Antimicrobial Resistance
National Research Programme

www.nrp72.ch
Wildhainweg 3, P.O. Box, CH-3001 Berne

**Swiss National Science Foundation**
Programmes division
National Research Programmes
+41 (0)31 308 22 22
nrp72@snf.ch

**Full Proposal: Research Plan**

**Second call of the NRP 72 "Antimicrobial Resistance"**

Division IV, National Research Programmes (NRP)

| | |
|---|---|
| **Corresponding applicant**<br>Name, first name | Harbarth, Stephan (HUG) |
| **Further applicant(s)**<br>Name, first name | François, Patrice; Schrenzel, Jacques; Pugin, Jérôme |
| **Project title**<br>(English) | An interventional study to evaluate the impact of a rapid screening strategy in improving nosocomial ESBL and CPE control in critically ill patients |

Please indicate to which research module your project belongs:

☒ **Module 3:** AMR control: interventional studies

# 1. RESEARCH PLAN

## 1.1. STATE OF RESEARCH IN THE FIELD

### 1.1.1. EXTENDED-SPECTRUM BETA-LACTAMASES (ESBL) AND CARBAPENEMASES: AN INCREASING NOSOCOMIAL HEALTH THREAT

ESBL-producing Enterobacteriacae (ESBL-PE) are now spreading worldwide.[1] The dissemination of multiple resistant clones[2,3] is related to ESBL-PE food chain contamination and silent household transmission[3–7] and worldwide, through travelers,[4] immigrants and visiting friends' relatives.[8–11] It is further supported by an increasing human,[4,12] animal [3,4,7] and environmental[13] reservoir. Hospitals[14] are impacted by this problem, and further nosocomial spread takes place through patient-to-patient transmission,[14] via the hands of healthcare workers,[14,15] but also other facilitating factors such as antibiotic selection pressure,[16] contaminated food[17] or the inanimate healthcare environment.[12,18] However, the detailed epidemiology and transmission routes may differ depending on the types of ESBL-PE.

Digestive carriage by ESBL-PE places patients at high risk of antibiotic-resistant nosocomial infection. Bloodstream infections[19] caused by ESBL-PE increase length of hospital stay[19] and mortality, through an increased delay before adequate empirical therapy regimen is administered.[20,21] We have recently shown in a large European cohort study that ESBL-PE bacteraemia significantly increases the risk of death (adjusted hazard ratio 1.63; 95% CI: 1.13–2.35), the length of stay (4.9 days; 95% CI: 1.1–8.7) and the cost compared with susceptible strains.[22]

Although still rare in Switzerland,[23] there is also a worldwide increase in carbapenemase-producing Enterobacteriacae (CPE), causing a major public health problem. We recently conducted an international prospective cohort study in 10 low- and middle-income countries (LMIC) with endemic CPE occurrence.[24] After adjusting for potential confounders, carbapenem resistance was associated with increased probability of in-hospital mortality (aHR, 1.65; 95% CI 1.01-2.69) and decreased probability of discharge alive (aHR, 0.65; 95% CI 0.49-0.88). Thus, among patients with bacteremia caused by Enterobacteriaceae in LMIC, carbapenem resistance is associated with adverse health outcomes and adds to the global burden of AMR.

## 1.1.2. ESBL/CPE CONTROL MEASURES IN HOSPITALS, THE COSTS OF SUCH MEASURES AND THE CURRENT UNMET DIAGNOSTIC NEEDS

ESBL-PE control measures may include standard precautions or contact precautions, including also transfer to single rooms or cohort isolation depending on the nosocomial acquisition rate of a particular EBSL-PE. Targeted screening for asymptomatic ESBL-PE carriers may be warranted, but remains a matter of debate.[25] However, in the intensive care unit (ICU) setting, early and rapid identification of critically ill patients colonized with ESBL-PE and subsequent prevention of patient-to-patient spread of ESBL-PE through proper infection control remain potentially useful interventions to control ESBL-PE cross-infection. Thus, targeted ESBL-PE screening upon ICU admission may help to identify unknown ESBL carriers, prevent transmission and could help to reduce time to adequate treatment in case of ESBL infection. Nevertheless, current microbiologic screening methods to detect previously unknown ESBL-PE carriers are slow and cumbersome. This delay impacts the discontinuation of pre-emptive isolation measures among patients at high risk of ESBL-PE carriage and lead to a human and monetary cost.

Indeed, isolation measures act as a barrier disrupting the interaction between patients and caregivers, impeding the quality of care and probably leading to depression and anxiety.[26] In a prospective cohort study, attending physicians were observed during the morning round to examine only 35% of patients in contact isolation (31 patients) when they examined at the same time 73% of the patients not in contact isolation (108 patients).[27] Another study conducted in both surgical intensive care unit (ICU) and surgical wards of a university hospital reported an overall contact time decreased in isolated patients, from 19 +/-4 to 34 +/-7 min/h. (P=0.5) despite the same severity of illness (APACHE II >10).[28] More concerning, in a cohort gathering patients hospitalized and diagnosed with a congestive heart failure, preventable adverse events occurred more frequently in those isolated for MRSA colonization and infection (n= 72) compared to matched controls without isolation (n= 144) (23 vs 5 per 1000 days, P<0.001).[29] Ultimately, several studies observed an association between isolation and an increased anxiety and depression as well as a decreased patient satisfaction with care.[30,31] However these results are still controversial due to the poor design of these studies. Overall, isolation measures also drive a financial cost, intrinsic to their implementation, but also due to their impact on the healthcare worker labour and the patient care.[32]

Diagnostic screening methods for detection of ESBL-PE carriers include both phenotypic and molecular methods. Even if phenotypic methods are efficient and inexpensive for detecting the most frequent pathogens,[33] their performance remains dependent on the sampling quality[34] and volume, and suffer major diagnostic delays in routine daily practice of at least 48 hours due to traditional culture-based systems and working hour restrictions.[35] Molecular methods address some of these pitfalls, sparing the culturing effort, and resulting in a reduced turn-around time.[36,37] They can also decrease the required volume of sampling because of their improved sensitivity,[21] but their breadth is limited to the selected molecular targets. Their cost-effectiveness is still unclear, but might be attractive when compared to the cost of unnecessary isolation.[38] In order to improve effective ESBL-PE control strategies in the ICU setting, there is a need for a fast, sensitive, and reasonably specific but also cost-effective screening test.[39]

To further complicate matters, there is ongoing controversy whether carriers of ESBL-producing E.coli still require contact precautions, in contrast to other ESBL-PE, such as Klebsiella spp and Enterobacter spp.[40] Availability of new data on nosocomial E. coli transmission questions the complete cessation of contact precautions to contain the spread of ESBL-producing E.coli in units with immunocompromised, critically ill or elderly patients with extended length of stay.[40]

In contrast to the ongoing debate about ESBL E.coli screening and contact isolation, most experts agree that patients at risk of CPE carriage (e.g. patient transferred from hospitals in hyper-endemic regions, like Italy or Greece) should be screened for CPE carriage upon ICU admission and placed into preemptive isolation, until negative screening results are obtained. However, current microbiologic culture methods are slow and not adapted to the rapid turnover in busy tertiary care ICU settings.

Overall, there is an ongoing need for a fast, reliable and inexpensive diagnostic screening method for ESBL-PE and CPE strains of major concern in Switzerland requiring contact precautions in the ICU setting. A novel strategy with 2 rapid diagnostic methods could allow individualizing and speeding up the implementation of appropriate infection control measures, or discontinue preemptive isolation as fast as possible. First, the loop-mediated isothermal amplification reaction (LAMP) is an isothermal molecular amplification method already developed and previously validated in our institution (see section 1.3.).[41] Compared to conventional PCRs for the detection of the main ESBL-PE and CPE types, this new technique is faster and potentially more cost-beneficial, but with a similar diagnostic accuracy.

### 1.1.3. LOOP-MEDIATED ISOTHERMAL AMPLIFICATION TEST

LAMP is a molecular amplification method using a DNA polymerase, Bst polymerase, providing self-replication and strand displacement through the formation of a loop with the help of 4 primers spanning 6 locations on the original DNA target.[42] Details of the LAMP method can be found at the following website: http://loopamp.eiken.co.jp/e/index.html.[43] This technique does not use thermal cycles as PCR[44] and its enzyme is less susceptible to inhibitors than the Taq polymerase,[45,46] faster,[44] and as sensitive[47] and specific as home-made qPCR assays.[46] The robustness and cost-effectiveness of LAMP may therefore be useful in the routine microbiology laboratory or as a delocalized assay.[48] LAMP reagents have already been approved for the detection of different types of DNA and RNA viruses but also bacteria with good accuracy.[47] This technique has also been evaluated for the detection of ESBL-PE and carbapenemases through several studies. In 2014, Thirapanmethee et al compared LAMP with PCR and Double Disk Synergy Test (DDST) for various clinical specimens and found a high specificity and better analytical sensitivity than conventional PCR in targeting the bla-CTX-M9 gene.[49] Performance comparison between LAMP and chromogenic agar using enriched broths was similar for the main ESBLs genes,[50] and demonstrated concordance with DDST in the case of phenotypic compatibility with carbapenemases,[51] yet with a substantially shorter turn-around time (TAT).

## 1.2. RESEARCH BY APPLICANTS AND LOCAL EPIDEMIOLOGY

### 1.2.1. EXPERIENCE OF OUR INTERDISCIPLINARY TEAM IN THE FIELD

The research groups of the applicants involved in the present proposal have a long and particularly fruitful collaborative history regarding the development and clinical evaluation of molecular assays for the control of multi-resistant bacteria in our institution.[52,53] In 2003, an interventional study conducted in our ICUs compared the median time intervals from admission to notification of test results between a multiplexed real-time qPCR assay and standard microbiological tests during MRSA screening.[54] A few years later, a prospective, interventional cohort study using a cross-over design was conducted in 6 surgical wards to determine the benefits for using the same rapid test as above as a universal screening test for MRSA carriage.[55] This clinical trial was funded by the SNSF and has until now generated 6 publications about the clinical, health-economic and epidemiologic evaluation of a rapid, universal MRSA screening strategy.[55–60]

In the context of increasing emergence and spread of ESBL-PE and CPE, the infection control program and the microbiological laboratory at HUG have already extensively contributed to the research agenda. We previously conducted epidemiologic studies,[61] prevalence surveys and clinical trials facilitated by systematic screening programs implemented in routine for specific services[62] or for certain studies[63]. Molecular typing has been used in studies to investigate outbreaks and the virulence of bacterial strains.[62,64] Other studies conducted by the present group of investigators focused on the impact of antibiotic use on emergence of ESBL-PE[65], and an oral decolonization regimen on intestinal ESBL-PE carriage through a randomized placebo-controlled clinical trial.[66]

### 1.2.2. PERSONAL CONTRIBUTION TO RESEARCH IN THE FIELD

Prof Stephan Harbarth (MD, MS) is a well-known infectious disease epidemiologist with extensive research experience in the field of antimicrobial use and resistance. In the past, he has devoted substantial efforts toward control of resistance through improved infection control and antibiotic use. He is currently hospital epidemiologist, board-certified attending physician in infectious diseases and director of the antibiotic stewardship programme at HUG, with more than 150 original articles about issues related to control of antimicrobial resistance in peer-reviewed journals. He has successfully conducted or contributed to several controlled trials and RCTs in the area of infection control and infectious diseases.[67,68] Moreover, S.H. is the principal investigator of an EU-funded multicentre RCT that examines the administration of antibiotics and faecal microbiota transplantation to decolonize carriers of ESBL-PE and CPE (NCT02472600). Finally, he has recently contributed to the new CPE guidelines issued by WHO and SwissNoso.
Prof Jacques Schrenzel (MD) is associate professor of medicine at the University of Geneva, Faculty of Medicine; director of the Bacteriology Laboratory and head of the Genomic Research Laboratory (www.genomic.ch). Both laboratories provide a panel of highly specialized tests, and integrate new technologies and new diagnostic assays to clinical care whenever this is relevant for patients or public health. The bacteriology research team conducts clinical, diagnostic and translational investigations, in different fields of microbiology.
PD Dr Patrice François (PhD) has been directly involved in the creation and development of the Genomic Research Laboratory, a state-of-the-art laboratory equipped with the required platforms to study infectious agents at the genome level. This laboratory develops and deploys assays to follow the local epidemiology of pathogens and improve

the diagnosis of MDROs of clinical interest.[69–71] His research group is familiar with epidemiological and clinical research projects in the field of infectious diseases. It uses molecular tools and technologies that allow assessing microbes at different levels, e.g. the transcriptome and proteome levels. These tools have been successfully used for epidemiologic studies of different MDROs.
Prof Jérôme Pugin (MD) has received his medical degree from the University of Geneva in 1984 and currently works as the director of the Adult Intensive Care Service at HUG. He is board certified both in Internal Medicine and in Intensive Care Medicine. He is also heading the laboratory of basic research of the Intensive Care Service, hosted by the Faculty of Medicine Research Facilities (Department of Microbiology and Molecular Medicine, Centre Médical Universitaire, University of Geneva). His research has been funded in part by the Swiss National Foundation for Scientific Research for over 25 years. His area of research is sepsis and severe bacterial infections in critically ill patients. Prof. Pugin is internationally recognized as an opinion leader in this field. He has published over 100 original papers in peer reviewed journals. In particular, Prof. Pugin has extensively studied and published on host-bacteria interactions, and the relevance of various biological markers in the care of patients with suspected bacterial sepsis.

### 1.2.3. LOCAL ESBL-PE AND CPE EPIDEMIOLOGY

ESBL-PE are now endemic in Switzerland. At HUG, it represents the most pressing threat of antibiotic resistance to patient safety, with more than 100 episodes of bacteremia caused by ESBL-PE in 2015. A recent WGS study (manuscript submitted) documented 62% of *E.coli* ST131 (including 37% of ST131 H30) among 89 bloodstream isolates caused by ESBL-producing *E.coli* during that year. Among those *E.coli* ST131 isolates, 53 of 56 (95%) belonged either to the CTX-M-1 or CTX-M-9 groups with the following distribution: 30 CTX-M-15 (CTX-M-1 group), 19 CTX-M-27 (CTX-M-9 group), 3 CTX-M-14 (CTX-M-9 group) and 1 CTX-M-24 (CTX-M-9 group).

An observational, prospective study found in 2010 among 13 internal medicine units a prevalence of 4.8% ESBL-PE carriers (1'072 patients included) and 4.4% ESBL-PE acquired in the hospital (473 patients).[63] A more recent observational study from 2013 to 2015 found a prevalence of 10.6% (226/2136 patients) of ESBL-PE carriage on admission in 4 different services, including 73.4% of *E.coli* (166/2136 patients).[72] The beta-lactamase CTX-M-15 (belonging to the CTX-M group 1) was also particularly prevalent in HUG among index patients.[73] In the R-Gnosis WP5 trial, among the 4 participating sites, HUG had the highest on-admission prevalence of ST131 (44%), compared to Berlin (22%), Utrecht (19%) and Madrid (14%). Importantly, among 147 tested ESBL-PE, 142 (97%) belonged to the CTX-M-1 or CTX-M-9 groups. More specifically, among the 66 non-*E.coli* ESBL-PE, only 4 contained SHV-12 (n=2) or TEM (n=2) ESBL enzymes.

In our ICUs, we perform on-admission screening of high-risk patients and weekly routine screening to detect previously unknown ESBL-PE and CPE carriers. In this setting, the mean weekly prevalence of ESBL-PE carriage was 10.2% in 2016, with an average of 2.4 newly identified ESBL-positive patients per week in 2016.

The Swiss antibiotic surveillance system (anresis.ch) observed an increasing number of CPE throughout Switzerland, from 69 isolates in 2013 to 142 isolates in 2016 (mainly *E.coli* and *K.pneumoniae*), with the highest number of CPE isolates identified in Geneva.[74] Not surprisingly, the HUG is at increased risk for CPE importation through international patients and repatriated U.N. workers. In a recent national CPE surveillance report, Geneva had a significantly higher CPE isolation rate compared to the rest of Switzerland (RR 1.65; 95% CI: 1.02-2.68).[23]

## 1.3. DETAILED STUDY PLAN

### 1.3.1. STUDY PURPOSE

The overarching goal of this proposal is to improve the early and rapid detection of patients colonized with ESBL-PE of infection control concern and/or CPE, and implementation of adequate and individualized preventive measures (e.g. starting/stopping of pre-emptive isolation) in one ICU of a Swiss University Hospital with endemic ESBL-PE occurrence and sporadic CPE importation.

## 1.3.2. SPECIFIC STUDY AIM AND HYPOTHESES

This study aims to evaluate the effectiveness of 2 novel and rapid tests as an innovative screening strategy to improve the implementation or discontinuation of ESBL-PE and CPE control measures among critically ill patients. We will test the specific hypotheses that a screening program with a novel diagnostic strategy enabling early detection of ESBL-PE and CPE carriage in ICUs at HUG can:

1. Decrease unnecessary isolation-days for patients suspected to be colonized with ESBL-PE and CPE, but who are negative by screening;
2. Decrease the time between patient screening and contact isolation of previously unknown ESBL-PE and CPE carriers in this high-risk setting;
3. Decrease the risk of nosocomial cross-transmission of ESBL-PE;
4. Reduce unnecessary costs from an institutional perspective and provide a cost-effective screening option.

## 1.3.3. STUDY POPULATION AND SETTING

This study will be conducted from January 2018 to December 2020 in 2 ICUs at HUG, a tertiary care hospital in Switzerland performing 18'687 rectal swabs (2016) for ESBL-PE screening purposes. The 34-bed mixed medical and surgical ICUs at HUG admit over 2'500 critically ill patients per year with a mean length of stay of 3.8 days. The ICUs are separated into 2 sectors on the same floor (niveau Opéra vs niveau Juliard). In this service, during 2016, 3'395 rectal screening swabs were performed for either on-admission or weekly screening purposes.

Both sectors have identical infection control guidelines and are supervised by the same medical team. Each unit has a limited number of isolation rooms. Compliance with standard infection control precautions is at a satisfactory level; in 2016, hand hygiene compliance was 65%. Current ESBL-PE prevalence on weekly screening varies, on average, between 8 and 15%. By contrast, MRSA on-admission prevalence has substantially decreased from 6.7% in 2004 to less than 1 new case of ICU-acquired MRSA colonization or infection per month since January 2016. For rapid MRSA on-admission screening, we use a PCR-based assay that has already accelerated substantially the identification of MRSA carriers and decreased unnecessary preemptive isolation.[54]

## 1.3.4. SPECIFIC PREVENTIVE MEASURES FOR ESBL-PE AND CPE

Currently, when an ICU patient infected or carrying a non-*E.coli* ESBL-PE and/or CPE strain is detected, specific measures are immediately implemented in order to prevent transmission of the pathogen or its resistant gene(s). In addition to standard precautions, gloves and hydrophobic overcoats are required before contacting the patient and its environment.[75] If respiratory samples are positive, droplet precautions (e.g. mask) are mandatory before each close contact with the patient.[76] Extensive terminal cleaning is performed after a non-*E.coli* ESBL- or CPE-positive patient discharge.[77] Contact precautions have been abandoned for all ESBL-producing *E.coli* carriers because of their low potential for nosocomial cross-transmission and because of their main acquisition source (community from the food chain and not hospital).

Contact precautions might also be deployed in a preemptive manner, for patients previously known as carriers, for contacts of a patient newly detected as a carrier during ICU stay and for transferred patients at risk before obtaining negative results. As a reminder, a pre-emptive isolation day is defined as a day during which an ICU-patient stayed under contact precautions while awaiting the results of the on-admission ESBL-PE and carbapenemases screening.

For identified CPE carriers, physical isolation in a single side room with dedicated medical devices is mandatory. Dedicated material, devices and surfaces have to be disinfected daily and after patient discharge. Non-dedicated material has to be disinfected after usage. Lingerie and waste are treated separately. Visits to the patient and its movements in the hospital are strictly controlled.[78] Specific preventive measures for CPE carriers are implemented during the entire hospitalization, until discharge.

## 1.3.5. CLINICAL TRIAL

### 1.3.5.1. Study design

This prospective, interventional, quasi-experimental interventional study will be prepared, conducted and analyzed over 3 years (January 2018 to December 2020), in order to compare a rapid ESBL-PE and CPE screening strategy with the currently used routine diagnostic method.

The true trial phase encompasses a 12-months intervention period, followed by a 1-month Wash-Out period and a 6-months control period. The intervention period will introduce the ESBL/CPE control program based on the novel rapid diagnostic strategy, in addition to current culture-based microbiological tests. The control period will then only use culture-based microbiological tests. Practical and scientific reasons (e.g. group-level transmission effects) make it impossible to randomly assign individual patients to one of the study groups. A cross-over design in this situation was impossible due to ethical reasons. The waiver of informed consent is necessary for a study targeting ecological effects in infection control of critically ill patients, but was not granted by ethical committees, considering potential risks from the screening strategies (unknown diagnostic performances in our epidemiological context).

Inclusion criterion: All screening samples performed during the routine surveillance of ICU will be included in our study, in order to generalize the results of this research to the whole population of patients admitted in ICUs.

Exclusion criterion: For the same reasons mentioned above, there will be no formal exclusion criteria. Furthermore, the study would suffer from significant bias if there was any exclusion criteria, as the routine surveillance and the infection control program have to be implemented on a group level and not at the individual patient level.

### 1.3.5.2. Main intervention

In the intervention period, a novel ESBL/CPE screening strategy will be implemented according to the rapid diagnostic laboratory method described below.

Infection control guidelines established at our institution will not be modified during the entire study period, including contact precautions of identified carriers of ESBL-PE other than *E.coli* and CPE; spatial separation of patients into cohorts in case of large clusters; decontamination of the environment; guidelines to adapt perioperative prophylaxis of identified ESBL/CPE carriers; and monthly feedback of ESBL/CPE surveillance results to the concerned units.

### 1.3.5.3. Main outcome measures

Unnecessary time (in *days*) patients spent under preemptive contact precautions will be measured as primary endpoint, and stratified by diagnostic strategy (novel vs. current diagnostic method). As secondary outcomes, we will determine (1) time (in *hours*) to notification and implementation (or discontinuation) of contact isolation measures and (2) nosocomial ESBL/CPE transmission events, which will be analyzed using the results of the weekly screening program performed on a routine basis in all sectors of the ICU. Outcome measures will be computed for the intervention and control groups separately and compared. Important aggregate-level data will also be collected in order to adjust for potential confounders.

### 1.3.5.4. Sample collection, processing and notification of results

Screening process and sample collection

Previously established ESBL/CPE screening criteria will not be modified in the 2 participating ICU sectors. The current screening recommendations include the following populations:

a) Transferred patients at risk of ESBL/CPE carriage (risk-based, targeted screening)
b) Patients previously identified and already known as ESBL/CPE carriers (risk-based, targeted screening)
c) Contacts and room-mates of patients newly detected as carrier through a positive clinical culture or weekly screening (risk-based, targeted screening)
d) Any patient hospitalized in the 2 ICU sectors on Monday morning (universal weekly screening)

Rectal swabs (eSwab™, Copan) will be collected by trained ICU nurses during weekly screening for each ICU patient and at arrival of a patient at risk of ESBL/CPE carriage. Trained personnel will also fill in the laboratory request via a computerized order entry system and assure timely transport of specimens to the routine microbiology laboratory. Current experience from our ICUs shows that screening performance is excellent (compliance, 80-90%) and does not interfere with routine work activities. Sequential screening will be performed for high-risk patients given their high pre-test probability to be MDROs carriers.

Microbiological work-up and notification

Rectal swabs will be sampled and addressed to the bacteriology laboratory in a timely fashion. Swabs will be processed in two different workflows according to the period (cf. intervention period vs. control period). Those obtained during the control period will be processed using currently used, conventional phenotypic methods, as described in section 1.3.6. Swabs obtained from patients during the intervention period will be separated into 3 parts to be processed in parallel. The first part will be run by the LAMP technique and a second will be processed by standard bacteriology methods for pathogen identification and quality assurance purposes (i.e. confirmed presence of *E.coli*), and the third part will be stored at -20°C and could be used to resolve any potential discordant results between molecular and phenotypic approaches. The bacteriology laboratory will process non-stop all diagnostic samples related to the study during weekdays until 17h00 and Saturdays until 14h00.

### 1.3.5.5. Data collection and definitions

#### 1.3.5.5.1. Microbiological data:

The screening samples (rectal swabs) will be collected during the routine MDRO surveillance program performed in the ICU. This routine screening program, in place for 15 years, advocates screening weekly on every ICU patient, at admission for patients at risk, for roommates of a newly positive ESBL-E and/or CPE, after detection of a clinical culture positive for ESBL-E and/or CPE, and iteratively before stopping the contact precautions of a high-risk patient.

#### 1.3.5.5.2. Health-related data:

TATs for ESBL/CPE screening and work up will be recorded for both study phases with the help of computerized laboratory databases and stored in a log file for the purpose of statistical analysis. Retrieved parameters will include the following time intervals (in *h*):

1) Time from screening to sample delivery to the laboratory;
2) Time from arrival at the laboratory to reporting of results;
3) Time from result notification to implementation or discontinuation of contact precautions.

We will document all new cases of ESBL/CPE detected during ICU stay. A previously unknown ESBL/CPE case will be defined as any patient in whom ESBL/CPE will be isolated for the first time on ICU admission or during ICU stay. A transmission event will be defined as the acquisition of ESBL-PE or CPE proven by a positive screening test for patients with previously negative results in clinical cultures and screening swabs, including the weekly ICU screening.

Clinical data for the analysis of this trial will be based on information routinely collected for infection control surveillance and stored in the electronic health record (e.g. origin of patient, microbiologic data, length of stay, infection type etc.) without use of a specific case report form.

### 1.3.5.5. Data code and storage

The data collected might contain sensitive information (Admission/discharge from ICU, presence of a MDRO pathogen...), which are however already collected for routine MDRO surveillance purposes at HUG. According to the specified procedure of Swissethics concerning the use of Microsoft Excel in research, we will copy the data in an Excel file secured with a password and will frequently save PDF dated copies, which will again be dated and signed manually by the project direction. We therefore be able to track all the changes. Printed copies will be locked at the study center. Only the research team will have an access to these copies. The Case Report Form collecting this data will not contain any personal information susceptible to directly identify a patient. These data will therefore be linked to the patient through a code included in a separate Excel file. This file will be securely stored in the server of the Geneva University Hospitals. We cannot anonymize the data here for institutional surveillance purposes. The microbiologic data relative to this study will not be coded as they contain the same information as the data retrieved in routine.

As this study is monocentric and a very few persons will have an access to the database, the data changes will be very rare. Therefore, we will use a simple excel file as a Case Report Form to collect the health-related and microbiological data, very closely related to similar files used at Service PCI. Only the research team will have access to them. The microbiological material will be the same as used in routine and will therefore be stocked under the routine condition.

Once the research is over, the health-related data will be anonymized to be archived in the research file. This anonymization will be achieved through the destruction of the code (separate Excel file). Without this code, the data stored cannot be crossed to identify any patient and are thus considered as anonymized. The microbiological data will be conserved as they contain the same information as the one retrieved in practice and might help to perform prevalence study and outbreak investigation.

### 1.3.5.6. Sample size determination

Regarding the primary outcome and based on an expected overall ESBL-PE carriage rate of 20-40% among high-risk patients, we hypothesize that among 1'000 preemptive isolation days in our ICUs, up to 70% are unjustified because of either the absence of any ESBL/CPE carriage or the presence of non-ST131 ESBL-producing E.coli isolates. After the introduction of our novel screening approach, we estimate a reduction of these unnecessary isolation days from 70% to less than 30%. According to the Z test, the sample size is estimated at 202 isolation days that are necessary to prove a significant decrease of the unjustified proportion of isolation days. Considering an average preemptive isolation time of 2 days for a screened patient, 101 patients with pre-emptive isolation have to be included. This sample size will be easily reached within the planned trial period of 2x6 months.

### 1.3.5.7. Statistical analysis

Statistical analysis will be performed in the 3rd year of the study (2020); no interim analyses are planned for the present trial. Time intervals in between the different time points will be expressed as medians and compared using nonparametric tests. Chi-square or Fisher's exact test will be used to compare categorical variables and Student's t-test, for continuous variables. Outcome measures will be computed for both periods and compared. Adjusted incidence-density ratios of ESBL-PE acquisition will be calculated by segmented Poisson regression analyses, using the Generalized Estimating Equation approach in order to adjust for clustering effects. We will attempt to adjust for various confounders documented at the aggregate level in both ICUs: ESBL-PE colonization pressure; antibiotic usage; severity of illness (SAPS score); patient-to-nurse ratio and hand hygiene compliance. The Kaplan-Meier method will describe the time to implementation or discontinuation of contact precautions in each study group, and eventual transmission events observed. All data will be stored in a specifically designed data base, and data analyses will be performed with STATA and R.

## 1.3.6. DIAGNOSTIC STRATEGY IN THE CONTROL PERIOD

### 1.3.6.1. Screening tests for ESBLs and CPE

For CPE and ESBL-PE screening from rectal swabs we will routinely use in parallel three media: chromID ESBL (BioMérieux), MacConkey agar with ertapenem, meropenem and imipenem disks (10 µg), and chromID® OXA-48. All colonies with distinct morphotypes will be identified by matrix-assisted laser desorption ionization–time of flight (MALDI-TOF) mass spectrometry and the antibiotic susceptibility profil of each isolate will be determined by the disc diffusion method using EUCAST breakpoints and recommendations.[82] For ESBL confirmation, we will use double-disk synergy tests (DDST20 and DDST30). In doubtful cases, ESBL & AmpC Screen Kit 98008 (Rosco Diagnostica) will be used as a second line confirmatory test. These different phenotypic tests ensure a broad coverage with a high sensitivity and specificity for ESBL-PEs,[83] but take at least from 24h to 48h for the whole process, depending on the time of day/week.

For CPE confirmation, we will use a chromogenic OXA-48 plate concomitantly to chromID ESBL and the McConkey plates to improve the screening performance for the carbapenemase OXA-48.[84] RAPIDEC® CARBA NP and the same LAMP tests as used during the interventional period targeting the main carbapenemases will be used as CPE confirmation tests.

## 1.3.7. DIAGNOSTIC STRATEGY IN THE INTERVENTION PERIOD

### 1.3.7.1. Screening methods

The novel diagnostic strategy will combine the molecular information yielded by LAMP and phenotypic information to identify the pathogen and confirm the absence of an ESBL-PE or CPE. LAMP will first detect the main genes coding for ESBLs and carbapenemases directly on rectal swabs. Samples will always be processed in parallel with conventional methods in order to keep a gold standard and to quantify potentially false negative results leading to inadequate early discontinuation of pre-emptive contact precautions.

a) LAMP assay:
The LAMP eazyplex SuperBug CRE assay (AxonLab, UK) used in this study is a qualitative genotypic test and comprises a freeze-dried and ready-to-use mixture for an isothermal amplification reaction. It covers ESBLs and carbapenemases of the CTX M-1 and CTX M-9 families, VIM, NDM, KPC families, and OXA-48 (-48, -181) from Gram-negative bacteria.[85] The provided materials are the primers detecting these targets and the master mix and enzyme for performing multiplex assays based on isothermal amplification. Visualization of results is provided in this assay through real-time fluorescence measurement of a fluorescent dye bound to double stranded DNA using the GENIE® II instrument.[86] The process of this LAMP assay takes about 20 to 30 min. All of the LAMP assays developed have an analytical sensitivity estimated by the manufacturer to be around 10CFU/reaction ($10^3$ bacteria/mL), with a technical analytical sensitivity measured at 8.1CFU/reaction for assays targeting CTX M-1 or M-9.

As a basis, the LAMP technology has already proved to be robust,[46] cost-effective,[47] real time[46,87] and performant for detecting ESBLs and carbapenemases on screening isolates.[51] The specific LAMP eazyplex superbug CRE assay also proved solid performances in the literature for the detection of various ESBLs- and carbapenemases-producing Enterobacteriaceae on isolates.[88] A UK study compared in 2015 the diagnostic performance between the Eazyplex SuperBug Complete A kit performed on the GENIE II platform with a reference standard using PCR assays and a commercial microarray (Check-MDR CT102) on 450 clinical isolates from various bacterial species. The overall test sensitivity and specificity were reported as 95.5% and 100% respectively, although it missed the detection of 18/102 OXA-48 variant carbapenemases genes. The delivery of a modified test "Eazyplex SuperBug complete B kit" resolved later this issue and identified the 18 OXA-181 producers.[89] Another Spanish study compared the Eazyplex SuperBug CRE kit performed on the GENIE II platform with minimum inhibitory concentration profiles, the modified Hodge test and double-disc synergy tests to identify carbapenemases and ESBLs, but also with conventional PCR assays and sequencing to characterize these resistant enzymes. This study performed on 94 genotypically characterized carbapenemases-producing strains and 45 clinical isolates observed a 100% agreement between the Eazyplex SuperBug CRE system results and the PCR and sequencing results. Another 100% agreement was found between the inferred phenotype of clinical isolates and the Eazyplex SuperBug CRE system results.[51] Other studies reported acceptable results during exams performed on direct samples. LAMP demonstrated similar performances on isolates and cultures, for the direct detection of *E.coli* on fresh urine samples.[45]

b) For patients at a very-high risk for MDRO screening:
Specific care are defined for patients at a very-high risk to carry an ESBL or a CPE. Risk profiles are ascertained at a case-by-case basis based on the patient profile and current epidemiological situation. Rational behind this specific care rely on a high pre-test probability of this specific population. In order to increase the negative predictive value of the screening method, control measures will only be de-implemented in the intervention arm after several repetition of the LAMP test. Current practices also require three negative results of these tests before preemptive isolation cessation.

### 1.3.7.3. Interaction with other MDROs screening policies and infection control measures in the ICU.

Other MDROs screened in the ICU surveillance program are MRSA, VRE and MDR-*Acinetobacter baumanii*. MRSA and VRE screening practices, as well as related infection control measures will not be modified during this study. Screening detection for MDR *Acinetobacter baumanii* will be slightly modified with the use of a PCR targeting *Acinetobacter baumanii* for already defined high-risk patients. Effectively, there is a high prevalence in high-risk patients of MDR-Acinetobacter, justifying the use of a rapid screening test in this situation. Prevalence of VRE in this specific population is lower.

### 1.3.8. HUMAN STUDY CONSIDERATIONS

The division head of the ICU is active member of this research team and agreed to the aim and interventions of this study. The current study protocol will be submitted for approval by the institutional review board (IRB). As a diagnostic quality improvement study, informed consents from the patients will not be required, based on our previous experience with the Geneva central ethics committee. Indeed, previous activities to ensure surveillance of MDRO transmission and cross-infection have been approved by the IRB at HUG, which considered them as an integral part and cornerstone activity to reach the main goal of the infection control program: to reduce MDRO infection rates, and improve quality of care and patient safety.

An annual safety report will be submitted once a year to the local Ethics Committees. This report will contain information from both ICUs about any unforeseen events related to the experimental diagnostics and trial procedures, including potential adverse outcomes related to premature withdrawal of contact precautions.

## 1.4 TIMETABLE AND MILESTONES

Beginning of the study: 1 January 2018
Project initiation (workflow establishment, staff training): January to December 2018
Intervention phase: January 2019 to June 2019
Washout period: July – August 2019
Control phase: September 2019 to February 2020
Data analysis: March 2020 to December 2020
Abstract preparation for international meetings (ECCMID 2021): Fall 2020
Preparation and submission of final manuscript and report to SNSF/PNR72: December 2020

GANTT chart:

| Years | Months | MILESTONES OF THE CLINICAL TRIAL | | | | |
|---|---|---|---|---|---|---|
| | | Study initiation | Intervention | Washout period | Control | Analysis |
| 2018 | January – March | | | | | |
| | April – June | | | | | |
| | July – Sept | | | | | |
| | Oct - December | | | | | |
| 2019 | January – March | | | | | |
| | April – June | | | | | |
| | July – Sept | | | | | |
| | Oct - December | | | | | |
| 2020 | January – March | | | | | |
| | April – June | | | | | |
| | July – Sept | | | | | |
| | Oct - December | | | | | |
| 2021 | January – March | | | | | |
| | April – June | | | | | |
| | July – Sept | | | | | |
| | Oct - December | | | | | |

In 2018, the study will be prepared and the novel tests implemented and validated on a routine basis in the bacteriology laboratory. During the first interventional period in 2019 (12 months), ICU will use the novel diagnostic strategy while after the wash-out period and during the control period, ICU will implement contact precautions according to the standard microbiologic method. During the analysis phase, the main objectives of this clinical trial will be analyzed sequentially, with the cost-effectiveness analysis performed during the last stage.

# 2. IMPLEMENTATION

## 2.1. PREVIOUS ACHIEVEMENTS IN KNOWLEDGE AND TECHNOLOGY TRANSFER

Prof Harbarth is an active member of the infection control program at HUG (Director: Prof. Pittet), a WHO Collaborating Centre for patient safety and infection control. The program has significant experience with translational research. The Geneva model for hand hygiene is now implemented in more than 180 countries in the context of WHO's "Clean Care is Safer Care" campaign.[90] In the context of the previous SNSF call NRP 49 ("antimicrobial resistance"), Prof Harbarth has conducted one of the first clinical trials examining the impact of universal screening for MRSA at hospital admission on nosocomial infections in surgical patients.[55] This article has been cited over 400 times in the literature and has influenced practices and guidelines with regard to MRSA control in many countries around the world.

The group of Prof Harbarth is currently conducting several clinical and epidemiological studies to evaluate key questions related to the control of the acquisition, transmission and infection by MDROs. He participates in several ongoing large-scale EU-funded studies (R-GNOSIS, AIDA, COMBACTE) and coordinates the DRIVE-AB project to address this public health threat. All these projects had or will have impact on clinical guidelines and international policy making. In particular, our group has experience with influencing policy-making at the highest level. The latest and probably most striking example is the release in June 2016 of the European Council recommendations on AMR control, approved by all EU health ministers, which mention explicitly the achievements of the DRIVE-AB project (http://drive-ab.eu) coordinated by Prof Harbarth: "*16. The council of the European Union UNDERLINES that in order to stimulate the development of new antimicrobials, alternative therapies and (rapid) diagnostics, EU and global coordination and cooperation on research programs and incentives are needed and RECOGNISES the work done by the Innovative Medicines Initiative (IMI) project DRIVE-AB (Driving reinvestment in research and development and responsible antibiotic use) …*"

## 2.2. ACTIVITIES PLANNED

Prof Harbarth is extensively involved in the strategic planning and supervision of the "*Strategy on Antibiotic Resistance, Switzerland*" (StAR) and NOSO action plans, accepted by the Federal Council and implemented by the Federal Office of Public Health in Switzerland. Measure 4.1 of the StAR programme clearly states that antibiotic resistance must be detected as early as possible and its spread prevented. In human medicine, this requires reducing the risk of patients introducing resistant microorganisms into a hospital or nursing home, for instance through rapid screening upon admission to detect previously unknown carriers of MDROs. This measure of the StAR is tightly linked to measure M-3 of the NOSO Strategy, which focuses on early detection systems within institutions. The proposed intervention of this clinical trial addresses this issue, and if successful could be a promising method to ensure these aims on a national level, after adaptation to local resources, epidemiological needs and infrastructure.

Dissemination of the results will be prepared through several channels. The results obtained during this study will be shared through an executive summary and a detailed final report to those who need to know within the institution. Abstracts will be presented to national and international meetings of several societies: intensive care medicine, infectious disease, and infection control. Scientific original papers will be submitted to peer-reviewed journals.

Results of the trial could be generalised and applied to other healthcare institutions in Switzerland. These results might be shared to contribute to the implementation of this diagnostic strategy, after taking into account differences in baseline ESBL-PE and CPE prevalence. Therefore, direct contact will be established with key stakeholders in other Swiss hospitals and microbiology laboratories to discuss whether a screening strategy based on similar rapid diagnostic tests should be evaluated and implemented at a wider scale.

## 2.3. IMPLEMENTATION PARTNERS: REFERENCES AND CONTRIBUTIONS PLANNED

This is an interdisciplinary project involving the infection control program, the laboratory of clinical microbiology and the ICUs at HUG. The principal investigators of this research have already extensively contributed to the research agenda about MDRO control including also ESBLs-PE/CPE prevention as well-known infectious disease epidemiologists. Thus, the results of this project will certainly find a large audience within and outside the academic setting.

As mentioned above, Prof Harbarth is member of the StAR-M core group and will make sure that the findings of this trial will also be widely disseminated among public health stakeholders and policy makers in the Federal Office of Public Health.

# 3. SIGNIFICANCE

## 3.1. SCIENTIFIC SIGNIFICANCE

### 3.1.1. THE IMPLEMENTATION OF A NOVEL DIAGNOSTIC STRATEGY IN A MICROBIOLOGICAL LABORATORY

This proposal allows application of a novel rapid diagnostic strategy as a routine screening tool to detect ESBL-PE/CPE colonization and improve patient safety by rapid identification and isolation of ICU patients carrying these multi-resistant bacteria. It will also allow earlier discontinuation of preemptive isolation in high-risk patients with negative screening results. Thus, this project may help to establish screening recommendations for national ESBL control guidelines and promote technological development with a direct impact on clinical care.

### 3.1.2. AN EFFECTIVENESS STUDY EVALUATING THE LAMP TECHNOLOGY IN A REAL-LIFE CLINICAL SETTING

As underlined in a recent NICE medtech innovation briefing (02.2017), none of the studies evaluated so far the LAMP easyplex superbug CRE assay in clinical practice, using rectal swabs. This research provides a unique opportunity to conduct an effectiveness study on LAMP in the ICU setting with clinically relevant outcomes. This study might also stimulate the future research agenda in order to integrate a cost-efficient and robust test in various clinical settings, which might be used later in LMICs to contribute to the international effort of prevention against emergent antibiotic resistance.

## 3.2. SOCIAL AND ECONOMICAL SIGNIFICANCE

The implementation of rapid molecular tests as a routine screening for ESBLs-producing *Enterobacteriaceae* will improve our epidemiological knowledge on the prevalence and spread of the circulating strains. Currently, this knowledge is only supported by phenotypic information, providing the information on resistance and susceptibility, but barely helpful to investigate nosocomial or community outbreaks.

Consequences of unnecessary isolation days on patient care and costs are well-known,[91,92] as well as the burden of infection caused by ESBL-producing bacteria.[19,93] A novel diagnostic strategy potentially able to reduce these consequences might decrease human and financial burden due to antibiotic resistance. Therefore, the results of this study should be of valuable help for those in charge of infection control, for hospital administrators, and for those managing budgets of large healthcare organizations.

Finally, Switzerland, like other European countries, is on the edge of more widespread spread of CPE at a local or regional level. It's low CPE prevalence is good news in that preventive measures will be largely cost-effective and more likely to succeed than if high CPE levels were already present. Therefore, it is likely that the proposed project may help to establish screening recommendations for national CPE control guidelines.

Ultimately, this project will promote technological development with a direct impact on daily clinical practice, by contributing to the earlier recognition of MDRO carriers and improved quality of patient care.

## 4. BIBLIOGRAPHY


1. Chong Y, Shimoda S, Yakushiji H, et al. Community spread of extended-spectrum β-lactamase-producing Escherichia coli, Klebsiella pneumoniae and Proteus mirabilis: a long-term study in Japan. J Med Microbiol 2013;62(Pt 7):1038–43.

2. Birgy A, Levy C, Bidet P, et al. ESBL-producing Escherichia coli ST131 versus non-ST131: evolution and risk factors of carriage among French children in the community between 2010 and 2015. J Antimicrob Chemother 2016;71(10):2949–56.

3. Oteo J, Pérez-Vázquez M, Campos J. Extended-spectrum [beta]-lactamase producing Escherichia coli: changing epidemiology and clinical impact. Curr Opin Infect Dis 2010;23(4):320–6.

4. Haverkate MR, Platteel TN, Fluit AC, et al. Quantifying within-household transmission of extended-spectrum β-lactamase-producing bacteria. Clin Microbiol Infect Off Publ Eur Soc Clin Microbiol Infect Dis 2016;

5. Rodríguez-Baño J, López-Cerero L, Navarro MD, Díaz de Alba P, Pascual A. Faecal carriage of extended-spectrum beta-lactamase-producing Escherichia coli: prevalence, risk factors and molecular epidemiology. J Antimicrob Chemother 2008;62(5):1142–9.

6. Ben-Ami R, Rodríguez-Baño J, Arslan H, et al. A multinational survey of risk factors for infection with extended-spectrum beta-lactamase-producing enterobacteriaceae in nonhospitalized patients. Clin Infect Dis Off Publ Infect Dis Soc Am 2009;49(5):682–90.

7. Lo W-U, Ho P-L, Chow K-H, Lai EL, Yeung F, Chiu SS. Fecal carriage of CTXM type extended-spectrum beta-lactamase-producing organisms by children and their household contacts. J Infect 2010;60(4):286–92.

8. Valverde A, Turrientes M-C, Norman F, et al. CTX-M-15-non-ST131 Escherichia coli isolates are mainly responsible of faecal carriage with ESBL-producing Enterobacteriaceae in travellers, immigrants and those visiting friends and relatives. Clin Microbiol Infect Off Publ Eur Soc Clin Microbiol Infect Dis 2015;21(3):252.e1-4.

9. Wickramasinghe NH, Xu L, Eustace A, Shabir S, Saluja T, Hawkey PM. High community faecal carriage rates of CTX-M ESBL-producing Escherichia coli in a specific population group in Birmingham, UK. J Antimicrob Chemother 2012;67(5):1108–13.

10. Laupland KB, Church DL, Vidakovich J, Mucenski M, Pitout JDD. Community-onset extended-spectrum beta-lactamase (ESBL) producing Escherichia coli: importance of international travel. J Infect 2008;57(6):441–8.

11. Ostholm-Balkhed A, Tärnberg M, Nilsson M, et al. Travel-associated faecal colonization with ESBL-producing Enterobacteriaceae: incidence and risk factors. J Antimicrob Chemother 2013;68(9):2144–53.

12. Khan AS, Dancer SJ, Humphreys H. Priorities in the prevention and control of multidrug-resistant Enterobacteriaceae in hospitals. J Hosp Infect 2012;82(2):85–93.

13. Korzeniewska E, Korzeniewska A, Harnisz M. Antibiotic resistant Escherichia coli in hospital and municipal sewage and their emission to the environment. Ecotoxicol Environ Saf 2013;91:96–102.

14. Brusselaers N, Vogelaers D, Blot S. The rising problem of antimicrobial resistance in the intensive care unit. Ann Intensive Care 2011;1:47.

15. Gupta A, Ampofo K, Rubenstein D, Saiman L. Extended Spectrum β Lactamase-producing Klebsiella pneumoniae Infections: a Review of the Literature. J Perinatol 2003;23(6):439–43.

16. Hilty M, Betsch BY, Bögli-Stuber K, et al. Transmission dynamics of extended-spectrum β-lactamase-producing Enterobacteriaceae in the tertiary care hospital and the household setting. Clin Infect Dis Off Publ Infect Dis Soc Am 2012;55(7):967–75.

17. Calbo E, Freixas N, Xercavins M, et al. Foodborne Nosocomial Outbreak of SHV1 and CTX-M-15-producing Klebsiella pneumoniae: Epidemiology and Control. Clin Infect Dis 2011;52(6):743–9.

18. Muzslay M, Moore G, Alhussaini N, Wilson APR. ESBL-producing Gram-negative organisms in the healthcare environment as a source of genetic material for resistance in human infections. J Hosp Infect 2016;

19. Stewardson A, Fankhauser C, De Angelis G, et al. Burden of bloodstream infection caused by extended-spectrum β-lactamase-producing enterobacteriaceae determined using multistate modeling at a Swiss University Hospital and a nationwide predictive model. Infect Control Hosp Epidemiol 2013;34(2):133–43.

20. Tumbarello M, Sanguinetti M, Montuori E, et al. Predictors of mortality in patients with bloodstream infections caused by extended-spectrum-beta-lactamase-producing Enterobacteriaceae: importance of inadequate initial antimicrobial treatment. Antimicrob Agents Chemother 2007;51(6):1987–94.

21. Pitout JDD, Laupland KB. Extended-spectrum beta-lactamase-producing Enterobacteriaceae: an emerging public-health concern. Lancet Infect Dis 2008;8(3):159–66.

22. Stewardson AJ, Allignol A, Beyersmann J, et al. The health and economic burden of bloodstream infections caused by antimicrobial-susceptible and non-susceptible Enterobacteriaceae and Staphylococcus aureus in European hospitals, 2010 and 2011: a multicentre retrospective cohort study. Euro Surveill Bull Eur Sur Mal Transm Eur Commun Dis Bull 2016;21(33).

23. Ramette A et al. Carbapenemase-producing Enterobacteriaceae in Switzerland from 2013 to 2016; 2017;

24. A. Stewardson, K. Marimuthu, S. Sengupta, A. Allignol, M. Carvalho, T. Walsh, S. Harbarth. The impact of carbapenem resistance on outcomes of bloodstream infections with Enterobacteriaceae in developing countries: an international prospective cohort study (OS 460). 2017;

25. Tacconelli E, Cataldo MA, Dancer SJ, et al. ESCMID guidelines for the management of the infection control measures to reduce transmission of multidrug-resistant Gram-negative bacteria in hospitalized patients. Clin Microbiol Infect Off Publ Eur Soc Clin Microbiol Infect Dis 2014;20 Suppl 1:1–55.

26. Fätkenheuer G, Hirschel B, Harbarth S. Screening and isolation to control meticillin-resistant Staphylococcus aureus: sense, nonsense, and evidence. The Lancet 2015;385(9973):1146–9.

27. Saint S, Higgins LA, Nallamothu BK, Chenoweth C. Do physicians examine patients in contact isolation less frequently? A brief report. Am J Infect Control 2003;31(6):354–6.

28. Evans HL, Shaffer MM, Hughes MG, et al. Contact isolation in surgical patients: a barrier to care? Surgery 2003;134(2):180–8.

29. Stelfox HT, Bates DW, Redelmeier DA. Safety of Patients Isolated for Infection Control. JAMA 2003;290(14):1899–905.

30. Tarzi S, Kennedy P, Stone S, Evans M. Methicillin-resistant Staphylococcus aureus: psychological impact of hospitalization and isolation in an older adult population. J Hosp Infect 2001;49(4):250–4.

31. Morgan DJ, Diekema DJ, Sepkowitz K, Perencevich EN. Adverse outcomes associated with Contact Precautions: a review of the literature. Am J Infect Control 2009;37(2):85–93.

32. Sprague E, Reynolds S, Brindley P. Patient Isolation Precautions: Are They Worth It? [Internet]. Can. Respir. J. 2016 [cited 2018 Feb 2];Available from: https://www.hindawi.com/journals/crj/2016/5352625/

33. Wiegand I, Geiss HK, Mack D, Stürenburg E, Seifert H. Detection of Extended-Spectrum Beta-Lactamases among Enterobacteriaceae by Use of Semiautomated Microbiology Systems and Manual Detection Procedures. J Clin Microbiol 2007;45(4):1167–74.

34. Queenan AM, Foleno B, Gownley C, Wira E, Bush K. Effects of Inoculum and β-Lactamase Activity in AmpC- and Extended-Spectrum β-Lactamase (ESBL)-Producing Escherichia coli and Klebsiella pneumoniae Clinical Isolates Tested by Using NCCLS ESBL Methodology. J Clin Microbiol 2004;42(1):269–75.

35. Gazin M, Paasch F, Goossens H, Malhotra-Kumar S, MOSAR WP2 and SATURN WP1 Study Teams. Current trends in culture-based and molecular detection of extended-spectrum-β-lactamase-harboring and carbapenem-resistant Enterobacteriaceae. J Clin Microbiol 2012;50(4):1140–6.

36. Huang T-D, Bogaerts P, Ghilani E, et al. Multicentre evaluation of the Check-Direct CPE® assay for direct screening of carbapenemase-producing Enterobacteriaceae from rectal swabs. J Antimicrob Chemother 2015;70(6):1669–73.

37. Lau AF, Fahle GA, Kemp MA, Jassem AN, Dekker JP, Frank KM. Clinical Performance of Check-Direct CPE, a Multiplex PCR for Direct Detection of blaKPC, blaNDM and/or blaVIM, and blaOXA-48 from Perirectal Swabs. J Clin Microbiol 2015;53(12):3729–37.

38. Wintermans BB, Reuland EA, Wintermans RGF, Bergmans AMC, Kluytmans J a. JW. The cost-effectiveness of ESBL detection: towards molecular detection methods? Clin Microbiol Infect Off Publ Eur Soc Clin Microbiol Infect Dis 2013;19(7):662–5.

39. Viau R, Frank KM, Jacobs MR, et al. Intestinal Carriage of Carbapenemase-Producing Organisms: Current Status of Surveillance Methods. Clin Microbiol Rev 2016;29(1):1–27.

40. Tschudin-Sutter S, Lucet J-C, Mutters NT, Tacconelli E, Zahar JR, Harbarth S. Contact precautions for preventing nosocomial transmission of ESBL-producing Escherichia coli - a point/counterpoint review. Clin Infect Dis Off Publ Infect Dis Soc Am 2017;

41. A. Cherkaoui, G. Renzi, P. François, J. Schrenzel. Extended-spectrum b-lactamase (ESBL) and carbapenemases detection directly from rectal swabs with the rapid isothermal amplification-based eazyplex SuperBug CRE assay. 2016;

42. Notomi T, Okayama H, Masubuchi H, et al. Loop-mediated isothermal amplification of DNA. Nucleic Acids Res 2000;28(12):E63.

43. Notomi T, Mori Y, Tomita N, Kanda H. Loop-mediated isothermal amplification (LAMP): principle, features, and future prospects. J Microbiol Seoul Korea 2015;53(1):1–5.

44. Mori Y, Kanda H, Notomi T. Loop-mediated isothermal amplification (LAMP): recent progress in research and development. J Infect Chemother 2013;19(3):404–11.

45. Hill J, Beriwal S, Chandra I, et al. Loop-Mediated Isothermal Amplification Assay for Rapid Detection of Common Strains of Escherichia coli. J Clin Microbiol 2008;46(8):2800–4.

46. Francois P, Tangomo M, Hibbs J, et al. Robustness of a loop-mediated isothermal amplification reaction for diagnostic applications. FEMS Immunol Med Microbiol 2011;62(1):41–8.

47. Parida M, Sannarangaiah S, Dash PK, Rao PVL, Morita K. Loop mediated isothermal amplification (LAMP): a new generation of innovative gene amplification technique; perspectives in clinical diagnosis of infectious diseases. Rev Med Virol 2008;18(6):407–21.

48. Fang X, Liu Y, Kong J, Jiang X. Loop-Mediated Isothermal Amplification Integrated on Microfluidic Chips for Point-of-Care Quantitative Detection of Pathogens. Anal Chem 2010;82(7):3002–6.

49. Thirapanmethee K, Pothisamutyothin K, Nathisuwan S, Chomnawang MT, Wiwat C. Loop-mediated isothermal amplification assay targeting the blaCTX-M9 gene for detection of extended spectrum β-lactamase-producing Escherichia coli and Klebsiella pneumoniae. Microbiol Immunol 2014;58(12):655–65.

50. Anjum MF, Lemma F, Cork DJ, et al. Isolation and detection of extended spectrum β-lactamase (ESBL)-producing enterobacteriaceae from meat using chromogenic agars and isothermal loop-mediated amplification (LAMP) assays. J Food Sci 2013;78(12):M1892-1898.

51. García-Fernández S, Morosini M-I, Marco F, et al. Evaluation of the eazyplex® SuperBug CRE system for rapid detection of carbapenemases and ESBLs in clinical Enterobacteriaceae isolates recovered at two Spanish hospitals. J Antimicrob Chemother 2015;70(4):1047–50.

52. Francois P, Renzi G, Pittet D, et al. A novel multiplex real-time PCR assay for rapid typing of major staphylococcal cassette chromosome mec elements. J Clin Microbiol 2004;42(7):3309–12.

53. Francois P, Huyghe A, Charbonnier Y, et al. Use of an automated multiple-locus, variable-number tandem repeat-based method for rapid and high-throughput genotyping of Staphylococcus aureus isolates. J Clin Microbiol 2005;43(7):3346–55.

54. Harbarth S, Masuet-Aumatell C, Schrenzel J, et al. Evaluation of rapid screening and pre-emptive contact isolation for detecting and controlling methicillin-resistant Staphylococcus aureus in critical care: an interventional cohort study. Crit Care Lond Engl 2006;10(1):R25.

55. Harbarth S, Fankhauser C, Schrenzel J, et al. Universal screening for methicillin-resistant Staphylococcus aureus at hospital admission and nosocomial infection in surgical patients. JAMA 2008;299(10):1149–57.

56. Harbarth S, Huttner B, Gervaz P, et al. Risk factors for methicillin-resistant Staphylococcus aureus surgical site infection. Infect Control Hosp Epidemiol 2008;29(9):890–3.

57. Harbarth S, Sax H, Uckay I, et al. A predictive model for identifying surgical patients at risk of methicillin-resistant Staphylococcus aureus carriage on admission. J Am Coll Surg 2008;207(5):683–9.

58. Murthy A, De Angelis G, Pittet D, Schrenzel J, Uckay I, Harbarth S. Cost-effectiveness of universal MRSA screening on admission to surgery. Clin Microbiol Infect Off Publ Eur Soc Clin Microbiol Infect Dis 2010;16(12):1747–53.

59. De Angelis G, Allignol A, Murthy A, et al. Multistate modelling to estimate the excess length of stay associated with meticillin-resistant Staphylococcus aureus colonisation and infection in surgical patients. J Hosp Infect 2011;78(2):86–91.

60. Huttner B, Robicsek AA, Gervaz P, et al. Epidemiology of methicillin-resistant Staphylococcus aureus carriage and MRSA surgical site infections in patients undergoing colorectal surgery: a cohort study in two centers. Surg Infect 2012;13(6):401–5.

61. Stewardson AJ, Renzi G, Maury N, et al. Extended-Spectrum β-Lactamase–Producing Enterobacteriaceae in Hospital Food: A Risk Assessment. Infect Control Amp Hosp Epidemiol 2014;35(4):375–83.

62. Agostinho A, Renzi G, Haustein T, et al. Epidemiology and acquisition of extended-spectrum beta-lactamase-producing Enterobacteriaceae in a septic orthopedic ward. SpringerPlus 2013;2(1):91.

63. Pasricha J, Koessler T, Harbarth S, et al. Carriage of extended-spectrum beta-lactamase-producing enterobacteriacae among internal medicine patients in Switzerland. Antimicrob Resist Infect Control 2013;2:20.

64. Ruppé E, Olearo F, Pires D, et al. Clonal or not clonal? Investigating hospital outbreaks of KPC-producing Klebsiella pneumoniae with whole-genome sequencing. Clin Microbiol Infect Off Publ Eur Soc Clin Microbiol Infect Dis 2017;

65. Vernaz N, Huttner B, Muscionico D, et al. Modelling the impact of antibiotic use on antibiotic-resistant Escherichia coli using population-based data from a large hospital and its surrounding community. J Antimicrob Chemother 2011;66(4):928–35.

66. Huttner B, Haustein T, Uçkay I, et al. Decolonization of intestinal carriage of extended-spectrum β-lactamase-producing Enterobacteriaceae with oral colistin and neomycin: a randomized, double-blind, placebo-controlled trial. J Antimicrob Chemother 2013;68(10):2375–82.

67. Nobre V, Harbarth S, Graf J-D, Rohner P, Pugin J. Use of procalcitonin to shorten antibiotic treatment duration in septic patients: a randomized trial. Am J Respir Crit Care Med 2008;177(5):498–505.

68. Nthumba PM, Stepita-Poenaru E, Poenaru D, et al. Cluster-randomized, crossover trial of the efficacy of plain soap and water versus alcohol-based rub for surgical hand preparation in a rural hospital in Kenya. Br J Surg 2010;97(11):1621–8.

69. Lee AS, Macedo-Vinas M, François P, et al. Impact of combined low-level mupirocin and genotypic chlorhexidine resistance on persistent methicillin-resistant Staphylococcus aureus carriage after decolonization therapy: a case-control study. Clin Infect Dis Off Publ Infect Dis Soc Am 2011;52(12):1422–30.

70. Von Dach E, Diene SM, Fankhauser C, Schrenzel J, Harbarth S, François P. Comparative Genomics of Community-Associated Methicillin-Resistant Staphylococcus aureus Shows the Emergence of Clone ST8-USA300 in Geneva, Switzerland. J Infect Dis 2016;213(9):1370–9.

71. van der Mee-Marquet N, Diene SM, Chopin F, Goudeau A, François P. Enigmatic occurrence of NDM-7 enzyme in the community. Int J Antimicrob Agents 2016;47(6):505–7.

72. Fankhauser C, Schrenzel J, Prendki V, et al. Prevalence of extended-spectrum betalactamase producing–Enterobacteriaceae (ESBL-E) carriage on admission at Geneva University Hospitals (HUG). Antimicrob Resist Infect Control 2015;4(Suppl 1):P120.

73. Fankhauser C, Zingg W, Francois P, et al. Surveillance of extended-spectrum-beta-lactamase-producing Enterobacteriaceae in a Swiss Tertiary Care Hospital. Swiss Med Wkly 2009;139(51–52):747–51.

74. Centre suisse pour le contrôle de l'Antibiorésistance. Anresis.ch [Internet]. 2015 [cited 2016 Nov 6];Available from: http://anresis.ch/

75. Mesure spécifique CONTACT - Acteurs & rôles | VigiGerme® [Internet]. [cited 2017 May 3];Available from: http://vigigerme.hug-ge.ch/mesure-specifique-contact-acteurs-roles

76. Mesures spécifiques goutelettes - Généralites | VigiGerme® [Internet]. [cited 2017 May 3];Available from: http://vigigerme.hug-ge.ch/mesures-a-appliquer/mesure-specifique-gouttelettes-generalites

77. MN. Chraïti, W. Zingg. Désinfection des locaux (code rouge). 2016;

78. MN. Chraïti, B. Huttner, A. Iten. Protocole de prise en charge d'un patient porteur d'Enterobactérie productrice de carbapenemases (CPE): sous-types KPC, OXA, NDM, VIM, IMP*. 2016;

79. von Dach E, Morel CM, Murthy A, et al. Comparing the cost-effectiveness of linezolid to trimethoprim/sulfamethoxazole plus rifampicin for the treatment of methicillin-resistant Staphylococcus aureus infection: a healthcare system perspective. Clin Microbiol Infect [Internet] Available from: http://www.sciencedirect.com/science/article/pii/S1198743X1730099X

80. De Angelis G, Murthy A, Beyersmann J, Harbarth S. Estimating the impact of healthcare-associated infections on length of stay and costs. Clin Microbiol Infect Off Publ Eur Soc Clin Microbiol Infect Dis 2010;16(12):1729–35.

81. Wassenberg MWM, Kluytmans J a. JW, Box ATA, et al. Rapid screening of methicillin-resistant Staphylococcus aureus using PCR and chromogenic agar: a prospective study to evaluate costs and effects. Clin Microbiol Infect 2010;16(12):1754–61.

82. The European Committee on Antimicrobial Susceptibility Testing. Breakpoint tables for interpretation of MICs and zone diameters. Version 7.1 [Internet]. 2017;Available from: http://www.eucast.org

83. P. Lagacé-Wiens, F. Tailor and al. Evaluation of a chromogenic medium for extended-spectrum beta-lactamase (ESBL)-producing Enterobacteriaceae. 2010;

84. Zarakolu P, Day KM, Sidjabat HE, et al. Evaluation of a new chromogenic medium, chromID OXA-48, for recovery of carbapenemase-producing Enterobacteriaceae from patients at a university hospital in Turkey. Eur J Clin Microbiol Infect Dis Off Publ Eur Soc Clin Microbiol 2015;34(3):519–25.

85. Eazyplex SuperBug CRE. 2015;

86. Genie® II - Molecular diagnostics - Human medicine - Hospital and private laboratories - Axonlab Česko [Internet]. [cited 2017 May 3];Available from: http://www.axonlab.com/CZ_eng/Hospital-and-private-laboratories/Human-medicine/Molecular-diagnostics/Genie-R-II#product

87. Mori Y, Kitao M, Tomita N, Notomi T. Real-time turbidimetry of LAMP reaction for quantifying template DNA. J Biochem Biophys Methods 2004;59(2):145–57.

88. Medtech innovation briefing. Eazyplex SuperBug kits for detecting carbapenemase-producing organisms [Internet]. 2017;Available from: https://www.nice.org.uk/advice/mib94

89. Findlay J, Hopkins KL, Meunier D, Woodford N. Evaluation of three commercial assays for rapid detection of genes encoding clinically relevant carbapenemases in cultured bacteria. J Antimicrob Chemother 2015;70(5):1338–42.

90. Pittet D, Hugonnet S, Harbarth S, et al. Effectiveness of a hospital-wide programme to improve compliance with hand hygiene. Infection Control Programme. Lancet Lond Engl 2000;356(9238):1307–12.

91. Sprague E, Reynolds S, Brindley P. Patient Isolation Precautions: Are They Worth It? Can Respir J 2016;2016:e5352625.

92. Abad C, Fearday A, Safdar N. Adverse effects of isolation in hospitalised patients: a systematic review. J Hosp Infect 2010;76(2):97–102.

93. Leistner R, Gürntke S, Sakellariou C, et al. Bloodstream infection due to extended-spectrum beta-lactamase (ESBL)-positive K. pneumoniae and E. coli: an analysis of the disease burden in a large cohort. Infection 2014;42(6):991–7.