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Can teaching young people lead to increased knowledge about chlamydia and give them knowledge about what protects against chlamydia. Intervention study with control group.

Tina Louise Schmock, Hygiene nurse, MMS

Fredensborg Municipality

Abstract

Background: Chlamydia Infection is one of the most common bacterial sexually transmitted infections in Denmark. In the period 2018 - 2022, there has been an increased prevalence of Chlamydia among young people aged 15-29 years.

Research question: Assess whether educating young people aged 16-20 years can lead to increased knowledge about chlamydia and provide them with knowledge about how to protect themselves against chlamydia.

Materials and methods: This was an intervention study with a control group, and a total of 329 questionnaires were completed. The intervention group and the control group each completed two identical questionnaires.

The intervention group (n=87) completed the first questionnaire before the intervention. After the intervention, the intervention group completed the second questionnaire. The control group (n=78) completed the first questionnaire followed by no intervention. Two weeks later, they completed the second questionnaire (n=77).

Results: Out of the total number of young people who answered the first questionnaire (n=165), 40% had their first sexual debut before the age of 16. Further 70.9% of the young people had previously received sex education in primary school. The young people in the intervention group increased their knowledge of where chlamydia can infect tissue by 132.4% after the education, while the control group remained unchanged.

Conclusion: The pilot study has shown that intervention in the form of classroom education can increase young people's knowledge about chlamydia and what protects them against chlamydia. Therefore, it is important to prioritize and optimize education about sexually transmitted infections before young people become sexually active.

Implications: The results of the study should be considered among policy makers, because new thinking in the primary care sector will be needed to reduce chlamydia transmission.

Costs and effectiveness associated with contact tracings due to unexpected findings of methicillin-resistant *Staphylococcus aureus* in Oslo University Hospital

Abstract

Background: Methicillin-resistant Staphylococcus aureus (MRSA) is a challenge in most healthcare-settings. Due to strict MRSA-policy, the prevalence of MRSA has been low in Scandinavian countries and the Netherlands. In Norway, the national guidelines are comprehensive with extensive contact tracings in all hospitals when unexpected findings occur.

Aim: The aim of this study was to evaluate the contact tracings performed in Oslo University Hospital, and to estimate the costs and effectiveness of this procedure.

Materials and methods: Data and results from contact tracings performed between 2016 and 2019 were collected. MRSA-screening cultures of exposed cases and the estimated time consumed by nurses involved were included as costs.

Results: Altogether, 443 contact tracings were performed during the study period with 6188 exposed cases. Including 5859 healthcare workers (HCW) and 329 patients or parents staying with their children, categorized as patients. 11 secondary cases were detected, of these only 3 among HCW. The risk of being colonized was 0.05% for a HCW and 2.4% for patients. Seven secondary cases occurred due to an outbreak in a Newborn Intensive Care Unit, all of them patients or parents.
Estimated costs for all contact tracings were EUR 258,882. Costs per secondary finding were EUR 23,535, and costs for detecting one secondary case among HCW were EUR 80,938.

Conclusions: We found this procedure time consuming and resource-intensive, with only a small number of secondary cases detected, and suggest the current guideline should be revised.

Development of rapid culture-independent mass spectrometry-based proteomics methods for improving urinary tract infection diagnostics

Roger Karlsson[1,2,3,*], Beatriz Pineiro Iglesias[1], Leonarda Achá Alarcón[2,3], Sali El-Ali[1], Niklas Eklo[1], Gelio Alves[4], Yi-Kuo Yu[4], Edward R.B. Moore[1,2,3,5]

[1]Clinical microbiology, Sahlgrenska University Hospital, Sweden, [2]Department of infectious diseases, Department of Biomedicine, Sahlgrenska Academy, University of Gothenburg, Sweden, [3]Center for Antibiotic Resistance Research (CARe) at the University of Gothenburg, Sweden, [4]National Center for Biotechnology Information, National Library of Medicine, National Institutes of Health, Bethesda, Maryland 20894, United States, [5]Culture Collection of the University of Gothenburg (CCUG), Sweden *Poster presenter

Urinary tract infection (UTI) is a very common infection, which can also lead to serious conditions, such as sepsis. Globally, sepsis is a leading cause of death in hospitalized patients and rapid diagnosis is crucial in order to provide proper treatment. Traditional methods for microbial identification at clinical microbiology laboratories involve the use of culturing clinical samples and isolating pure cultures, processes that take time. This delay often leads to prescription of broad-spectrum antibiotics as treatment of the infection. The most common pathogen causing UTIs is *Escherichia coli,* which is prone to acquire antibiotic resistances, such as Extended Spectrum βeta Lactamases (ESBL).

To improve diagnostics and reduce unnecessary use of antibiotics, a rapid, culture-independent method called proteotyping has been developed, relying on the identification of biomarkers using nano-liquid chromatography in combination with tandem mass spectrometry (LC-MS/MS). In this study, proteotyping was employed to analyze patient urine samples without prior culture. Ten urine samples, where ESBL *E.coli* was identified as the cause of infection, were processed by simple and rapid differential centrifugation steps, followed by a short enzymatic digestion of proteins into peptides. The LC-MS/MS data was analyzed using a bioinformatic pipeline called Microorganism classification and Identification (MiCId) and a curated database for species identification, developed at NIH, USA. Additionally, the antibiotic resistance traits were identified through CARD and ResFinder databases.

Biomarker peptides unique for *Escherichia coli* was identified and the most predominant resistances found by the traditional phenotypic resistance profiling was confirmed at peptide/protein level. In conclusion, direct, culture-independent bacterial identification and antibiotic profiling of patient urine samples was possible using the proteotyping workflow.

Proteotyping of *Clostridioides difficile* and detecting expression of toxins using mass spectometry

Beatriz Pineiro Iglesias[1], Lucas Lundbäck[2], Edward R.B. Moore[1,2,3,4], Gelio Alves[5], Yi-Kuo Yu[5], Roger Karlsson[1,2,3]

[1]Clinical microbiology, Sahlgrenska University Hospital, Gothenburg, Sweden, [2]Department of infectious diseases, Institute of Biomedicine, Sahlgrenska Academy, University of Gothenburg, Sweden, [3]Center for Antibiotic Resistance Research (CARe) at University of Gothenburg, Sweden, [4]Culture Collection of the University of Gothenburg (CCUG), Sweden, [5]National Center for Biotechnology Information, National Library of Medicine, National Institutes of Health, Bethesda, United States

Clostridioides difficile is a bacterial species often associated with antibiotic treatment. Many strains produce toxins, which can cause disease. Toxins are also relevant in diagnostics as well as epidemiology. Some *C. difficile* variants are considered more virulent, and there are multiple approaches to perform typing of *C. difficile*.

Our aim was to investigate whether proteomics and mass spectrometry (proteotyping) can be used for typing of *C. difficile* and hence to distinguish between separate outbreaks, caused by different strains. Furthermore, we wanted to investigate whether proteotyping can be used to detect expression of *C. difficile* toxins.

The experimental design included six *C. difficile* strains, three from PCR ribotype 001 and three strains from PCR ribotype 014. Samples were then analyzed using liquid chromatography tandem mass spectrometry, resulting in the identification of peptides and proteins. Around 25000 peptides and 2300 proteins were identified in the *C. difficile* strains. Analysis using the MiCId bioinformatics pipeline, showed the typing capability and possibility to show similarities and dissimilarities of strains in outbreak clusters. Furthermore, the toxins A and B, as well as proteins related to antimicrobial resistance, were detected.

In conclusion, the results indicate that there is a lot of potential in further studying *C. difficile* using proteomics and mass spectrometry. The proteotyping approach not only allows for identification at the species and strain level, but also detection of expressed phenotypic antimicrobial resistance and traits of virulence, including the expression of toxins. Proteotyping thus provides the potential for highly sensitive and rapid diagnoses of infectious bacteria, without the time-consuming cultivation steps, which could have a significant impact on the overall treatment of infectious diseases.

Title:
Spontaneus loss of MRSA carriage in children under two years of age, without the use of topical or systemic eradication treatment

Student:
Judit Christensen 2024
Institutionen för biomedicin, Sahlgranska akademi, Göteborgs universitet

Supervisor:
Dennis Back Holmgaard consultant, MD, Ph.D. Klinisk Mikrobiologisk afd. Region Sjælland Denmark

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Background Epidemiological data for Denmark in the period 2007-2022 from the Statens Serum Institut shows that children below the age of 1 year pose the second highest risk of contracting MRSA (Methicillin-resistant *Staphylococcus aureus*) during hospitalization in a hospital environment. In terms of risk they are exceeded only by hospitalized patients on a medical ward. During the period 2007-2022, 679 children and 64 health workers were infected with MRSA in neonatal wards in Denmark.
In the Danish recommendations, treatment with topical and systemic antibiotics is omitted in asymptomatic children below the age of 2 years, as these are presumed to become spontaneously MRSA-free before the age of 2. However, evidence for this practice is sparse**.**

Aim To investigate whether children diagnosed with MRSA spontaneously lose MRSA carrier status before they reach the age of 2 years and whether Clonal Complex (CC) or pharyngeal colonization plays a role in the length of MRSA carrier status**.**

Method Retrospective register study of 119 children in the period 2018-2022. Data are presented in the form of tables and diagrams and described descriptively**.**

Results Data for achieving spontaneous carrier freedom show a success rate of 93.1%. There was too little data to demonstrate differences at the Clonal Complex level. Pharyngeal colonization was not associated with reduced loss of carrier status at 24 months**.**

Conclusions: The study supports the current Danish national MRSA guidelines, which recommend not to intervene on asymptomatic MRSA carriers in the first two years of life. In the study, it has not been possible to assess the influence of the CC type on the loss of carrier status in the first two years of life, as 16 different CC types were represented in the study. There is no statistical significance in relation to whether the MRSA carrier state was localized to the throat or another primary location. In children, pharyngeal carrier state is thus not associated with prolonged carrier state**.**

Surveillance of enteroviruses

Marianela Patzi Churqui[1,2*] and H Wang [1,2], T tunovic[2], L Enache[3], M Andersson[1,2], K Rembeck[2], K Nyström [1,2], M Lindh [1,2], H Norder [1,2]

[1] University of Gothenburg, Sahlgrenska Academy, Institute of Biomedicine, Department of Infectious Diseases, Gothenburg, Sweden, author's affiliation, City, Country, [2] Sahlgrenska University Hospital, Department of Clinical Microbiology, Västra Götaland Region, Gothenburg, Sweden,[3] Gryaab AB, Gothenburg, Sweden

Enteroviruses (EVs) belong to the *Picornaviridae* family. The genus Enteroviruses comprises about 12 species of Enterovirus (A–L), and 3 species of Rhinovirus (A–C), which are transmitted by respiratory and fecal-oral routes. EV infections in humans vary from mild respiratory and gastrointestinal illnesses to severe diseases, such as meningitis, myocarditis, and paralysis. In temperate climates EV infections peak in late summer and early fall, primarily among children and young adults. EVs are secreted in feces and in the nasopharynx and those causing neurological disorders, as meningitis, encephalitis and paralysis, are also present in cerebrospinal fluid (CSF). This study aims to identify and compare the circulating EVs in wastewater and in clinical samples to investigate if outbreaks can be predicted. Patient samples, such as feces, CSF, nasopharyngeal secretions or blood were analyzed for detection for EV, and the virus in positive samples were typed by sequencing partial VP1 and 5′-UTR regions. Additionally, weekly wastewater samples with increased EV RNA were analyzed by NGS. Preliminary results from the surveillance of viruses during the pandemic indicate that in 2022, EV peaked in wastewater on weeks 11, 22, 28, and 33. Sequencing the virus in of the wastewater and clinical samples from the same period showed the spread of certain types such as CV-A6, CV-A16, E30 and CV-B5. Additionally, the NGS analysis of some wastewater samples revealed the presence of additional circulating EV types, including E7, EV-D68, EV-C104, EV-C116, which have potential to cause serious and widespread outbreaks. Surveillance of EV in wastewater relate to the findings in patient samples and can help in fill knowledge gaps in the dynamics of seasonality of EVs and potentially predict future outbreaks.

The influence of relative humidity on the sterility of sterile pouches

Tove Windh, 2024

Department of Biomedicine, Sahlgrenska Academy, University of Gothenburg

Supervisor: Anders Edebo, MD PhD

Background

In healthcare, sterilized medical equipment, often packaged in sterile pouches, is used. High relative humidity can compromise the microbiological barrier of these pouches, leading to loss of sterility. When high relative humidity is detected in surgical and sterile units, resterilization of medical equipment becomes necessary, incurring costs, time, and environmental concerns.

Research question

Does *S. epidermidis* penetrate sterile pouches during a 24-hour incubation at 60%, 85% or 90% relative humidity?

Materials and methods

Method development involved test to explore different time intervals, temperatures, relative humidity levels, as well as assessing different bacterial strains and concentrations. The aim was to establish a standardized method for contaminating intervention pouches. Following method development, a series of tests at 60%, 85%, and 90% relative humidity levels for one day were conducted to detect any differences in growth on the inside of sterile pouches between different humidity levels. The intervention group consisted of sterile pouches contaminated with *S. epidermidis* externally. The control group consisted of sterile pouches without contamination, and bags with thumbprints were also included. The experiment was replicated four times.

Results and Conclusion

In total, samples from inside 47 sterile pouches in the intervention group, 24 sterile pouches in the control group avd 12 sterile pouches with thumbprints from the different humidites were analyzed. Bacterial growth was observed inside sterile pouches on seven out of occasions. Bacterial growth occurred across all groups and humidity levels, with only two instances involving *S. epidermidis* av 60-61% relative humidity and 85% relative humidity. Drawing the conclusion that *S. epidermidis* penetrates sterile pouches at 60%, 85% and 90% relative

humidity cannot be made at this point. Occational findings of unrelated species inside the sterile pouches likely indicate contamination during some part of the experiment.