme has enormous expertise and skills in the development of biomedical science and research infrastructures and projects in medical schools, universities and agencies (Govt/private) settings.

Abstract

Genital infection by the obligate intracellular bacterium *Chlamydia trachomatis* is the most common bacterial STD worldwide with major public health concern. The rising infections some populations could predispose to HIV-related AIDS and HPV-associated cervical dysplasia, thus the urgency to develop a prophylactic vaccine as the most reliable and cost effective to achieve the greatest impact on control. Although vaccines are needed to prevent the oculo-genital diseases of *C. trachomatis*, the role of natural immunity after an infection is unclear. However, infected hosts appear to develop immunity, although temporary, and experimental vaccines have yielded significant protective immunity in animal models, fueling the impetus for a vaccine. Since infections cause sequelae, the functional relationship between infection- and vaccine-induced immunity is unclear. We hypothesized that infection- and vaccine-induced immunity are functionally distinct, particularly in the ability to prevent sequelae due to the differential profile of immune effectors induced. *Chlamydia* immune mice, generated by either a previous infection or vaccination, exhibited a significant degree of protective immunity, marked by a lower intensity and abbreviated course of infection. However, vaccinated mice were protected from infertility whereas pre-infected mice were not. Thus, infection-induced immunity does not prevent the pathologic process leading to infertility. In addition, T cell subsets and more prominently the CD8 T cells, play a major role in *Chlamydia*-induced infertility. Furthermore, the substantial loss of the ability of plasmidless *C. trachomatis* to induce infertility in mice and the significant resistance of IL-10 deficient mice against *Chlamydia*-induced infertility has lead to the working model that *chlamydial* disease involves the activation of specific immunopathogenic inflammasome platforms and T cell-mediated immune responses by gene product(s) of the *chlamydial* cryptic plasmid. The results have significant implications for the immunopathogenesis of *chlamydial* disease and new vaccine strategies.



Koen Quint

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Curriculum Vitae

K. Quint (k.d.quint@umail.leidenuniv.nl, born 15-12-1984). At this moment he is in his final year for the Master of Biomedical Science as well as Medical school. During his study he was a coworker for DDL Diagnostics Laboratory, where he developed a Ct- amplification, detection and genotyping assay. This was the start of his PhD project. In 2007 he did a 6 months internship at the Weill Medical College of the Cornell University in NY. He investigated the association between the different HPV types and the natural course of a LSIL lesion. The role of Ct in the course of LSIL lesions was also investigated.
At this moment he is working in collaboration with the National Cancer Institute (USA) on a project to investigate the role of Ct as co-factor in development of cervical cancer and with the VU Amsterdam on serovar distribution studies in different populations.

Abstract

BACKGROUND: Although infections with carcinogenic HPV are a necessary cause of cervical cancer, infections are extremely common relative to cancer. It is postulated that co-factors might increase the risk of progression to cervical precancer and cancer. Some, but not all studies have shown *Chlamydia trachomatis* (Ct) to be a risk factor for cervical cancer.
OBJECTIVE: To assess the role of Ct in cervical precancers and cancer.
METHODS: We identified 327 cumulative cases of prevalent (n=185) or incident (n=142) histological CIN2/CIN3/cancer (CIN2+) in the Proyecto Epideiologico Guanacaste (PEG) cohort, and sampled 10% of subjects at enrollment (n=1100) as controls. Type-specific cervical HPV status over multiple visits was determined using PCR amplification and reverse line-blot hybridization. Ct DNA was determined using a novel serovar-specific PCR-based Ct detection and genotyping assay (Labo Biomedical Products, Rijswijk, The Netherlands) on cervical samples. Corresponding plasma was used to determine Ct IgG and IgA status (Medac Diagnostika, Hamburg, Germany). Behavioural factors were determined from questionnaires.
RESULTS: The measures of HPV and Ct positivity were strongly associated, as expected for two sexually transmitted agents. There was no association between

Ct DNA or serology and CIN2+ among carcinogenic HPV DNA-positive cases and controls (OR=0.77, 95%CI=0.42-1.41 for Ct DNA-positivity; OR=1.09, 95%CI=0.85-1.41 for Ct seropositivity).
CONCLUSIONS: We did not find an association between Ct DNA or serology positivity and CIN2+ among carcinogenic HPV-positive women. The details of the analysis suggest that the previous positive finding between Ct and cervical cancer could be due, in part, to confounding by HPV status.

Tjaco Ossewaarde, MD, PhD

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Curriculum vitae

Dr. Ossewaarde studied medicine at the University of Leiden from 1973 to 1981. During his study he was actively involved from 1976 to1978 in laboratory research into cancer. His military service from 1982-1983 lead him to the National Institute of Public Health in Bilthoven studying the occurrence of Q fever among UNIFIL military personnel in Lebanon. From 1983 to 1987 he followed a specialist training in medical microbiology, especially clinical virology, at the University of Utrecht (prof. J. Verhoef and prof. A. Hekker). He wrote a PhD thesis on *Chlamydia trachomatis* laboratory research in 1994. At the Hogeschool Utrecht he teached virology from 1985 to 2002. He was employed by the National Institute of Public Health from 1987 to 2001 and participated in clinical virology, public health research, STD research (*Chlamydia trachomatis*), and chronic diseases research (*Chlamydophila pneumoniae* and atherosclerosis). In 2001 he joined the Department of Medical Microbiology and Infectious Diseases of the Erasmus MC at Rotterdam. In 2005 he moved to the Maasstad Ziekenhuis in Rotterdam, while continuing teaching students at the Erasmus MC (0.1 fte employment).

Abstract

In this presentation we will review the whereabouts of the *Chlamydia* family since the tree was greatly expanded a decade ago. Phylogenetically, *Chlamydiae*, *Lentisphaerae*, and *Verrucomicrobia* have now found each other in one group connecting environmental microorganisms to animal and human pathogens. A great diversity has been documented.

Chlamydia (and *Chlamydophila*) species have been well studied as pathogens in man and animals. Most infections involve the mucosal lining, while some become systemic. Well defined tools exists for their detection. But not so for many of their related cousins, the chlamydia-likes. The natural habitat of most of them is unknown, although a few have been described in more detail, even connecting them to diseases in men and animals. One might postulate that each host, being man, animal or even a one cellular organism, harbors at least one member of the *Chlamydia* family as a pathogen or symbiont and is usually susceptible to cross-infection with other members.

Amsterdam, 17 november 2009

Respiratory and reproductive tract diseases caused by *Chlamydia* (and *Chlamydophila*) species share characteristics with similar diseases caused by *chlamydia*-likes. Respiratory tract infections in man ranges from subclinical infection to severe pneumonia necessitating ventilator assistance. Reproductive tract infections might cause fetal impairment and abortion.
We will review some (dis)similarities between the classical *Chlamydiae* and the *chlamydia*-likes in these types of infection with emphasis on the *chlamydia*-likes.

Delphine Beeckman, PhD

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Curriculum vitae

Delphine S.A. Beeckman was born on August 20th 1982 in Ghent (Belgium). After finishing Latin-Mathematics in 2000 at the Koninklijk Atheneum III – Ghent, she studied Bio-Engineer option Cell and Gene Biotechnology at Ghent University. In October 2005, she started her PhD in the laboratory of Immunology and Biotechnology of the Animal Cell at Ghent University, funded by the Research Foundation – Flanders (FWO – Vlaanderen). The research mainly focused on the role of the Type III secretion system in the biology and intracellular pathogenesis of *Chlamydophila psittaci*. She successfully defended her PhD in May of this year and is currently working as a Post-doctoral Fellow (FWO - Vlaanderen) in the same lab. She currently examines the role of Type III secretion in the immune response elicited by *Chlamydophila psittaci* infections. The aim is to gain insight in the fundamental molecular mechanisms involved in the observed differences in virulence between *Chlamydophila psittaci* strains, in both the avian and human host.

Abstract

Within a few days post infection of SPF turkeys, highly pathogenic *Chlamydophila* (*Cp.*) *psittaci* genotype A and D strains can be found in blood monocytes/macrophages, while this effect is less pronounced for infection with a milder genotype B strain. To elucidate on the observed difference, we studied the developmental cycle of avian *Cp. psittaci* strains of varying virulence in a matched avian monocyte/macrophage cell line (HD11) by electron microscopy and immunofluorescence and determined the gene transcription of 26 Type III secretion related genes and six control genes upon infection of HD11 cells. *Cp. psittaci* strains used were 84/55 and 92/1293 (highly virulent), CP3 (low virulent) and 84/2334 (phylogenetically intermediate between *Cp. psittaci* and *Cp. abortus*). The genotype A (84/55) and D (92/1293) strains 1) clearly induced actin recruitment to the site of entry, 2) initiated host cell degeneration at earlier time points, and 3) survived and proliferated better when compared to the milder CP3 strain. Strain 84/2334, genetically intermediate between *Cp. psittaci* and *Cp. abortus*, did not induce actin recruitment. Limited mRNA transcripts for the cell

division genes ftsW and ftsK were in agreement with the observed low replication of *Cp. psittaci* in these host cells. The results also indicated that genes coding for the structural components of the Type III secretion system were transcribed earlier compared to an infection in epithelial cells. We postulate that upon infection of blood monocytes/macrophages, *Cp. psittaci* deliberately limits its replication and immediately arms itself to infect other cells elsewhere in the host, whilst using the monocytes/macrophages as a quick transport vehicle. *Chlamydophila psittaci* and avian pathogenic *Escherichia* (*E.*) *coli* infections contribute to the respiratory disease complex observed in turkeys. Secondary infection with *E. coli* exacerbates *Cp. psittaci* pathogenicity and augments *E. coli* excretion. The innate immune response initiated by both pathogens in their avian host is unknown. We therefore determined the cytokine responses following *Cp. psittaci* infection and *E. coli* superinfection of avian monocytes/macrophages by examining gene transcripts of IL-1β, IL-6, CXCLi2 (IL-8), CXCLi1 (K60), IL-10, IL-12α/β, IL-18, TGF-β4 and CCLi2 at 4 h post inoculation with different *Cp. psittaci* strains or 4 h post treatment with avian *E. coli* LPS of *Cp. psittaci* pre-infected HD11 cells. The same *Cp. psittaci* strains as mentioned above were included in the study. At 4 h post chlamydial infection, an increased expression of IL-1β and IL-6 as well as CXCLi2, CXCLi1 and CCLi2 was observed compared to levels in uninfected HD11 controls. This effect was less pronounced for the milder CP3 strain. The pro-inflammatory response of *Cp. psittaci* infected cells to *E. coli* LPS was significantly lowered compared to uninfected controls, especially when the cells were pre-infected with highly virulent *Cp. psittaci* strains. In both experiments, exceptionally high IL-10 and no TGF-β4 responses were observed, and we propose that this could induce macrophage deactivation and NF-κB suppression. Consequently, pro-inflammatory and Th1-promoting responses to both the primary *Cp. psittaci* infection and *E. coli* would be inhibited, thus explaining the observed aggravated in vivo pathology.

Vitaly Smelov, MD

Faculty of Medicine, St. Petersburg State University & St. Petersburg State University Outpatient Clinic, St. Petersburg, Russia

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Curriculum vitae

Vitaly Smelov was born in Neustrelitz, Germany in 1970 and was graduated from the Military Medical Academy, St. Petersburg, Russia in 1993. Since 1997 he is a certified urologist and interested in the studies in the "grey area" between urological diseases and sexually transmitted infections. He also has a clinical background in the field of urogenital tuberculosis (City Tuberculosis Hospital) as well as chronic inflammation of the prostate and long-term antibacterial treatments of such patients.

In 2002-2004 he worked as a researcher in Nijmegen, the Netherlands. Being a winner of the European Urological Scholarship Program for the period of 2003-2004, he got lab skills and was involved in the studies on the development of new prostate cancer markers.

Since 2004 V. Smelov is a stuff member of the Faculty of Medicine, St. Petersburg State University and, meanwhile, works as an urologist in 2 STI clinics. He initiates or becomes involved in several research projects together with people from the VU University and GGD Amsterdam and was awarded with the UNESCO-ASM Travel Award and EUSP Clinical Fellowship in 2006 and 2007, respectively. V. Smelov is a PhD fellow at the VU University, Amsterdam since 2006 and his thesis is planned to be defended in the spring 2010.

Abstract

The data on serovar distributions of *Chlamydia trachomatis* – the most diagnosed sexually transmitted infection (STI) worldwide – are important for epidemiologic purposes and transmission studies but are completely lacking in Russia. This is primarily the result of suboptimal diagnostics, case reporting, and surveillance systems. However, in recent years, several DNA and RNA amplification systems (NAATs) have been developed and widely used in Russia in *C. trachomatis* diagnostics, the first prevalence figures have been published and screening studies have been initiated.

The preliminary and already published data on *C. trachomatis* prevalence within different groups in Russia will be presented.

Amsterdam, 17 november 2009

With the current study we are the first to determine the serogroup and serovar distribution in Russian women and men. In addition, the obtained serogroups and serovars are compared with two independent large distributions assessed in the Netherlands.

Stephan Verweij, BSc

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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 bachelor internship at the department of pathology, laboratory Immunogenetics, he worked with *Chlamydia trachomatis* (CT) and found some relations between serovars and serological responses. These findings will be published in 'Drugs of Today, supplement B' (November 2009). In March 2009 he presented his findings at the Maastrichts Medical Students Research Congress 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 and is working at the laboratory of Immunogenetics of the VU medical center on serological response studies in patients having CT.

Abstract

Chlamydia trachomatis serovars are divided into three serogroups, namely serogroup B, serogroup I (intermediate) and serogroup C, and subsequently into 19 different serovars. Worldwide, serogroup B is the most prevalent followed by serogroup I. Clear differences have been observed in the duration of infection and growth kinetics between serovars from different serogroups in murine and cell culture models. Reasons for these observed differences are bacterial and host related, and are not well understood. The aim of this study was to determine the differences in immunoglobulin (Ig) G responses between the three serogroups in a group of patients infected with different serovars. Serovars were assessed from 235 *C. trachomatis* positive patients and quantitative IgG responses were determined. Analyses of variance were used to compare the IgG responses between the three serogroups. Of the serovars, 46% were B group (with serovar E the most prevalent: 35.3%), 39.6% were I group, and 14.3% were C group. A highly significant difference in serologic response was shown when comparing the mean IgG concentrations (AU/mL) of patients having serovars in the most prevalent serogroup compared to the other serogroups: B = 135, C = 46, and I =

60 (B *vs.* C, and B *vs.* I; P < 0.001). In conclusion, the most prevalent serovars generate the highest serologic responses.

Prof. Paul Savelkoul, PhD

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Curriculum Vitae

Prof. Dr. Paul HM Savelkoul graduated in Biology (specialization Medical Biology B5*) in 1988 at the University of Utrecht, The Netherlands. After graduation a PhD study was started at the department of veterinary Bacteriology, University of Utrecht under auspices of prof. dr Ben van der Zeijst. In 1992, he received his PhD-degree on the development of detection and characterization of adhesion factors of *Bordetella*. Subsequently, in 1992 a position at the University hospital Maastricht was accepted to set up a molecular laboratory for HLA typing. This was achieved after three years and the HLA typing was performed on a molecular basis for solid organ transplantation purposes. In 1995 he joined the group of prof. dr. Christina Vandenbroucke-Grauls at the VU University in Amsterdam as an assistant professor. Within the department of Medical Microbiology & Infection Control he was involved in setting up a molecular typing laboratory, molecular diagnostics, microbiological research & education. In 2000 he was appointed associated professor in molecular microbiology at the VU University medical center in the same department as head of the molecular diagnostics and molecular epidemiology. In 2006 he was appointed professor in molecular epidemiology at the same institute and head of the translational molecular research of the department. He has published over 100 papers in the fields of molecular microbiology, molecular epidemiology, HLA typing and human genetics related to basic research and applied and translational research.
Currently, he is involved in clinical molecular diagnostics both in the field of infection control and patient care. Research is focused on three subjects: bacterial blood stream infections, chronic gastrointestinal diseases and molecular epidemiology especially the spread of epidemiological strains and mobile DNA elements.

Abstract

The VU University hospital was invited to evaluate the new CT/NG test (Cobas® 4800 CT/NG Test) to be able to test the new kit in a routine setting for CE-IVD registration. A state of the art comparator test was chosen (Abbott m2000). In Groningen a The Laboratory of Infectious Diseases was able and willing to collect samples and sent the samples to Amsterdam. In this lab the Abbott test was used as routine test. The comparison was carried out in 2 stages. The early evaluator

stage was meant to be able to get proof of principle in a routine setting and to make some practical changes in performance thus ensuring success during CE-IVD stage. The second stage was a large prospective study. We will report the evaluation and discuss general performances for the different clinical specimens evaluated.

Björn Herrmann, PhD

Department of Clinical Microbiology, Uppsala University Hospital, Uppsala, Sweden

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Curriculum Vitae

Björn Herrmann is associate professor in Clinical Microbiology at Uppsala University, Sweden. He gradutated in Microbiology at Umeå University 1982 and after two years work with methanogenic bacteria at the Swedish University of Agricultural Sciences he shifted to clinical bacteriology at the Academic Hospital, Uppsala. He has also had commissioned work at the Swedish Institute for Infectious Disease Control and is currently linked to the institute as responsible for diagnostics of *Chlamydia trachomatis* infections.
In clinical microbiology he has mainly worked with *Chlamydial* infections and been interested both in diagnostics and epidemiological aspects. In recent years molecular epidemiology has been a focus and after having shown the limitations of conventional *ompA* genotyping his group developed a multilocus sequence typing system for short term epidemiological purposes. The emergence of the new “Swedish” variant of *C. trachomatis* is at present a concern. A side branch of *Chlamydia* research is investigations of wildlife birds and this has led to findings and hypothesis of the spread of *Chlamydophila psittaci* infections.
Another research field is species identification and detection of bacteria based on the “universally” present *rnpB* gene, which has highly conserved and hypervariable regions that enable diagnostics of any kind of microorganism. This has resulted in papers about *Legionella*, *Streptococcus*, *Haemophilus*, *Chlamydia*, and *Candida*.

Abstract

Genotyping of *Chlamydia trachomatis* is limited by the low sequence variation in the genome, and no adequate method is available for analysis of the spread of *chlamydial* infections in the community. We have developed a multilocus sequence typing (MLST) system based on five target regions and compared it with analysis of ompA, the single gene most extensively used for genotyping. Sequence determination of 16 reference strains, comprising all major serotypes, serotypes A to L3, showed that the number of genetic variants in the five separate target regions ranged from 8 to 16. The genetic variation in 47 clinical *C. trachomatis* isolates of representative serotypes (14 serotype D, 12 serotype E,

11 serotype G, and 10 serotype K strains) was analyzed; and the MLST system detected 32 variants, whereas 12 variants were detected by using ompA analysis. Specimens of the predominant serotype, serotype E, were differentiated into seven genotypes by MLST but into only two by ompA analysis. The MLST system was applied to *C. trachomatis* specimens from a population of men who have sex with men and was able to differentiate 10 specimens of one predominant ompA genotype G variant into four distinct MLST variants. Our MLST system can be used to discriminate *C. trachomatis* strains and can be applied to high-resolution molecular epidemiology. In the current presentation an update on current studies using this system will be provided.

Laura van Dommelen, MD

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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 will finish in July 2010. Her interest in sexually transmitted infections started by a research project on syphilis serology. She subsequently started a thesis project on STI testing, with *Chlamydia trachomatis* point-of-care tests as focus of this presentation.

Abstract

Objective Infection by *Chlamydia trachomatis* (CT) is the most prevalent sexually transmitted disease (STD) worldwide. The most frequently used diagnostic test for CT is a nucleic acid amplification test (NAAT), which is highly sensitive and specific. To further shorten time delay until diagnosis has been made, in order to prevent CT spread, the use of point of care (POC) tests could be the way forward. Three POC tests, Handilab-C, Biorapid *CHLAMYDIA* Ag test and QuickVue *Chlamydia* test, were evaluated regarding diagnostic performance in comparison with NAAT.

Design All women, above the age of 16 years old, consulting at an STD clinic between September 2007 and April 2008, were asked to participate. Women were asked to complete a short questionnaire and to collect 6 self-taken vaginal swabs (SVS). SVS 2 was used for NAAT and SVS 3 to 5 were randomized for the different POC tests. SVS 1 and 6 were used for determining quantitative CT load to validate the use of successively SVS. All POC tests were performed without knowledge of NAAT results. NAAT was used as the 'gold standard'.

Setting This study was performed at the STD clinic of the South Limburg Public Health Service and the Laboratory of Medical Microbiology of the University Hospital Maastricht, in The Netherlands

Participants 772 women were included.

Results CT prevalence was 11% in our population. Sensitivities of the Biorapid *CHLAMYDIA* Ag test, QuickVue *Chlamydia* and Handilab-C test were 17%, 27% and 12% respectively.

Conclusions Our results indicate that CT POC tests, due to the very low sensitivities, are not ready for widespread use.

Servaas Morré, PhD

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Curriculum vitae

Servaas A. Morré, PhD, who is working on *Chlamydia trachomatis* for 12 years, graduated at the VU University, the Netherlands, in Biochemistry and Molecular Biology in 1994. He worked on plat genetics at The Zaadunie (NL), on Drosophila genetics in Potugal (Universidade Do Porto, Laboratório de Genética Molecular), and on the on the processing of ribosomal RNAs in *Saccharomyces cerevisiae* (VU University, NL). 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, he extended *Chlamydial* research in the Department of Infectious Diseases, The City of Hope Medical Center, California, USA (Dr. Jim Ito and Dr. Joseph Lyons). The 1st of November 2001, he joined the Laboratory of Immunogenetics, VUmc. His research is focused on the immunogenetics of infectious diseases including HPV, sepsis, and periodontitis, but with still special attention to *Chlamydia trachomatis*. Together with Prof. Salvador Peña and Dr. Sander Ouburg, he organized for the 4th time the “Annual Amsterdam Chlamydia Meeting” in Dec 2007. In July 2005 he was a member on the ISSTDR Scientific Committee and organized amongst others the workshop “Immunogenetics of *Chlamydia trachomatis* Infections”. He was organizing Committee member of 6th Meeting of the European Society for *Chlamydia* Research, University of Aarhus, Aarhus, Denmark, July 1-4, 2008. He is coordinator of the International Chlamydial ICTI consortium and since 2007 he is Scientific Consortium Director, of the European Framework Programme 6 (FP6) “Contribution of molecular epidemiology and host-pathogen genomics to understand *Chlamydia trachomatis* disease (Acronym: EpiGenChlamydia). Since the first of January 2008 he is the head of the Laboratory of Immunogenetics.

Abstract

The Netherlands has a longstanding history for over 30 years on *Chlamydia trachomatis* studies performed in the field of epidemiology, detection and immunopathogenesis. The first PubMed Retrievable publication was in 1979 from J.L. Schuller and the first person defending his thesis on *C. trachomatis* was Kie

Amsterdam, 17 november 2009

H. Tjiam in 1987. In 2010 the current generation of Chlamydiologists will defend their thesis: Janneke den Hartog, Ingrid Rours and Vitaly Smelov. In the current presentation and overview will be give on the Dutch *C. trachomatis* lines performed in the past, current lines and groups, and our position in the world.

Hannelore Götz, MD, PhD

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Curriculum vitae

Dr Hannelore Götz studied medicine at the Free University of Amsterdam. She was clinically trained in surgery and gynaecology and obtained a certificate in tropical medicine. She worked 3 years in Namibia as District Medical Officer. Since 1995 she works in Public Health in the Netherlands, and is specialised in Infectious Disease Control and Epidemiology (EPIET). Since 2000 she works at the Municipal Public Health Service Rotterdam- Rijnmond at the department of infectious disease control. Her PhD in 2006 was about *Chlamydia* screening. She worked 2 years as regional consultant infectious disease control in South Holland. Currently her responsibilities are now focused on STI HIV control. She is project leader of CSI in Rotterdam and the Medical Head of the STI clinic Rotterdam.

Abstract

In April 2008, a selective systematic, internet-based *Chlamydia* Screening started among 16-29 year old citizens in Amsterdam, Rotterdam and South-Limburg. The programme aims to predict the effect of yearly screening on the prevalence of *Chlamydia trachomatis* (Ct) and related complications in the target population. Here we present results from the first year of screening.

The main outcome parameters, participation and Ct-positivity rate in the screened population, were compared among sub-groups within each screening region.

In the first round, 261,053 young people were invited in the 3 regions. 52,347 requested testing kits of which 78% were returned to the laboratories, hence finally 16% of invitees was tested for Ct. A total of 1733 *Chlamydia* infections was identified, equivalent to a 4.2% positivity rate.

* Women were more likely to participate than men (21% versus 10%). The positivity rate among women was higher than among men (4.4% versus 3.8%).

* Young people from 16 to 19 years old were less likely to participate but more likely to test positive than participants of 20 to 29 years old (participation 12% versus 17%, positivity 7.3% versus 3.8%).

The first round of the large scale *Chlamydia* screening has been successfully concluded. One out of six invitees participated and one in 25 participants was found positive for *Chlamydia*.

Symposium Organizer
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Curriculum vitae

Servaas A. Morré, PhD, who is working on *Chlamydia trachomatis* for almost 12 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 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 *Chlamydial* 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*, and also HIV (Prof. S. Danner & Dr. M. Agtmael)), periodontitis (collaboration with ACTA) and sepsis (collaboration AZM). Studies on Human Papilloma Virus (HPV) infections have been initiated together with Prof. C.J.L.M. Meijer in 2006. Together with Prof. Salvador Peña, he organised the “First Mini-symposium *Chlamydia trachomatis* Infections” in December 2004 and in December 2008 we organize already our fifth “Annual Amsterdam Chlamydia Meeting”. In July 2005 at the 16th Biennial meeting of the International Society for Sexually Transmitted Diseases Research (ISSTDR) 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 will be the organizer of the 7th Meeting of the European Society for *Chlamydia* Research in 2012 in Amsterdam.

Together with Tjaco Ossewaarde and Yvonne Pannekoek, he coordinates the Dutch *Chlamydia* Working Party. 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: EpiGenChlamydia)“ with 20 European, African and US groups. This consortium had his first meeting on 12 December 2007. As a partner he is participating in two other European FP6 programmes. Finally, 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 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 *Chlamyda trachomatis* were allocated to the VU University Medical Center.

Amsterdam, 17 november 2009

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

Table I: *PhD theses in the Netherlands*

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. Gijsen*	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.

Janneke E. den Hartog	Maastricht University
Ingrid Rours	Erasmus University Rotterdam
Caroline J. Bax	University of Leiden / Medical Center Haaglanden
Arnold Catsburg	VU University Amsterdam
Vitaly Smelov	St. Petersburg State Medical University, Russia and VU University Amsterdam
Koen Quint	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

Attendants:

Title	Last name	Surname	Affiliation	E-mail
Prof.	Bailey	Robin	LSHTM	Robin.bailey@lshtm.ac.uk
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Dr.	Bloembergen	Peter	Isala Clinics	p.bloembergen@isala.nl
Drs.	Bom	Reinier	Municipal Health Service Amsterdam	rbom@ggd.amsterdam.nl
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Dr.	Bruisten	Sylvia	Municipal Health Service Amsterdam	sbruisten@ggd.amsterdam.nl
	Bruyneel	Geert	Gen-Probe	geertb@gen-probe.nl
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	Coul, op de	Eline	RIVM, Bilthoven	eline.op.de.coul@rivm.nl
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	Diaz	Melissa	SOA – Aids Foundation	MDiaz@soaaids.nl
Ir.	Dickx	Veerle	University Ghent	Veerle.dickx@ugent.be
Drs.	Dommelen, van	Laura	azM	laura.van.dommelen@mumc.nl
Dr.	Dreesbach	Karen	Medac	k.dreesbach@medac.de
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	Lagae	Stephanie	University Ghent	stefanie.lagae@ugent.be
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Dr.	Morré	Servaas	VUmc, Amsterdam	samorretravel@yahoo.co.uk
	Nuijten	Piet	Nobilon	piet.nuijten@sp.nobilon.com
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	Oud	Elise	VUmc, Amsterdam	elise_oud@hotmail.com
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Drs.	Quint	Koen	LUmc, Leiden	k.d.quint@gmail.com
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Title	Last name	Surname	Affiliation	E-mail
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	Smelov	Vitaly	St. Petersburg State University	vitsmelov@yahoo.com
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	Vermeulen	Hans	Roche Diagnositics	
Ir.	Verminnen	Kristel	University Ghent	Kristel.Verminnen@UGent.be
	Verweij	Stephan	VUmc, Amsterdam	s.p.verweij@vumc.nl
Dr.	Vries, de	Henry	AMC & GG&GD A'dam	h.j.devries@amc.uva.nl
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The Abbott m2000 system is a flexible automated platform for NA extraction and RT PCR for the detection of C. trachomatis and N. gonorrhoeae and other microbial targets. The m2000 system can run Abbott RT IVD CE-marked assays as well as home-brew assays.

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Future STI / *Chlamydia* Meetings

11th IUSTI World Meeting
9-12 November 2009, Cape Town, South Africa
www.iusti.org

12th International Symposium on Human *Chlamydia* Infections
June 20th – 25th, 2010, Fuchsl, Austria

7th Annual Amsterdam *Chlamydia* Meeting
December 2010, Amsterdam The Netherlands

19th ISSTDR Meeting
July 10th – 13th, 2011, Québec, Canada
www.isstdrquebec2011.com / www.isstdr.org

12th IUSTI World Meeting
November 2nd – 5th 2011, New Delhi, India
www.iusti.org

5th Chlamydia Basic Research Society (CBRS)
2011, (Los Angeles) USA

7th European *Chlamydia trachomatis* meeting
2012, Amsterdam, The Netherlands

20th ISSTDR Meeting
2013, Vienna, Austria
www.isstdr.org

Amsterdam, 17 november 2009

Announcement

7th
Annual Amsterdam *Chlamydia* Meeting

December 2010

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

We hope to welcome you all in 2010

Notes

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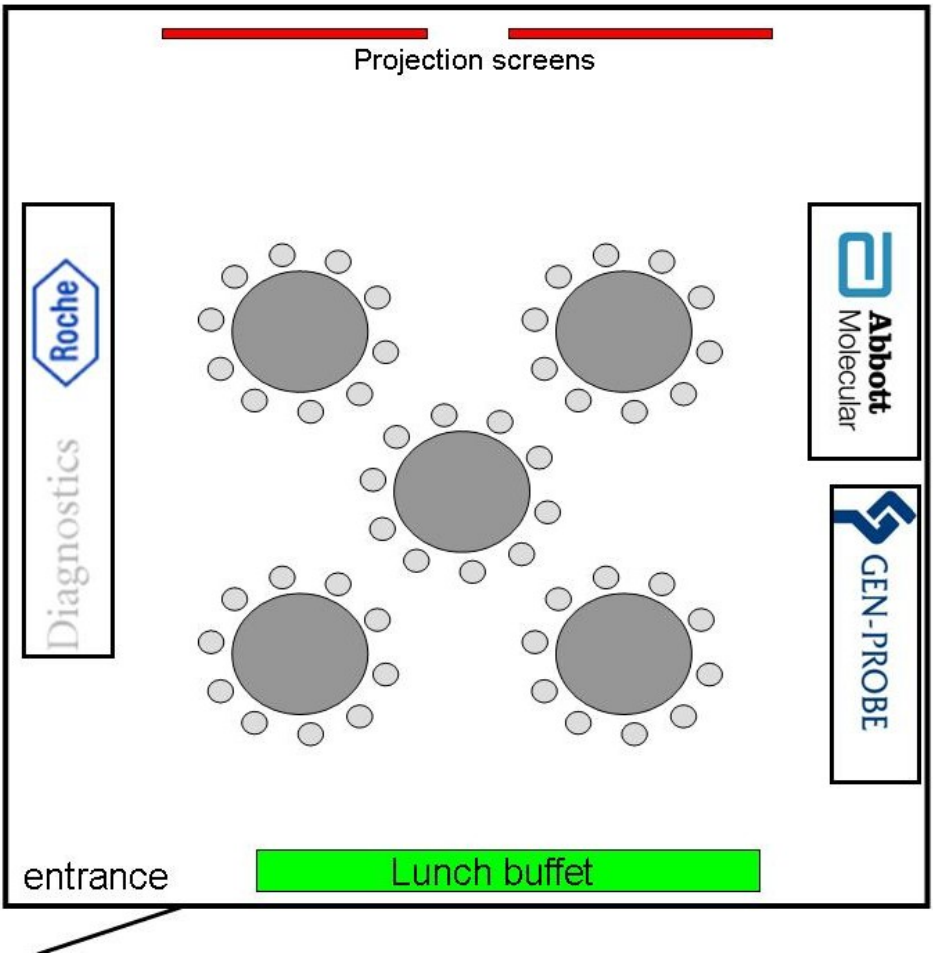
Floor plan (1st Floor, rooms 1+2)



Amsterdam, 17 november 2009

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Lay out & design, odd jobs:
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The organisers are indebted to Amine Karimi, Roel Heijmans, Stephan P. Verweij, and Elise V. Oud for their valuable assistance.

Accreditation is requested for this symposium from:

- *The Dutch Society for Medical Microbiology (NVMM)*
- *The Dutch Society of Obstetrics and Gynaecology (NVOG)*
- *The Dutch Society for Dermatology and Venerology (NVDV)*
- *The Dutch Association of Specialists for Gastroenterology-Hepatology (MDL)*
- *Accreditation Bureau Social Medicine (AbSg)*

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