24. DOSCH-Symposium

15. BIS 17. MAI 2017

VELDEN / KÄRNTEN

KURZFASSUNGEN DER BEITRÄGE

ÖSTERR. GESELLSCHAFT FÜR HYGIENE, MIKROBIOLOGIE UND PRÄVENTIVMEDIZIN
c/o MAW, 1010 WIEN, FREYUNG 6

INHALTSVERZEICHNIS

ORAL PRESENTATION

Aktion Saubere Hände Tirol
Theresa Geley 4

Benefits and risks of remnant "active ingredients" in alcohol-based handrubs
Günter Kampf 5

Antibiotikaverbrauch in der Humanmedizin, Österreich 2016
Stefanie Kirchner, Franz Allerberger 6

Efficacy of two antiseptic regimens on skin colonization of insertion sites for two different catheter types: a randomized, clinical trial
Juergen Thomas Lutz, Isabel Victoria Diener, Kerstin Freiberg, Robert Zillmann, Kija Shah-Hosseini, Harald Seifert, Bettina Berger-Schreck, Hilmar Wisplinghoff 7

AHALT-PPS: Point prevalence survey on healthcare-associated infections and antimicrobial use in Austrian long-term care facilities
Luigi Segagni Lusignani, Sneschana Neschkova, Magda Diab-Elschahawi, Elisabeth Presterl 8

Tod durch Totalprothese - Gesundheitsrisiko Prothesenplaque
Patricia-Anca Steinmassl, Otto Steinmassl, Herbert Dumfahrt, Ingrid Grunert 9

POSTER

Antimicrobial activity of octenidine against multidrug-resistant Gram-negative pathogens
Rocio Alvarez Marin; Marta Aires de Sousa; Nicolas Kieffer; Patrice Nordmann and Laurent Poirel 11

Measuring alcohol-based hand rub volume used by healthcare workers in practice
Ojan Assadian, Curtis J. Donskey, Trina F. Zabarsky, Albert E. Parker 12

Verhinderung der bakteriellen Einsaat in das Operationsgebiet durch antimikrobielle chirurgische Handschuhe
Ojan Assadian, Helga Paula, Romana Klasinc, Edda Skrinjar, Afshin Assadian 13

Mikrobielle Belastung von langärmeligen Arztkitteln – eine Stichprobe
Michael Berktold, Guido Hinterberger, Astrid Mayr, Dorothea Orth-Höller und Cornelia Lass-Flörl 14

Self-disinfecting surfaces and the activity against *Staphyloccocus aureus* ATCC 6538 under real life conditions
Christina Brühwasser, Holger Heinrich, Cornelia Lass-Flörl, Astrid Mayr 15

Octenidine against *Staphylococcus aureus* epidemic clones: an alternative strategy
Teresa Conceição, Hermínia de Lencastre, Marta Aires-de-Sousa 16

Inaktivierung von Norovirus (MNV-1) auf antimikrobiellen Oberflächen
Andreas Fritz, Wilfried Posch, Doris Wilflingseder und Cornelia Lass-Flörl 17

Survey in Austrian Healthcare Institutions: Systematic evaluation of medical devices on their compatibility to disinfectants
Hartl Christopher, Krejci Marion, Bergmann Andrea, Spoljaric-Lukacic Lidija, Laupenmühlen Fiona, Eichinger Daniela, Freundlinger Thomas, Colbert Brigitta, Hansal Selma, Ettenberger-Bornberg Gabrielle 18

Changing the epidemiology of Verotoxin-producing *E. coli* (VTEC) in Austria by introducing a new molecular based diagnostic procedure in one single laboratory in 2016
Markus Hell, Alexandra Wojna, Elisabeth Mustafa-Korninger, Christiane Schausberger, Franz Allerberger, Burkhard Springer, Sabine Schlager 19

Epidemiology of urinary tract infections in HIV positive patients at a tertiary care hospital in Central Europe (2011 - 2016)
Romana Klasinc, Armin Rieger, Elisabeth Presterl, Thomas Wrba, Magda Diab-Elschahawi 20

Infection Prevention and Control 4.0?
Walter Koller und Klaus-Peter Adlassnig 21

Masernelimination in Europa- wie kann das Ziel bis 2020 erreicht werden
Kreidl Peter, Decristoforo Petra, Luckner-Hornischer Anita, Iris Broksch, Würzner Reinhard, Daniela Schmid 22

Detection of bloodstream pathogens by real-time PCR compared to standard conventional blood culture in patients with sepsis
Bettina Kuppelwieser, Ludwig Knabl, Dorothea Orth-Höller 23

Activity of N-chlorotaurine against *Staphylococcus aureus* and Pseudomonas aeruginosa in a long-term in-vitro biofilm model
Markus Nagl, Andrea Windisch, Débora Coraca-Huber 24

Platelet activation in the course of nosocomial *Candida*: in vivo study using a mouse model and patient samples
Günter Rambach, Claudia Eberl, Ilse D. Jacobsen, Christine Dunker, Magdalena Hagleitner, Cornelia Lass-Flörl, Romuald Bellmann, Ingo Lorenz, Mathias Ströhle, Cornelia Speth 25

Nachweis der desinfizierenden Wirkung von Waschverfahren (Detection of the disinfecting effect of washing processes)
Arno Sorger, Sladana Jusic 26

Prevention of fungal infection by antimycotic drugs can modulate activity and function of human platelets
Cornelia Speth, Bianca Salzgeber, Magdalena Hagleitner, Martin Hermann, Cornelia Lass-Flörl, Günter Rambach 27

Analysis of resistance genes in *Candida albicans* and *Candida glabrata* using Next Generation Sequencing
Kathrin Spettel, Wolfgang Barousch, Athanasios Makristathis, Alexander Hirschl, Marion Nehr, Brigitte Selitsch, Michaela Lackner, Peter-Michael Rath, Jörg Steinmann, Birgit Willinger 28

Comparison of microbiological laboratory methods for the diagnosis of dermatophytes
Alexander Tolios, Erika Wimmer, Iris Zeller, Brigitte Selitsch, Erika Steiner, Wolfgang Barousch, Birgit Willinger 29

Comparison of *Fusarium* Diagnosis Methods – Evaluation of Cultivation, Sequencing and Proteomic Analysis
Alexander Tolios, Iris Zeller, Marion Nehr, Brigitte Selitsch, Kathrin Spettel, Wolfgang Barousch, Birgit Willinger 30

A suspected case of Lassa fever: a bundle of problems
Elisabeth Ullrich, Christoph Koidl, Franz F. Reinthaler, Marianne Wassermann-Neuhold, Robert Krause, Andrea .J. Grisold 31

Determination of reversion frequency of clinical thymidine-dependent Small-Colony Variants of *Staphylococcus aureus*
Gernot Zarfel, Clemens Kittinger, Bettina Folli, Lilian Masoud-Landgraf, Daniela Toplitsch 32

ORAL PRESENTATION

Aktion Saubere Hände Tirol

Theresa Geley

Tiroler Gesundheitsfonds

Der Tiroler Gesundheitsfonds startete vor 5 Jahren gemeinsam mit dem Institut für Hygiene der Medizinischen Universität Innsbruck und der Tiroler Gebietskrankenkasse das Projekt „Aktion Saubere Hände Tirol“. Da in Österreich kein derartiges Programm umgesetzt ist, beteiligte sich Tirol an der deutschen Aktion dieser Kampagne der Weltgesundheitsorganisation WHO. Ziel des Projektes ist es, die Compliance für Händehygiene der Akteure des Tiroler Gesundheitswesens zu verbessern und dadurch die Patientensicherheit in den Gesundheitseinrichtungen zu erhöhen. Der Tiroler Gesundheitsfonds finanziert im Rahmen des Projektes die im Rahmen des Kooperationsvertrages mit der deutschen Kampagnenleitung anfallende Kosten, Kosten für tirolspezifische Drucksorten und die Organisation und Ausstattung der Erfahrungsaustausche. Im Projektzeitraum wurden konsekutiv alle bettenführenden Krankenanstalten, alle Alten- und Pflegeheime und das Rettungswesen in das Projekt integriert. Weiters wurde in Zusammenarbeit mit der Landessanitätsdirektion ein Rahmenhygieneplan für Pflegeheime ausgearbeitet und alle Pflegeheime auf diesen und natürlich auch auf die Umsetzung einer richtigen Händehygiene geschult. Die Teilnahme der Einrichtungen an der AKTION Saubere Hände erfolgt auf freiwilliger Basis. Mit Stand Jänner 2017 waren alle 8 Tiroler Akutkrankenhäuser und 8 Tiroler Pflegeheime als Mitglieder der Aktion Saubere Hände in Deutschland registriert und nehmen an der Erfassung des Desinfektionsmittelverbrauches als Surrogatparameter für die Überwachung der Händehygiene teil. Seit dem Projektstart im Jänner 2012 finden jährlich in Innsbruck Erfahrungsaustausche für die teilnehmenden Einrichtungen statt, die seit 2016 als gemeinsame Veranstaltung von Krankenanstalten und Pflegeheimen abgehalten werden. Diese Veranstaltungen wurden von den Teilnehmern als sehr befruchtend wahrgenommen und unterstützen die interdisziplinäre, sektorenübergreifende Zusammenarbeit.

Benefits and risks of remnant “active ingredients” in alcohol-based handrubs

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Some alcohol-based handrubs contain additional “active ingredients” such as chlorhexidin (CHG) or mecetronium etilsulfate (MES).The 2016 hand hygiene KRINKO recommendation, however, says that these formulations cannot be recommended for hygienic hand disinfection because they do not improve the efficacy but have a risk for side effects. The use of these hand rubs for surgical hand disinfection is an unsettled issue because the overall evaluation of benefits and risks is not clear. These ingredients may slow down the re-growth under the surgical glove. EN 12791 allows measuring such a sustained efficacy. In a recent review all published efficacy data for surgical hand disinfection with such alcohol-based hand rubs were analyzed. Two data sets were found for handrubs with 0.5 or 1% CHG applied for 3 min, no sustained efficacy was found. 23 datasets were found for handrubs with 0.2% MES. No sustained efficacy was found when applied for 1 min (1 dataset), 1.5 min as currently recommended (14 datasets) and 2 min (1 dataset). When applied for 3 min a sustained efficacy was detected in 3 out of 7 datasets. A new study provides evidence that identical formulations with and without 0.2% MES have a similar efficacy in surgical hand disinfection after 3 h. With a lack of sustained efficacy the possible risks come into the focus. Use of CHG is associated with a risk of acquired bacterial resistance especially when it is used for various types of application (hand hygiene, skin antisepsis, oral rinse, body wash). Even anaphylactic reactions have been reported. MES is a type of detergent and may contribute to a mild skin irritation. If there is no measurable benefit such as a sustained efficacy in surgical hand disinfection but some associated risks it seems to be adequate to critically review the current use of these handrubs.

Antibiotikaverbrauch in der Humanmedizin, Österreich 2016

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Der kausale Zusammenhang von Antibiotikaeinsatz und Resistenzentstehung bei Bakterien kann sowohl für Infektionen bei PatientInnen niedergelassener ÄrztInnen als auch für im Krankenhaus erworbene Infektionen belegt werden. Koordinierte Maßnahmen zur Hintanhaltung der Verbreitung antimikrobieller Resistenzen erfordern Surveillance-Systeme. Nur so ist es möglich, zu beurteilen, wie lokale und globale Resistenzsituationen auf einen geänderten Antibiotikaeinsatz und neue Infektionskontrollmaßnahmen reagieren. Die AGES (Österreichische Agentur für Gesundheit und Ernährungssicherheit GmbH) erhebt, rückwirkend seit 2010, den Verbrauch systemischer Antibiotika in Österreich. Datengrundlage bildeten Absatzzahlen des pharmazeutischen Großhandels. Während in der tierischen Lebensmittelproduktion ein Rückgang des Antibiotikaeinsatzes belegbar ist (Rückgang von 50,9 Tonnen im Jahr 2011 auf 45,7 Tonnen im Jahr 2015; Zahlen für 2016 noch nicht verfügbar) ist die Menge des Antibiotikaverbrauchs in der Humanmedizin [alle unter den ATC-Code "J01 zur systemischen Applikation" fallenden Substanzen] bislang nicht gesunken (Gesamtverbrauch 2011: 66,9 t; 2016: 72 t). Mit einem Antibiotikaverbrauch (in kg Wirksubstanz) von 72.020 kg fand sich im Jahr 2016 ein neuer Spitzenwert: 67% vom Gesamtwert gelangten im niedergelassenen Bereich zur Anwendung, 33% im stationären Bereich). Im Oktober 2015 wurde zudem vom Nationalrat angeregt, den Antibiotika-Verbrauch in Österreichs Krankenanstalten zu erheben. Die ÖGACH (Österreichische Gesellschaft für antimikrobielle Chemotherapie) hat in Zusammenarbeit mit der AGES und dem Robert Koch Institut ein Antibiotika-Verbrauch-Surveillance-System entwickelt, das es individuellen Krankenhäusern gestattet, im Rahmen ihrer Antibiotika-Stewardship-Bemühungen ihren Antibiotikaverbrauch zu erfassen, auf Stationsebene auszuwerten und sich anonymisiert mit gleichartigen Krankenanstalten zu vergleichen [https://www.ages.at/themen/ages-schwerpunkte/antibiotika-resistenzen/antibiotikaverbrauch-im-krankenhaus/]. Gemäß EU-weiter Vorgaben zur Eindämmung von Antibiotikaresistenzen wurde mit diesem Pilotprojekt ein österreichweit einheitliches Antibiotikaverbrauch-Meldeverfahren in Krankenanstalten etabliert, um statistisch abgesicherte Grundlagen für das Management des sorgfältigen Einsatzes von Antibiotika zur Verfügung zu haben. Derzeit nehmen an dieser Qualitäts-Sicherungsmaßnahme zwölf Krankenanstalten teil.

Efficacy of two antiseptic regimens on skin colonization of insertion sites for two different catheter types: a randomized, clinical trial

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Background: Catheter associated infections in central venous (CVC) and epidural catheters (EDC) are associated with severe complications. The extent of bacterial colonization around the insertion site is crucial for (systemic) infection. The use of a dermal antiseptic with remanent effect may be an option to reduce colonization and thus infections. In this prospective, randomized and blinded study we compared an octenidine containing skin antiseptic to a conventional, frequently used alcohol-based product. Primary outcome was defined as the amount of re-colonization of the skin (CFU/swab) as an indicator for bacterial contamination of the catheter.

Material & methods: After approval of the local ethics committee (Ärztekammer Nordrhein Nr. 2014222) 216 patients were enrolled in the study from July to December 2015 and three standardized skin swabs were conducted on each patient, respectively: 1) untreated skin; 2) after disinfection and catheter insertion; 3) 48h after catheter insertion. CFUs were determined on agar plates after 48h of incubation and logarithmized to detect any difference between #2 und #3.

Results: The bacterial recolonization of the skin, disinfected by using the octenidine containing alcoholic product, was significantly reduced after 48 hours (log 0.72; n=59; $p<0.001$) compared to the standard alcohol-based antiseptic (log 1.97; n=51). 106 patients were excluded from analysis due to manipulation, partial or complete loss of the sterile dressing on the insertion site during the study period, as defined in the protocol.

Conclusion: In our study, the rate of re-colonization was significantly reduced in the study group (octenidine). Based on present data, skin antiseptics with remanent actives should be considered for skin preparation prior to the insertion of central venous or epidural catheters. Furthermore, due to the high number of excluded patients, we recognized an unexpected, inadequate use of dressings to protect the catheter insertion sites, which had been optimized thereafter.

AHALT-PPS: Point prevalence survey on healthcare-associated infections and antimicrobial use in Austrian long-term care facilities

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Das „European Centre for Disease Prevention and Control (ECDC)“ hat 2016/2017 in 33 europäischen Staaten die dritte Punkt-Prävalenz-Studie organisiert, um nosokomiale Infektionen (NI), den Verbrauch von Antibiotika und Strukturindikatoren für Hygiene in Langzeitpflegeeinrichtungen anhand von standardisierten Definitionen zu erfassen. Die Erhebung wurde in ganz Österreich im Zeitraum von drei Monaten (September-November 2016) durchgeführt. Es nahmen insgesamt 14 Langzeitpflegeeinrichtungen mit einer durchschnittlichen Bettenanzahl von 212 Betten teil. In die AHALT-Studie wurden 2,625 Bewohnerinnen und Bewohner eingeschlossen. Davon waren 28% (748/2,625) über 85 Jahre, 43% (1,122/2,625) benötigten einen Rollstuhl oder waren bettlägerig, 50% waren desorientiert und 52% (1,370/2,625) waren inkontinent. Die Gesamtprävalenz der NI war 4,9% (130 NI in 128 Patienten). Die häufigsten NI waren Hautinfektionen (30%), Harnwegsinfektionen (17%), Pneumonien und andere tiefe Atemwegserkrankungen (16%). Überwiegend wurde Escherichia coli isoliert (6,2%), allerdings standen in 81,5% der Fälle keine mikrobiologischen Daten zur Verfügung. Fünfundsiebzig (2,8%) der erfassten Einwohner haben Antibiotika erhalten, davon wurden 90% zu therapeutischen Zwecken verschrieben. Am häufigsten erhielten die Bewohnerinnen und Bewohner mindestens ein systemisches Antibiotikum als Therapie für folgende Indikation: Harnwegsinfektion (32,4%), Atemwegsinfektion (25,4%) und Haut-/Wundinfektion (19,7%). Die am häufigsten verschriebenen Antibiotika waren Penicilline plus β-Lactamase-Inhibitoren (39%) und Fluorchinolone (22%). Infektionskontrollmaßnahmen kamen in mehr als 80% der Langzeitpflegeeinrichtungen zur Anwendung. AHALT-PPS bot die Möglichkeit, sich innerhalb von kurzer Zeit und mit verhältnismäßig wenig Aufwand einen Überblick über die aktuelle Situation der Pflege- und therapieassoziierten Infektionen und Antibiotikaeinsatz in österreichischen Langzeitpflegeeinrichtungen zu verschaffen. Um die reale Situation möglichst genau abzubilden, besteht die Notwendigkeit, diese Erhebungen anhand von gleichen Protokollen regelmäßig zu wiederholen.

Tod durch Totalprothese - Gesundheitsrisiko Prothesenplaque

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Ein Großteil der älteren Menschen trägt herausnehmbare Prothesen. Obwohl die Prothesen herausnehmbar sind, ist ein oft nicht unerheblicher Teil der Prothesenoberflächen von Biofilm besiedelt. Die erste Tiroler Mundgesundheitsuntersuchung unter Altenheimbewohnern, beispielsweise, hat gezeigt, dass durchschnittlich 43,9% der Prothesenoberflächen von Plaque besiedelt sind. Dieser Biofilm kann bekanntermaßen lokale Erkrankungen der Mundhöhle (Stomatitis, Karies, Parodontitis) begünstigen. Weniger bekannt ist aber, dass Prothesenplaque auch gravierende Auswirkungen auf die Allgemeingesundheit, vor Allem bei älteren oder immunkompromittierten Patienten, haben kann. So konnte besonders die Aspiration von Keimen aus dem Prothesenbiofilm eindeutig mit dem Auftreten von Pneumonien, einer der häufigsten Todesursachen bei geriatrischen Patienten, in Zusammenhang gebracht werden.

Studien haben gezeigt, dass die Reduktion des mikrobiellen Biofilms auf herausnehmbaren Zahnprothesen die Mortalität älterer Menschen reduzieren kann. Die drei wichtigsten Säulen hierfür sind 1.) Modifikation der Prothesenoberflächen hin zu geringerer mikrobieller Attraktivität, 2.) Verbesserung der Prothesenhygiene durch Schulung und professionelle Unterstützung und 3.) Reduktion der Prothesentragedauer, also nächtliche Prothesenkarenz. Die mikrobielle Attraktivität von Prothesenoberflächen zu reduzieren, wurde in der Vergangenheit vereinzelt erfolglos versucht. Erst die seit kurzem mögliche computergestützt-maschinelle (CAD/CAM) Fertigung von Prothesenbasen ist in dieser Hinsicht vielversprechend, da die industriell verarbeiteten Kunststoffe eine geringere Oberflächenrauigkeit sowie eine erhöhte Hydrophilie und geringere freie Oberflächenenergie aufweisen. Im Hinblick auf die Verbesserung der Prothesenhygienemaßnahmen ist in diesem Zusammenhang die Tatsache, dass kognitiv-motorische Screening-Tests nicht als Triagetool für die Beurteilung der Unterstützungsbedürftigkeit pflegebedürftiger Menschen bei der Mund- und Prothesenhygiene geeignet sind, besonders interessant. Da die personellen Ressourcen für eine generalisierte Mund- und Prothesenpflegehilfe meist nicht vorhanden sind, sollten Risikogruppen zumindest auf das nächtliche Prothesentragen verzichten.

POSTER

Antimicrobial activity of octenidine against multidrug-resistant Gram-negative pathogens

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Background: Multidrug-resistant Gram-negative (MRGN) pathogens pose a major and growing threat for health care systems, as therapy of infections is often limited due to the lack of available systemic antibiotics. Besides to the development of new antibiotics to cure the patient, deliberated prevention strategies within health care facilities are a key element in infection control. Application of well tolerated antiseptic molecules may be a very useful measure to be taken, not only to reduce the dissemination of methicillin-resistant *Staphylococcus aureus* (MRSA), but also MRGN on skin, wounds or mucous membranes, respectively. As decolonization strategies with regard to MRSA are already implemented in high-risk areas (i.e. intensive care units), this study aimed to investigate if the same protocol theoretically might be concomitantly efficient against common MRGN.

Materials/methods: A series of 5 different Gram-negative pathogens *(Escherichia coli, Klebsiella pneumoniae, Enterobacter cloacae, Acinetobacter baumannii, Pseudomonas aeruginosa)* was studied to prove efficacy of octenidine (OCT) under clinically relevant conditions according to an official test norm (EN13727). We used 5 clonally-unrelated isolates per species, including a single wild-type strain, and four MRGN isolates, corresponding either to the 3MGRN or 4MGRN definition of multidrug resistance. Several strains were adapted to Chlorhexidine digluconate, often used as standard antiseptic in decolonization protocols (MIC < 156.4mg/L). OCT susceptibility was evaluated with and without organic load.

Results: A contact time of 30sec or 1min was fully effective for all isolates by using different OCT concentrations (0.01% and 0.05%), with a bacterial reduction factor of >5 log systematically observed. Growth kinetics were determined with two different wild-type strains (*A. baumannii* and *K. pneumoniae*), proving a time-dependent efficacy of OCT, mirroring what has been previously observed for MRSA.

Conclusion: These results highlight that OCT, besides being a very effective agent against MRSA, may also be extremely useful in infection control strategies to eradicate emerging highly multidrug-resistant Gram-negative pathogens associated with nosocomial infections.

Measuring alcohol-based hand rub volume used by healthcare workers in practice

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Introduction: A critical variable impacting the efficacy of hand antisepsis is the volume of alcohol-based hand rub (ABHR) applied to the hands during a single HH event (i.e. application volume). It may be assumed that healthcare workers (HCWs) use application volumes recommended by manufacturers. However, manufacturer recommendations for application volume of ABHR vary based on the formulation properties and local requirements with typical recommendations varying from 1 mL – 5 mL. The objective of the study was to determine the mean application volumes of an ABHR used by HCWs during routine clinical practice.

Methods: A personal carriage bottle (125 mL) was engineered to electronically record the number of times product was dispensed. A total of 19 HCWs from five wards in an acute care hospital in Cleveland, OH USA completed the study. The mass of each bottle was measured before and after each shift and the number of uses was recorded from the lid after each shift. The total volume of ABHR used by each HCW during a shift was calculated by dividing the total mass used by the density of the product (0.8607 g/mL). The mean application volume was determined by dividing the total volume by the number of ABHR uses recorded.

Summary: Overall mean ABHR application volume over three shifts = 0.73 mL per HCW. Mean application volumes for individual HCWs ranged from 0.27 mL to 1.61 mL. The mean number of ABHR uses from the personal carriage bottle was 22 per HCW per shift (median 18) and ranged from 2 to 78.

Conclusions: Results of this pilot study suggest that ABHR application volumes in clinical practice are below typical manufacturer and WHO recommendations. This may lead to suboptimal reductions of microbial pathogens and have a negative impact on ABHR effectiveness in clinical practice.

Verhinderung der bakteriellen Einsaat in das Operationsgebiet durch antimikrobielle chirurgische Handschuhe

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Hintergrund: Die mikrobielle Kontamination des sterilen Operationsfeldes ist ein Risikofaktor für postoperative Wundinfektionen. Während Operationen sind Perforationen der sterilen Handschuhe mit bis zu 25% häufig. Ein antimikrobieller chirurgischer Handschuh mit einem inneren Chlorhexidin-basierten Beschichtungskomplex wurde zum Schutz des chirurgischen Personals entwickelt; Auf Grund der mikrobiellen Reduktionsleistung wurde die Hypothese aufgestellt, dass ein solcher Handschuh in der Lage ist, im Fall einer Perforation die bakterielle Einsaat in das Operationsfeld infolge der Reduktion der Hautflora zu verhindern.

Material und Methode: Die Studie wurde als doppel-verblindete randomisierte Studie durchgeführt [ISRCTN trial no.: CCT-NAPN-22137]. Es erfolgt eine Prüfung der mikrobiellen Hautflora nach Tragen eines antimikrobiellen Handschuhs (HS A) an einer, und eines nicht antimikrobiellen Handschuhs (HS B) an der anderen Hand von teilnehmenden Chirurgen bei gefäßchirurgischen Operationen. Bestimmt wurde der Unterschied der Bakterienzahl pro mL Handschweiß im Handschuh nach den Operationen.

Ergebnisse

Mittleres Patientenalter: 72 ± 9 Jahre (60 – 85 Jahre); Männer: n=7; Alter: 73 ±8 Jahre (60 – 84 Jahre); Frauen: n=4; Alter: 70 ± 11 Jahre (61 – 85 Jahre)

Gesamt-OP Dauer: 112 ± 43 Min. (50-185 Min.); Carotis-Endarterektomie (n=4):68 ± 15 Min., Revaskularisationen [A. com. Femoris und A. profunda femoris] (n=3): 129 ± 18 Min., Periphere Bypass-Operationen(n=4): 144 ± 37 Min.

HS A (antimikrobiell): Mittlere Erregerzahl pro mL Handschweiß im Handschuh (mean cfu/mL) 0,28 cfu/mL (median: 0,00; Max.: 2 cfu/mL); HS B (standard): 299,00 cfu/mL (median: 15,00; Max. 3.000 cfu/mL). Der Unterschied zwischen den Erregerzahlen lag bei 1,68 log10 cfu/ml, und war statistisch signifikant unterschiedlich (P = 0.006).

Schlussfolgerung: Während gefäßchirurgischer Eingriffe kann ein antimikrobieller Handschuh die Hautflora der Hand der Operateure signifikant reduzieren. Im Falle einer Handschuhperforation hat das Tragen eines antimikrobiellen Handschuhes damit das Potential, die Erregerkontamination des sterilen Operationsgebiets und eine Kontamination implantierter Fremdkörper zu verhindern. Damit könnte ein neues Konzept im Rahmen der Multibarriere-Strategie zur Prävention von postoperativen Wundinfektionen und zur Steigerung der Patentensicherheit vorliegen.

Mikrobielle Belastung von langärmeligen Arztkitteln – eine Stichprobe

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Langärmelige Arztkittel werden seit geraumer Zeit verdächtigt, aufgrund hoher Keimbelastung eine potentielle Streuquelle für diverse Erreger, darunter auch multiresistente Bakterien, zu sein. Zur Erhebung der mikrobiellen Belastung von langärmeligen Arztkitteln wurde am Landeskrankenhaus Innsbruck, einem 1.600-Betten-Krankenhaus der Maximalversorgung, eine Reihenuntersuchung getragener langärmeliger Arztkittel durchgeführt und die Keimbelastung erhoben. Im Rahmen der Untersuchung wurden 100 von Ärztinnen und Ärzten aller Fachrichtungen getragene Mäntel mittels Objektabklatschmethode auf bakterielle Kontamination untersucht. Hierfür wurde von jedem Mantel jeweils an der rechten und an der linken Ärmelunterseite ein Abklatsch mittels Kontaktplatte (Count-Tact® Agar, Biomérieux, Deutschland) gewonnen. Nach Bebrütung der Proben über Nacht bei 37 °C erfolgte eine Quantifizierung der gesamten Keimlast durch Zählung der koloniebildenden Einheiten (KBE) sowie eine Speziesidentifikation der gezüchteten Keime mittels Massenspektrometrie (MALDI-TOF-MS, Bruker Daltonic, Deutschland). Im Anschluss wurde bei allen Keimen, ausgenommen Keimen der normalen Hautflora, eine Resistenztestung gemäß EUCAST 7.0 Richtlinien durchgeführt um das Vorliegen multiresistenter Keime auszuschließen. In der Auswertung wurde pro untersuchten Ärmel eine mittlere Keimbelastung von 17 KBE/16cm² festgestellt. Pro Mantel ergab sich eine mittlere Keimbelastung von 33,9 KBE/16cm². In der Mehrzahl der untersuchten Mäntel (n=56) wurde linksseitig eine höhere Keimbelastung detektiert als rechtsseitig. Folgende Spezies konnten identifiziert werden: KNS (n=178), Tetrakokken (n=170), aerobe Sporenbildner (n=75), *Corynebacterium sp.* (n=15), *Staphylococcus aureus* (n=4), *Sphingomonas sp., Pseudomonas sp.* (jeweils n=2), *Acinetobacter baumannii, Acinetobacter ursingii, Streptococcus pyogenes* und *Streptococcus salivarius* (jeweils n=1). Es konnten keine multiresistenten Erreger detektiert werden. Die Ergebnisse der durchgeführten Untersuchung bestätigen die erwartete hohe Kontamination der Ärmel von langärmeligen Arztkitteln vor allem mit Keimen der normalen Hautflora. Da auch Keime mit hohem pathogenen Potenzial nachgewiesen werden konnten *(Staphylococcus aureus, Streptococcus pyogenes, Acinetobacter sp., Pseudomonas sp., Sphingomonas sp.)*, kann eine Gefährdung des Patienten bei Übertragung dieser Keime nicht ausgeschlossen werden.

Self-disinfecting surfaces and the activity against Staphyloccocus aureus ATCC 6538 under real life conditions

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Background: The environmental burden in clinical settings and the transmission of healthcare-associated infections through exogenous sources represent a drawback in patient care worldwide. Disinfection measures often lack effectiveness due to inadequate handling. Various self-disinfecting surfaces have been developed as a possible support to reduce surface contamination; however most of them which are currently available on the market have not yet been examined under real-life conditions.

Aim: In this study, we tested five different self-disinfecting surfaces, provided by Fritz Egger GmbH & Co. OG (Austria) (micro-patterned surfaces, zinc molybdenum surfaces, polyguanidin silica surfaces, surfaces with membrane-active polycations incorporated, membrane-active polycations acrylate surfaces). Our aim was to determine their activity against *Staphylococcus aureus (S. aureus)* ATCC 6538, as well as a long-term effectiveness against this pathogen after disinfection.

Method: By applying the dry-inoculation method we provided with study conditions that appropriately mimic the hospital environment.

Findings: Our investigation revealed either a post-antibiotic or killing effect from four out of the five surfaces against *S. aureus* before wipe-disinfection of the surface sample (micro-patterned surfaces, polyguanidin silica surfaces, surfaces with membrane-active polycations incorporated, membrane-active polycations acrylate surfaces). Lack of remaining effects was observed after disinfection with most of the surfaces. However, one sample type, the membrane-active polycations acrylate surfaces, displayed a remaining killing effect for 19 disinfection cycles.

Conclusion: These data suggest that this particular item could provide the basis for the development of improved surfaces with long-term and durable killing effect against various pathogens.

Octenidine against *Staphylococcus aureus* epidemic clones: an alternative strategy

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Mupirocin nasal ointment, together with chlorhexidine-based skin disinfectants, have been used for methicillin resistant *Staphylococcus aureus* (MRSA) decolonization, but emergence of resistance strains highlights the need for alternative strategies. In the present study we investigate the bactericidal activity of octenidine among international epidemic clones of *S. aureus*, including mupirocin resistant strains.

Eight *S. aureus* isolates belonging to different epidemic clones were included. Mupirocin resistance was tested by disk diffusion, E-test and by PCR amplification of *mupA* and *mupB*. The bactericidal efficacy of octenidine was determined with and without organic load (albumin) by a quantitative suspension test in accordance to EN13727.

Two MRSA isolatesshowed high-level mupirocin resistance (MIC>1024 mg/L and *mupA*), two MRSA isolates showed low-level mupirocin resistance (MICs = 24 and 32 mg/L) and the two remaining MRSA and the two MSSA were susceptible to mupirocin (MICs = 0.25 to 0.75 mg/L). Octenidine was effective (bacterial reduction of >6 log10) at a concentration of 0.001% (=10 ppm) after 30 sec of contact time for all isolates tested. With 0.6% albumin, bacterial reduction of >6 log10 was observed with 0.01% octenidine (=100 ppm) at the same contact time.

Octenidine is a promising alternative for *S. aureus* eradication, including mupirocin resistant strains belonging to different epidemic clones, and at significantly lower concentrations than those currently used in the clinical setting (0.05% to 0.1%).

Inaktivierung von Norovirus (MNV-1) auf antimikrobiellen Oberflächen

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Humane Noroviren (Familie *Caliciviridae*) gelten derzeit als häufigste Ursache von nicht bakteriell bedingten Gastroenteritiden in Gemeinschaftseinrichtungen. Weltweit sterben bis zu 8 Millionen Menschen an den Folgen einer Norovirus-Infektion. Durch den erhöhten Pflege- und Medikamentenaufwand von infizierten Patienten sowie einer eingeschränkten Bettenauslastung von betroffenen Einrichtungen entstehen zusätzliche ökonomische Belastungen für das Gesundheitssystem. Veranlasst durch die Fähigkeit von Noroviren für eine lange Zeit auf kontaminierten Oberflächen zu persistieren, werden in dieser Studie antimikrobielle Oberflächen der Firma Egger (Egger antimicrobial surfaces; EAS) in Bezug auf deren antivirale Wirksamkeit getestet. Zur Austestung gelangten Oberflächen mit den Wirkstoffen Zink-Molybdän und Polyhexamethylenguanidin (PHMG) sowie Standard-Vergleichsoberflächen ohne Wirkstoffbeschichtung. Für die Effektivitätstestung wurde das Surrogat-Virus MNV-1 (Murines Norovirus, Familie *Caliciviridae*) auf die zu testende Oberfläche aufgetragen und die reduzierende Eigenschaft der getesteten Oberflächen mittels Plaque-Assay und real-time PCR-Assay ermittelt. EAS mit PHMG-Zusatz (1% in wässriger Reinacrylat-Lösung) zeigten eine signifikante Reduktion ($p=0,0068$) des Virustiters um 0,5 log Stufen. Durch eine vorherige Behandlung der EAS (1% PHMG in wässriger Reinacrylat-Lösung) mit Incidin® Rapid (5%) und Ethanol (70%) konnte eine Titerreduktion von >2,58 log erreicht werden. Bei einer erneuten Kontamination der EAS konnte diese Reduktion nicht wiederholt werden (-0,13 log). Das Ergebnis deutet auf eine Veränderung der Oberflächenstruktur und somit auf eine kurzfristig erhöhte Verfügbarkeit des Biozids durch die Behandlung der EAS mit dem Desinfektionsmittel Incidin® Rapid hin. Aufgrund der geringen Infektionsdosis von humanen Noroviren (10-100 Viren) und der hohen Virus-Ausscheidung von infizierten Personen (1010 Kopien von viraler RNA/ Gramm Faeces) stellen die getesteten Oberflächen keinen ausreichenden Schutz im Sinne einer effizienten desinfizierenden Wirkung und somit Ausbruchsvermeidung dar. Für den praktischen Einsatz werden derzeitig gemeinsam mit der Firma Egger neue EAS entwickelt. Zusätzlich konnte gezeigt werden, dass Effektivitätstestungen von antimikrobiellen Flächen nicht nur die antivirale Wirksamkeit der zu testenden Oberflächen berücksichtigen sollen sondern Langlebigkeits- und Robustheitstests in Hinblick auf notwendige Reinigungs- und Desinfektionsmaßnahmen der EAS umschließen müssen.

Survey in Austrian Healthcare Institutions: Systematic evaluation of medical devices on their compatibility to disinfectants

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Hospitals and other health care institutions are increasingly confronted with specific hygienic challenges. The disinfection of surfaces plays a central role. Incorrect use of surface disinfectants or general material incompatibility can impair the surface material, as seen in the figures below. As a result, not only the appearance can be altered seriously but the defect can lead to a total loss of function and massive economic damage in the worst case.

The cause of these defects lies often with inadequate disinfection procedures provided by the manufacturer and/or distributor of the medical devices. Insufficient material compatibility testing can also lead to an incorrect selection of incompatible surface disinfectants.

Hygline GmbH hygiene issues & management and a group of committed experts evaluated the scope and situation concerning surface disinfectant induced damages of medical devices and furniture in various Austrian healthcare institutions. Additionally, medical devices and commonly used disinfectants were screened for material compatibility, processing procedures and handling.

OFI (Austrian Research Institute) together with other research partners (Happy Plating, ÖTI) carried out disinfectant compatibility tests with the most relevant active substance groups and materials in use. Standardized testing was developed which, in addition to the chemical exposure, had also to incorporate the wiping process, since the mechanical treatment was also regarded as critical.

In the course of the evaluation of 32 different healthcare institutions, including 20 hospitals, 16 defective medical devices and 17 pieces of defective pieces of furniture were found. In 93.8% of the medical devices and 76.5% of furniture surface disinfectants were identified as being responsible for the damage caused. Based on these finding a matrix incorporating relevant polymer materials and metals and their compatibility to active substances commonly used in disinfectants was devised.

Changing the epidemiology of Verotoxin-producing *E. coli* (VTEC) in Austria by introducing a new molecular based diagnostic procedure in one single laboratory in 2016

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Background: VTEC epidemiology has changed in many European countries the last years mainly due to introduction of new -molecular diagnostic tools. We report a rise of incidence of confirmed VTEC-positive samples within a short period of time (1st of August to 17th of November 2016) due to introduction of molecular based stool diagnostics in a single laboratory in Austria.

Methods: 69 stool samples of 3447 in total (2%) were tested positive for stx1/2-genes by multiplex PCR enteric panel technique on the BDMax system. These samples were sent to the National Reference Center for *Escherichia coli* including VTEC (NRCE) were 67 could be confirmed VTEC-positive. Confirmation through further molecular, cultural and serological techniques PCR, Antisera-Agglutination was performed in the NRCE.

Results: The confirmed VTEC-positive samples derive from 67 patients in total of which 55 where symptomatic (mild to severe bloody diarrhea) and 12 were identified as asymptomatic carriers. 55 of 3079 unformed stool samples (1.8%) and 12 of 368 formed stool samples (3.3%) derived from a screening program of employees of food processing companies. Age of patients ranged from 1 to 86 years. Two VTEC serotypes never described in Austria before could be detected; VTEC O176:H4 and O136:HNM.

Conclusions: The 67 confirmed VTEC -positive samples are counting for 33% of all positive samples in Austria up to now (67 of 204). In 2015 in total only 136 human samples were confirmed VTEC-positive in Austria. This clearly demonstrates the dramatic underreporting of this pathogen.

Epidemiology of urinary tract infections in HIV positive patients at a tertiary care hospital in Central Europe (2011 - 2016)

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Background: There is limited published data concerning recent epidemiology of urinary tract infections (UTI) in HIV-patients, thus we analysed independent risk factors for UTI in HIV positive individuals and antimicrobial resistance rates of *E. coli* to antimicrobial agents commonly used in UTI. To determine the prevalence of symptomatic urinary tract infections (UTI) in HIV-patients we performed a retrospective case-control study.

Methods: We included 313 HIV-patients, 101 with UTI and 212 age and gender matched controls, attending the HIV outpatient clinic at the Vienna University hospital (VUH) over a period from January 2011 to September 2016. The patients' specific data was gathered from the electronic database of the VUH. The statistical analysis was performed using SPSS Software Version 20.0.

Results: HIV infected individuals with CD4 count >200 cells/mm3 were less likely than HIV infected individuals with CD4 count <200 cells/mm3 to experience UTI (OR 0.811, 95% CI 0.712–0.923 vs. OR 2.555, 95% CI 1.553 – 4.205, respectively). The in vitro resistance rate of E. coli to antimicrobial agents was as follows: ciprofloxacin (41%), mecillinam (20.5%), trimethoprim (61%), ampicillin (67%), ampicillin/ clavulanic acid (23%), cefuroxime (17%), nitrofurantoin (2%), amikacin (0%), gentamicin (9.5%).

Conclusion: Immunological status (CD4 count) is an important parameter for risk assessment of UTIs in HIV-patients. The increased resistance rate of E. coli to commonly used antimicrobial agents needs to be considered when it comes to the management of UTI, additionally surveillance strategies should be implemented in HIV-patients.

Infection Prevention and Control 4.0?

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Infection Prevention and Control (IPC) is possible, as is proven by sufficiently resourced and structured infection control teams working with appropriate processes. Hence, IPC has become mandatory worldwide as a structured and resourced work item in healthcare of all developed and many other countries.

Surveillance of infection is a key tool in IPC. Surveillance demands (expensive) expert manpower if done conventionally. Therefore, targeted healthcare-associated infection surveillance or point prevalence surveillance often feature as a surrogate for full surveillance. But all patients in our healthcare systems deserve state-of-the-art IPC!

Intelligent IT makes this feasible, with automated real time processing of all relevant medical patient data, which are held and provided by automation-supported electronic documentation. Though IT in real life systems and their interoperability often lack or not suffice, a number of successful practical examples from infection surveillance and other medical fields are in effect.

Intelligent IT has become and will be indispensible in healthcare merely for optimizing allocation of scarce resources - provided that optimized and state-of-the-art healthcare remains an undisputable claim of the public. In near future, we expect strong impulses from IT systems, which link machine learning and structured knowledge. This development appears inevitable, as industry 4.0 seems to be.

Our question is how to address and overcome mental, clinical, technical and other evitable barriers for this “natural” development.

Masernelimination in Europa- wie kann das Ziel bis 2020 erreicht werden

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Nach Pocken und Polio hat sich die Weltgesundheitsorganisation die Ausrottung von Masern zum Ziel gesetzt. In der europäischen Region wurde dieses Ziel schon 2010 angepeilt, jedoch kam es in diesem Jahr zu einer der größten dokumentierten Epidemien in Europa, die vor allem Bulgarien betraf. Seither kommt es immer wieder zum Auftreten von zum Teil großen Ausbrüchen, unter anderem auch in Österreich. Ungeschütztes Gesundheitspersonal ist häufig für eine rapide Ausbreitung verantwortlich. Der Hauptgrund für das Auftreten von Ausbrüchen in Österreich sind unzureichende Durchimpfungsraten der Masern Mumps Röteln (MMR) Impfung, die vor allem durch Impfskepsis bedingt sind.

Während der europäischen Impfwoche 2016 wurden in Tirol verschiedene Aktivitäten durchgeführt, um Awareness zu steigern und die MMR Impfung wurde vermehrt angeboten. Es wurde bei verschiedenen durchgeführten Veranstaltungen ein Knowledge, Attitutes und Practices (KAP) Survey mittels „Convenience Sampling“ und eine Assessment des Impacts der Aktivitäten durchgeführt.

Während des „langen Nachmittags der MMR Impfung“ kam zu einem 17 fachen Anstieg verabreichter MMR Impfungen in Gesundheitsbehörden. Der KAP- Survey zeigte dass, die MMR Impfung am häufigsten (90%) von den Teilnehmern als wichtig eingestuft wurde, gefolgt von Diphtherie-Tetanus-Pertussis (85%), Hepatitis B (77%) Hepatitis A (67%) und FSME (76%). Sieben Prozent der Teilnehmer gaben an, Impfungen nicht für wichtig zu halten. Eine positive Einstellung zu alternativer Medizin (z.B. Homöopathie) (Prevalence Ratio (PR 1.31, 95% CI: 1.10-1.56)) und ein Alter über 50 Jahren (PR 1.22, 95% CI: 1.06-1.40) waren als einzige Determinanten signifikant mit einer negativen Einstellung zu Impfungen assoziiert.

Gesundheitspersonal ist die wichtigste Informationsquelle, die Eltern und Patienten von der Wichtigkeit von Impfungen überzeugen können. Kontinuierliche Aktivitäten, um die Awareness in der Bevölkerung, aber vor allem beim Gesundheitspersonal zu steigern und vermehrte Angebote zeigen eine positive Auswirkung auf die Impfakzeptanz und resultieren daher in einer besseren Impfcompliance.

Detection of bloodstream pathogens by real-time PCR compared to standard conventional blood culture in patients with sepsis.

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Fast detection of pathogens in bloodstream infection and early adequate antibiotic therapy are crucial for a better outcome of patients with sepsis. Conventional blood culture system is still considered as gold standard for diagnosis of bloodstream infection, however, low positivity rates and long time to result are limiting factors.

Thus, the aim of our study was to evaluate a real-time PCR method, the LightCycler SeptiFast assay, for diagnosis of bloodstream pathogens on clinical samples routinely send to our laboratory.

All blood samples, which were investigated for bloodstream pathogens by real-time PCR at our routine laboratory in the period of 2014 to 2015 were included in this retrospective study. Conventional blood culture (BactAlert) obtained at the same day +/-24 hours was used as reference method.

Altogether, 453 blood samples of critically ill patients with presumed sepsis were collected and processed on the same day of sampling. Two-hundred fifty-nine samples (57.2%) were concordant negative with both methods. In 194 positive samples (42.8%) pathogens were detected by either blood culture system or SeptiFast assay or by both methods. Forty-one samples (9,1%) were positive in both assays, 25 samples of these were concordant positive. The agreement rate of blood culture system and SeptiFast assay was 61%. The negative predictive value of SeptiFast assay versus blood culture was 0.88, the sensitivity was 0.54, and the specificity 0.69. The most frequently found pathogens in SeptiFast assay and blood culture were coagulase-negative Staphylococci, followed by Streptococcus sp., Staphylococcus aureus and E. coli.

In conclusion, the SeptiFast assay is a valuable tool for rapid diagnosis of bloodstream infections in addition to, but not as replacement for the blood culture test.

Activity of N-chlorotaurine against *Staphylococcus aureus* and *Pseudomonas aeruginosa* in a long-term in-vitro biofilm model

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Background: N-chlorotaurine (NCT) is an investigational endogenous antiseptic particularly suited for topical therapy of infections of sensitive body regions.
Aims: To investigate the activity against fresh and up to 4 months old biofilms of *Staphylococcus aureus* and *Pseudomonas aeruginosa*.

Methods: Biofilms of ATCC strains of S. aureus or P. aeruginosa were grown in vitro in special microtiter plates MBEC Biofilm Inoculator with 96 well base P&G25 (Innovotech Inc., Edmonton, Canada) for up to 4 months under weekly change of nutritient media. Samples each were removed weekly and treated with 1% NCT in phosphate buffer (pH 7.1) for 30 and 60 min or with plain buffer for 60 min at 37°C. Samples were fixed and subjected to electron microscopy and ultrasonicated and subjected to quantitative cultures, respectively.

Results: Electron microscopy revealed reproducible biofilms of both bacteria. The biomass became significantly more voluminous with *S. aureus* than with *P. aeruginosa*. Accordingly, the structural damage of the biofilm and single bacteria by NCT was more pronounced in *P. aeruginosa*, but clearly visible with both strains. Quantitative cultures showed a highly significant reduction in colony forming units by 0 – 2.5 (0 – 4.5) log10 for *S. aureus* and 0.9 - > 5.0 (4.2 - > 6.2) log10 for *P. aeruginosa* after 30 min (60 min) incubation in 1% NCT. The biofilm of *S. aureus* became more susceptible after a period of development of ≥10 weeks.

Conclusion: NCT has bactericidal activity against Gram-positive and Gram-negative bacteria in short- and long-term biofilms. As expected, more bulky biofilms are more resistant to attack than less bulky ones.

Platelet activation in the course of nosocomial *Candida*: in vivo study using a mouse model and patient samples

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Objectives: Platelets fulfil multifaceted functions in pathogenesis and host defence of infectious diseases such as invasive mycoses. In contrast to previous experiments with *Aspergillus fumigatus*, we found no significant activation of platelets by *Candida* spp. or secreted factors thereof in *vitro* (see abstract: Eberl et al). To study the situation in vivo, we analysed the putative stimulation of platelets by *Candida* in a mouse model as well as in a clinical study with candidemic patients.

Methods: Blood samples derived from candidemia patients or from mice infected intravenously with *C. albicans* were analysed for platelet activation and viability by flow cytometry and ELISA.

Results: Mouse model: *Candida* infection induced platelet activation in the animal model, as detected by the characteristic surface markers CD62P and CD63, binding of fibrinogen to platelets, and formation of microparticles. These findings were confirmed by enhanced concentrations of soluble CD62P. Moreover, the hyphal stage of *C. albicans* induced more pronounced effects on the platelets than the yeast stage.

Patients: Compared to healthy control patients, platelets derived from candidemia patients were clearly activated, as shown by enhanced numbers of microparticles and increased levels of CD62P and CD63. In parallel, an amine binding assay indicated a decrease of platelet membrane integrity and viability, a typical consequence of platelet activation *in vivo*.
The kinetic of these parameters differs between patients, which may be due to differences in medication and underlying diseases.

Conclusion: While platelets were not activated *in vitro* by *Candida* cells or secreted factors, this stimulation could be detected in a mouse model of invasive candidiasis as well as in patients suffering from candidemia. We hypothesize that *Candida* survival in the blood and establishment of sepsis is facilitated by the lack of direct platelet activation, but indirect mechanisms might induce platelet stimulation in vivo.

Nachweis der desinfizierenden Wirkung von Waschverfahren
(Detection of the disinfecting effect of washing processes)

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W.H.U. GmbH

The "Hygiene guideline for laundries that process laundry of health facilities" of the ÖGHMP requires in chapter 2.2 that the applied chemothermal procedures must be listed by ÖGHMP, VAH or RKI.
For the control of chemothermal washing procedures, the guideline requires in point 5.1.2.2. the use of germ carriers between bacteria-tight membranes. This is contrary to RAL 992, which allows only the use of open germ carriers.
We tested for several years open germ carriers (according to EN 16616) and closed germ carriers (according to the guideline of the ÖGHMP) together in different (both listed and not listed) wash procedures. Additional to the disinfecting performance we tested the open germ carriers also for cleaning performance.
The cleaning performance of the open germ carriers was in many cases greater than 99.9%. That means with a contamination of 8 log10 CFU per germ carrier it is not possible to demonstrate a disinfection effect of 5log10 reduction. In addition, *E. faecium* appears to be more easily "cleanable" than the contributing blood contamination.
If the open and closed germ carriers showed different results, the open germ carriers always showed a "better" disinfection than the closed carriers. In some cases we had a malfunction in the washing processes. These were recognized only with the closed germ carriers while open germ carriers farther showed a sufficient disinfecting effect.
The closed germ carriers allow a better distinction of the disinfection efficacy in washing processes. The requirement of the Guideline on the use of closed germ carriers is thus justified.

Prevention of fungal infection by antimycotic drugs can modulate activity and function of human platelets

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Aim:
Recent studies implied that the activity of human platelets might be modulated by some antimycotics used for infection prophylaxis and therapy. Since platelets are also part of the innate immunity this effect might influence the antifungal immune response and thus modulate the outcome of treated patients. For that reason we screened various antimycotic drugs for their effects on platelets.

Methods:
Platelets were mock-treated or incubated with increasing concentrations of various antimycotic drugs. Platelet activity was quantified by FACS detection of the surface markers CD62P and CD63. Morphological changes of platelets were analysed by confocal microscopy. The antifungal capacity of the platelets was investigated by calcofluor quantification of *Aspergillus* growth and life-time microscopy.

Results:
The antimycotic drugs amphotericin B (AmB) and caspofungin were both strong activators of human platelets and induced a time- and dose-dependent increase of the activation markers CD62P and CD63 on the platelet surface. A combinatorial effect between antimycotics and platelet-mediated antifungal killing could be demonstrated for *Aspergillus* hyphae. We further studied in more detail the effect of AmB, since this drug is a standard for most prophylactic and therapeutic regimens. Both liposomal and desoxycholate AmB were able to stimulate platelet activity. The relevant concentrations for this effect were within the physiological range in the patients.
In contrast, some azols such as fluconazole and voriconazole decreased the basic activity of the platelets and interfered with their thrombin-induced stimulation. Other drugs such as micafungin did not modulate the platelet activity at all.

Conclusions:
Antimycotic drugs can influence the activity of platelets and modulate platelet-mediated antifungal killing. Since these processes might crucially influence the course of invasive aspergillosis as well as other fungal diseases further studies to understand the interaction between platelets and antimycotics should be performed.

Analysis of resistance genes in *Candida albicans* and *Candida glabrata* using Next Generation Sequencing

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An increase in antifungal-resistant *Candida* strains has been reported in recent years. The aim of this study was to detect mutations in resistance genes using NGS technology and to determine the impact of the underlying mechanisms of azole and echinocandin resistance.
45 *Candida* strains were examined consisting of susceptible and resistant control strains and azole-resistant, echinocandin-resistant and multi-resistant clinical isolates. The MICs of anidulafungin, micafungin, caspofungin, fluconazole, posaconazole, voriconazole, itraconazole and isavuconazole were determined using the EUCAST microdilution. The genes ERG11, ERG3, TAC1 and GSC1 in *C. albicans*, as well as ERG11, CgPDR1, FKS1 and FKS2 in *C. glabrata* were sequenced using the Illumina MiSeq® Platform.
140 different mutations were identified, of which 53 were missense mutations. Out of these, 42 mutations were found to be presumably causal. 13 mutations have not been reported before. All all echinocandin-resistant *Candida* strains with a MIC at least two titers above the clinical breakpoint showed a mutation in the hotspot regions of FKS1/2 or GSC1. Usually, a cross resistance in echinocandins was found, only one *C. glabrata* strain (P667T in FKS2) showed an isolated susceptibility to micafungin despite high anidulafungin and caspofungin MICs. In contrast to *C. albicans* strains no ERG11 mutations were found in azole-resistant *C. glabrata* strains. Thus, point mutations in ERG11 are not involved in the development of azole resistance in these *C. glabrata* strains. Genetic variations in the transcription factor CgPDR1, which are associated with an overexpression of the efflux pumps CDR1/2, seem to be a more important cause for azole resistance. Loss-of-Function mutations in ERG3 due to premature stop codons seem to be important for resistance acquiring in *C. albicans*. The relevance of TAC1 remains unclear, because no isolated mutation was found in this transcription factor.

Comparison of microbiological laboratory methods for the diagnosis of dermatophytes

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Dermatophytes are fungi able to invade keratin-comprised tissues. While conventional diagnosis relies on selective culture and KOH-microscopy, genetic analyses can increase diagnostic sensitivity and specificity while reducing the necessary time until diagnosis.
For this project, samples with the clinical diagnosis of dermato-/onychomycosis were analyzed using selective fungal culture, 15% potassium hydroxide [KOH] microscopical testing, a commercial multiplex real time-PCR (“FTD Dermatophytes" by Fast-Track Diagnostics) and a panfungal PCR with subsequent ITS-region-sequencing. The combination of selective culture and KOH-microscopy was considered as diagnostic standard. Additionally, in a second step a combined gold standard was calculated (positive in any of the above mentioned tests) and compared to the above mentioned assays. Accuracy, sensitivity, marginal homogeneity (McNemar's test) and inter-rater agreement (Cohen's kappa) were calculated.
Of 195 unique samples, the number of samples testing positive for dermatophytes was n=47 (26%), n=60 (34%) and n=15 (8%) for selective culture or KOH-microscopy [culture/KOH], the multiplex real time-PCR [rt-PCR] and ITS-PCR with subsequent sequencing [sequencing], respectively. Compared to culture/KOH, the rt-PCR showed a sensitivity of 73% and an accuracy of 78% without significant difference in homogeneity. When comparing sequencing to culture/KOH, sensitivity was only 22%, whereas accuracy was 78%. McNemar's test showed highly significant differences between culture/KOH and sequencing. When comparing each test to the combined gold standard, sensitivity was 63%, 81% and 19%, whereas accuracy was 85%, 92% and 66% for culture/KOH, rt-PCR and sequencing, respectively. The negative predictive value was 79%, 88% and 63% and the Cohen's kappa was 0.66, 0.83 and 0.22 for culture/KOH, rt-PCR and sequencing, respectively. Our results show that the rt-PCR is a valuable addition to classical microbiological testing for dermatophytes while decreasing the time until diagnosis. Sequencing of the ITS-region can be used to identify individual species in a sample, although sensitivity and inter-rater agreement are significantly lower compared to other methods.

Comparison of *Fusarium* Diagnosis Methods – Evaluation of Cultivation, Sequencing and Proteomic Analysis

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Besides *Fusarium* being a fungal genus commonly affecting plants, cases of human fusariosis are known. In the last decades the number of fusariosis has been on the rise, often due to contact lens-associated keratitis or patients with immunodeficiency or immunosuppression after organ transplantation. For the identification of different *Fusarium* strains, micro- and macromorphology is helpful, but genetic or proteomic analysis is often required.
For this project we evaluated all cases of *Fusarium* infections at the General Hospital of Vienna (Medical University of Vienna) since 2007. We collected 50 isolates from 33 different patients since 2007. Identification was performed using micro- and macromorphological aspects after incubation for 3 weeks and confirmed by sequencing and proteomic analysis. In addition, the history of all patients was reviewed.
Most infections were contact lens-associated and isolated either from the contact lens fluid (33%) or directly from the eye (12%). 18% of *Fusarium* isolates were collected from wounds or surgical sites, 18% were taken from broncho-alveolar lavage (BAL) fluids or sputum from patients after lung transplantation.
All used methods were able to identify *Fusarium* species, although for strain analysis EF sequencing performed best, followed by MALDI-TOF proteomic analysis. ITS sequencing was inferior to both EF sequencing and MALDI-TOF proteomic analysis, although still superior to classic micro- and macromorphological analysis.
Although *Fusarium* infections in humans are rare, we could find 3-6 cases each year at the General Hospital of Vienna (Medical University of Vienna). 75% of infections are caused by *F. oxysporum* or *F. solani*. As a diagnostic tool, EF sequencing was in most cases equal or superior to MALDI-TOF proteomic analysis, although being more time-consuming. ITS sequencing was inferior to both MALDI-TOF proteomic analysis and EF sequencing, although in most cases sufficient to detect the genus *Fusarium* when further identification of the species was not necessary.

A suspected case of Lassa fever: a bundle of problems

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Background
Lassa fever is a viral hemorrhagic fever (VHF) that is endemic in West Africa. The virus is classified as Biosafety-Level- 4 (BSL-4) pathogen. According to the so-called Styrian-epidemic-plan, in case of a suspected BSL-4 infection a team of experts would manage the case. Not having a BSL-4 laboratory for Lassa in Austria the blood samples were sent to the Bernhard-Nocht-Institute in Hamburg for diagnosis.

Case
A 34-year old patient had been to Nigeria for professional purposes. Six days after returning to Austria he got subfebrile and consulted a general practitioner (GP), where he mentioned the possibility of Lassa fever, because of confirmed cases near his accommodation in Nigeria. The GP took blood samples and sent them to our microbiological laboratory. Due to patient's travel history the informed team of experts couldn't exclude Lassa fever. The transport of the blood samples to Bernhard-Nocht-Institute in Hamburg was organized, 36 hours later the result of the PCR was available and negative. The patient didn't develop further symptoms.

Problems
- Blood samples were transported per Post to a routine laboratory without prior information
- On the attached referrals Lassa fever was only one among plenty of other diagnoses. Until recognizing the risk of VHF the blood sample was already opened and in process.
- There was a delay in the information of the Health authorities
- The organization of the transport to Hamburg was definitively complex due to the lack of authorized and trained personal

Conclusion
Diagnosis of VHF is a rare case in Austria. In Styria a local plan exists but the knowledge about the Styrian-epidemic-plan is low. Sending blood samples not mentioning suspected BSL-4 viruses on the referrals endangers the microbiological staff. Transport to appropriate laboratories has to be checked and act out, to avoid any delay in diagnosis.

Determination of reversion frequency of clinical thymidine-dependent Small-Colony Variants of *Staphylococcus aureus*

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Background:
Staphylococcus aureus Small Colony Variants (SCVs) are known for being a method of adaptation to environmental stress like antibiotic therapy or the host immune system. The SCV phenotype can persist in eukaryotic cells for a long time and later colonize the host again extracellularly. *S. aureus* SCVs possess the ability to revert back to the wild type phenotype, which poses a problem in correct identification and treatment of the infection.

Aim:
In this study, we characterized the reversion frequency of clinical thymidine-dependent Small Colony Variants of *S. aureus* from the SCV phenotype to the wild type phenotype.

Material and Methods:
Twelve thymidine-dependent *S. aureus* Small Colony variants (TD-SCVs) isolated from Cystic Fibrosis patients were genetically characterized and were tested for their frequency of reversion in a novel experimental setting.

Conclusion:
All TD-SCVs tested for frequency of reversion showed a quite diverse ability to revert back to the wild type phenotype, ranging from up to 90% of revertants to 0%. The frequency of reversion was not dependent on the primary mutations that led to the SCV phenotype.
In conclusion, the ability of TD-SCVs to revert back to the wild type phenotype not only depends on the primary mutations in *thyA* which lead to their thymidine dependency, but also other factors. We suggest that this is due to different genetic backgrounds of the strains, resulting in a different reversion frequency.

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Druck: ROBIDRUCK, A-1200 Wien – www.robidruck.co.at