UNIVERSIDADE DE LISBOA
Faculdade de Medicina

"*Improving hand hygiene behaviour: from bench to bedside"*

Maria Daniela da Costa Pires

Orientador: Professor Doutor Didier Robert Pittet
Co-Orientador: Professor Doutor José Melo Cristino

Tese especialmente elaborada para obtenção do grau de Doutor em Medicina, especialidade de Doenças Infeciosas e Parasitárias

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As opiniões expressas nesta publicação são da exclusiva responsabilidade do seu autor.

Para o Tiago e a Heleninha

Para os meus pais e irmão

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Introduction

Healthcare associated infections and Antimicrobial resistance are public health concerns

Healthcare associated infections (HAI) are a common occurrence in healthcare and result in increased mortality, morbidity and significant costs for patients and healthcare systems [1,2].

According to the European Centre for Disease Prevention and Control (ECDC) 2016/2017 point prevalence survey (PPS), an estimated 6.5% of patients in European acute care hospitals had at least one HAI in any given day, summing up to around 4.5 million episodes each year [1]. The burden of the six most common HAI is higher than the combined burden of all other infectious diseases surveyed by the ECDC in Europe [2]. Furthermore, HAIs' burden is at least two to three times higher in low/middle- compared to high-income settings [3].

The alarming increase in antimicrobial resistance (AMR) further adds to the burden of HAI. Keeping the current pace, it is estimated that AMR will be responsible for over 10 million deaths worldwide by 2050 [4]. AMR jeopardises the accomplishments of modern medicine, including surgical procedures, organ transplants and cancer treatments, whose success relies heavily on the availability of effective antimicrobial prophylaxis and treatment of infectious complications, such as HAI [5,6]. Unfortunately, increasing levels of resistance among disease-causing pathogens have not been met by an equally fast development of new treatments for infectious diseases [7]. HAI is also a driver of AMR in healthcare, through an increased use of antimicrobials.

AMR has been recognized as one of the biggest threats to global health by the World Health Organization (WHO) [8,9] and the United Nations (UN) [10]. In 2015, the WHO global action plan on AMR was launched [9]. For the fourth time in its history, the 71st UN General Assembly, in September 2016, dedicated a high-level meeting on health topics, discussing the global threat of AMR [10]. Countries reaffirmed their commitment to the WHO global action plan to tackle AMR. Two of the five main objectives of this plan are the reduction of incidence of infections, through effective sanitation, hygiene and infection prevention measures, and the increase of knowledge in the field trough research [9].

The importance of Infection prevention and control

Infection prevention and control (IPC) is essential to tackle HAI and AMR spread in healthcare [11]. It is estimated that between 55 and 70% of HAI could be prevented by the effective implementation of IPC [12].

In 2015, the basis for an evidence-based definition of the essential elements of IPC programmes was launched [13]. Building upon that, the WHO recently issued guidelines on the Core Components of IPC, setting a new international standard on this matter [11]. Also, in accordance to the public health importance of multi-drug resistance in Gram-negative bacteria, the WHO recently performed a systematic review [14] and issued specific IPC guidelines on the subject [15].

Despite international guidance, IPC practices are often not well established and their implementation differ substantially between countries [16,17]. The wide gap at the level of IPC indicators, such as compliance with hand hygiene (HH), alcohol-based handrub (ABHR) consumption, availability of single rooms or IPC staffing are examples of the diverse and often sub-standard IPC practices in European countries [18]. Ultimately, this is reflected in the wide range of infection rates across Europe: the proportion of bloodstream infections with carbapenem resistant *Klebsiella pneumoniae* (KPC) ranges from 0% to 65% [19,20].

It remains a challenge to bridge the gap between evidence-based recommendations [13] and effective practice change [21]. Several studies have shown that written guidelines alone are not sufficient to promote adoption and implementation of evidence-based measures [22]. The best approaches to foster implementation of IPC across Europe remain to be determined [23–25].

Fostering effective IPC strategies is essential to reduce the burden of HAI and AMR. This is a public health priority.

Hand hygiene with alcohol-based handrub: a core element of IPC

The importance of hand hygiene in preventing infections has long been recognized [26]. Back in 1847, Ignas Semmelweis demonstrated for the first time that cleansing contaminated hands with an antiseptic solution reduced infections and provided safer obstetric care [27]. Interestingly, albeit Semmelweis had considerable evidence supporting the use of chlorinated lime solution as an antiseptic, he was not successful implementing the strategy [28].

Almost one hundred and fifty years elapsed until the scientific community

readdressed the need to create a vision focused on hand hygiene improvement and its direct effect on patient safety. Indeed, the importance of hand hygiene in healthcare re-emerged in the early 1960s with the landmark study on the prevention of *Staphylococcus aureus* (*S. aureus*) transmission by hand washing [29]. However, it was only in 1985 that the Centers for Disease Control and Prevention (CDC) issued the first national recommendation endorsing hand washing as the most important measure to prevent nosocomial infections [30].

The 1990s witnessed the beginning of a thorough implementation of Semmelweis' hand hygiene concept: the use of ABHR was identified as an alternative to hand washing with soap and water [31].

Around the same time, the landmark studies that would establish evidence on the advantages of using ABHR and on the effectiveness of a hospital-wide multimodal strategy to implement hand hygiene and reduce hospital-acquired infections, were being conducted at the Geneva University Hospitals (HUG) [32,33]. The so-called Geneva model [33], successfully pilot-tested in 4 countries [34], would became the model endorsed by the WHO guidelines and implementation tools (2009) [35,36]. These studies were also the stepping stone for transatlantic collaboration and the publication of the CDC guidelines for Hand Hygiene in Health-Care Settings (2002) [37].

In 2005, the WHO First Global Patient Safety Challenge *'Clean Care is Safer Care'* was launched under the expert advise of Professor Didier Pittet [38]. Ministers of Health were invited to pledge support to reduce HAIs trough the implementation of hand hygiene according to the multimodal strategy developed in Geneva.

In 2009, the worldwide campaign promoting HH in healthcare (http://www.who.int/gpsc/en/) WHO "*Save Lives: Clean Your Hands*", marked a significant shift towards the globalization of the hand hygiene with ABHR concept [35,36,39]. As part of this campaign, the 5th of May marks the WHO global hand hygiene day [6,40–42].

So far, 139 of the 194 WHO Member States have made formal statements pledging their support to *'Clean Care is Safer Care'*, more than 18,000 healthcare facilities in 179 countries have committed to improve HH and 50 countries have performed national campaigns (https://www.who.int/infection-prevention/campaigns/clean-hands/ccsc_ten-years/en/).

Several factors facilitated the worldwide dissemination of the ABHR hand hygiene concept. The development of the WHO ABHR formulation was essential for the implementation effort in the low and middle-income countries [43]. Indeed, it allows the

pharmacies of hospitals to produce ABHR formulation at a very low cost and with local products. In an unprecedent move, a positive "fatwa" was obtained from religious leaders in order to allow Muslim healthcare workers (HCWs) to use alcohol in their hands [44]. In 2011, an educational video was published at the New England Journal of Medicine (NEJM) in order to disseminate the "My 5 moments" concept [45]. The video was made freely available in 12 languages (https://www.who.int/gpsc/5may/hand_hygiene_video/en/). It was recently considered the NEJM's most viewed video ever [46].

A recent systematic review and meta-analysis evaluated the efficacy of interventions to improve hand hygiene, including secondary clinical outcomes [47]. Authors showed an association between an increase in hand hygiene compliance and a reduction in HAIs in 19 studies. Three high quality studies [48–50] found strong evidence that increased use of alcohol-based handrub was associated with a reduction of HAI in general and a reduced incidence of healthcare-associated methicillin-resistant *S. aureus* (MRSA) infections specifically. This probably reflects the contribution of hand hygiene in reducing cross-transmission [47]. Infections by Gram-negative pathogens may be less affected, as these are more often endogenous. However, hand hygiene plays a critical role in outbreak situations [51].

Finally, by reducing the burden of HAI, hand hygiene lowers antibiotic use in hospitals, and, as such, indirectly contributes to AMR containment in general. A systematic review performed by WHO underscores these findings [52].

WHO multimodal strategy to improve hand hygiene

Great efforts have been made to improve hand hygiene compliance among HCWs worldwide [35,36]. The WHO guidelines were issued with a guide to the implementation of the Multimodal strategy to improve hand hygiene. This includes extensive, comprehensive and freely available materials for assisting healthcare institutions in the implementation of hand hygiene best practices (https://www.who.int/infection-prevention/tools/hand-hygiene/en/) [36]. This strategy proved highly successful in the first pilot countries [34] and has been replicated worldwide [47].

The IPC programme of HUG proposed a model with 5 essential steps needed for the cross-transmission of pathogens though the hands of healthcare workers [53]. The model was used to identify 5 key moments in which a hand hygiene action could stop cross-transmission during clinical care. This was summarized in the widely used "My 5 moments for hand hygiene" or "When to handrub" poster (Figure 1) [54].

Another very important aspect of the strategy is the guidance on the performance of the hand hygiene action, the so-called "How to handrub" (Figure 1). This poster depicts the WHO recommendations on the hand hygiene action: 6-step technique, palmful of ABHR and 20 to 30 seconds of hand friction.

These posters became the most visible aspects of the WHO campaign to implement hand hygiene.

FIGURE 1. World Health Organization "My 5 moments for hand hygiene" and "How to handrub" posters.

The multimodal strategy to improve hand hygiene includes 5 components: System change, Training and Education, Evaluation and Feedback, Reminders in the workplace and Institutional safety climate. System change focuses on the availability of ABHR at the point of care. Training and Education emphasises the need to regular train HCWs on the "My 5 moments" concept ("When to handrub") and the hand hygiene gesture ("How to handrub"). Evaluation and Feedback preconizes regular evaluation of HH practices as well as the feedback of information. Reminders in the workplace consists in providing cues to HCWs throughout the institution. Safety culture consists in creating an environment at the healthcare institution focused on patient safety [36].

Importantly, a recent systematic review and meta-analysis showed that the increase in hand hygiene compliance and reduction in health care–associated infections are significantly higher when all elements of the multimodal strategy are applied together [47].

The WHO developed a hand hygiene self-assessment tool, allowing hospitals to

evaluate their status with regards to the implementation of each of the components of the strategy [55,56]. A WHO worldwide survey performed in 2011 and 2015 showed that Evaluation and Feedback and Institutional safety climate are amongst the less implemented components of the strategy [57].

Evaluation and Feedback: challenges

Evaluation of practices and feedback are essential components of the WHO Multimodal strategy to improve hand hygiene [36]. First, direct observation of hand hygiene practices yields performance data that permits to improve and adapt the hand hygiene implementation action plan of the institution. Second, the direct observation method also strongly contributes to education and training of frontline healthcare workers. Third, it underwrites institutional safety climate through routine feedback of data, target setting, and the regular presence of trained observers in the wards, reinforcing the institutional commitment to hand hygiene [58].

The WHO's method for direct observation of hand hygiene practices was developed at the University Hospitals of Geneva. Trained IPC practitioners perform regular, open, unobstructive observation of HCWs compliance with the "5 moments for hand hygiene" during clinical care. This method contemplates the immediate and direct performance feedback to HCWs. In addition, it also considers other forms of restitution of data, including aggregated feedback to the heads of units and hospital management. Compliance is calculated by dividing the number of hand hygiene actions performed when an opportunity exists by the number of hand hygiene opportunities according to the "My 5 moments" concept [59]. This method also allows for the direct observation of the hand hygiene action ("How to handrub"), but there is no current validated method to monitor it and calculate the adherence to its correct performance.

However, the WHO direct observation method is time-consuming, costly and prone to bias [60,61]. The most common critic to the hand hygiene data generated is the overestimation of compliance due to the presence of the Hawthorne effect. The Hawthorne effect is the change of behaviour of an individual when he/she is aware of being observed. It has not been possible thought to estimate the magnitude of this effect, as it seems variable according to the conditions of the observation [62,63].

These gaps have fuelled research and new technologies have emerged to complement the WHO direct method of HH observation. In this context, the interest on electronic

monitoring devices is on the rise [61]. Besides monitoring, many electronic devices have the potential to remind HCWs about hand hygiene and to provide feedback [61].

Wall mounted electronic ABHR dispensers allow for an accurate monitoring of the volume of ABHR dispensed in each HH event, and the frequency of ABHR dispensing events. Other systems have elegantly combined events recorded by ABHR dispensers with HCWs movements, namely room entrance and exit (taken as surrogate markers of WHO moments 1 and 4/5, respectively) [61]. In spite of all of these and other exciting advances, none of the systems available today are able to provide data on hand hygiene compliance, simply because it is nearly impossible for an automated system to accurately detect opportunities for hand hygiene [53].

There are also electronic systems developed to evaluate the quality of the HH action, based on ultraviolet light (Hand-in-Scan™, HandInScan Ltd., Hungary) [64] and video measurement technology (SureWash™, Glanta Ltd., Ireland) [65]. They have been successfully used for HH training purposes, but are not intended to continuously monitor the quality of HH action in the daily clinical practice.

Hand hygiene: campaign fatigue

Hand hygiene compliance remains sub-optimal, despite availability of national and international guidance and best efforts of IPC practitioners to improve it. It is estimated that HCWs perform HH according to the "My 5 moments" in only 40% of the time [66]. New strategies are needed to improve these figures [67].

Recently, several new ideas were tried to fight campaign fatigue and keep HCWs and IPC practitioners motivated in improving HH. Some examples of innovation are the hand hygiene relay [68] and the Train-the-Trainers programme [69]. In Hong Kong, the first hand hygiene sanitizing relay was conducted [70]. HCWs sequentially performed the "How to handrub" in a human chain around the hospital. Each hand hygiene action was evaluated and approved by a jury before the next HCW could proceed in the chain. This strategy did improve HH practices and was followed by a worldwide movement to beat the World Guinness Record set out by Hong Kong [67].

The IPC programme of the HUG developed an international 3-day course "Train-the-Trainers" aimed at fostering country capacity on hand hygiene promotion. This course addresses the gaps on training and homogenization of HH practices and aim at creating a domino effect in HH promotion within each country [69].

“How to handrub”

Little attention has been devoted to the adequate performance of the hand hygiene action (“How to handrub”), despite being as important as respecting the correct timing (“When to handrub”) in preventing cross transmission of microorganisms [71]. The appropriate performance of the hand hygiene action is crucial to ensure its antimicrobial efficacy [72–74,71].

Contributing factors to the less attention given to the “How to handrub” may include the lack of clear evidence-based guidance on its performance and the absence of validated tools to conduct its monitoring and foster its improvement amongst HCWs.

The WHO hand hygiene guidelines address several aspects related to the quality of the hand hygiene action [35,36]. A specific 6-step technique is recommended in the “How to handrub” poster (Figure 1). However, this technique was designed to ensure homogenous hand-surface coverage by applied hand hygiene agents [75]. It was not developed to be user-friendly or to address the most contaminated parts of hands. In addition, less precise information exists in the WHO guidelines regarding the volume of ABHR ("palmful") and duration of hand friction (“20 to 30 seconds or until dry”) required to perform an optimal hand hygiene action.

The CDC guidelines for hand hygiene are imprecise, mentioning that “if hands are dry before 10 to 15 seconds, an insufficient amount of ABHR has been used” [37]. There is no specific mention to specific a technique nor on the duration of hand rubbing.

Products entering the market in Europe are tested according to the European Norm 1500 (EN 1500) [76]. This test is based on comparisons between a gold standard, that consists in performing hand hygiene twice with 3 mL of isopropanol 60% v/v and 30 seconds of hand friction according to the WHO technique, and the new product, used as per manufactures’ specifications (the majority of which are tested with 3 mL, 30 seconds of hand rubbing and WHO technique). The reduction in bacterial counts achieved in artificially contaminated hands with *Escherichia coli* (*E.coli*) is then compared between the new product and the gold standard. If the new product achieves a 0.6 $\log_{10}$ non-inferiority margin in terms of reduction of bacterial count, then it is considered acceptable and can be commercialized.

The WHO, CDC and EN 1500 do not take into account what is the contamination usually present on the hands of healthcare workers during clinical care, what are the most contaminated parts of hands during clinical work or the correlation between hand contamination and the likelihood of cross-transmission. These concepts deserve further

investigation. In fact, we do not know what is the minimum reduction of bacterial load produced by a hand hygiene action to prevent cross-transmission.

Compliance with the World Health Organization "How to handrub" action is suboptimal. In a study performed at the Geneva University Hospitals, no HCW performed adequately the 6-step technique recommended by the WHO [55]. In another study in Basel, Switzerland, only 8.5% of HCWs performed correctly the 6-step WHO technique [77]. In addition, the volume of ABHR used by HCWs during clinical practice is very low, around 1 mL [78,79]. The duration of hand friction is also much inferior the recommended 20 to 30 seconds, more likely bellow 10 seconds [80].

Simplifying the hand hygiene action could help improving practices. Indeed, lack of time is repeatedly identified as a major factor negatively influencing adherence to hand hygiene [32]. The lack of time influences how much disponibility HCWs have to dispend on hand hygiene, which in turn influences the performance of the technique, the duration of hand friction and the volume used.

There is an emerging research interest on the "How to handrub". In fact, our group recently demonstrated the importance of the volume of ABHR in the antimicrobial efficacy of the HH gesture [81]. In addition, we have demonstrated for the first time that the hand size influences the antimicrobial efficacy of the hand hygiene action. HCWs with larger hands need more volume of ABHR than those with smaller hands in order to achieve the same antimicrobial efficacy.

According to other recently published studies, the hand hygiene technique is also a major determinant of the antimicrobial efficacy of hand hygiene actions. However, there is still controversy on the need to perform the WHO 6-step technique. Some studies point to a possible reduction to a 3-step technique, while others report a loss in antimicrobial efficacy with this simplification [82].

In addition, the optimal duration of hand rubbing remains to be determined. Indeed, there is no strong evidence backing WHO's 20 to 30 seconds recommendation of hand friction.

Using a wearable device to monitor and improve the HH action quality: SmartRub®

SmartRub® is a novel, wearable device capable of monitoring and providing real-time personalized feedback on the hand hygiene action (duration of hand friction and volume of ABHR provided) to HCWs during clinical care (Figure 2). This wearable device includes

a wristband and a pocket-size bottle of ABHR. SmartRub® was developed by an investigator-initiated partnership, including the IPC programme of the University Hospitals of Geneva, the "Haute école du paysage, d'ingénierie et d'architecture de Genève » and the start-up IQati™.

As we previously discussed, there is not enough evidence to back the WHO recommendations on "How to handrub". Therefore, in order to *program* SmartRub® monitoring and feedback, we first run a series of laboratorial-based experiments to define the parameters of an optimal « How to handrub ».

Finally, we tested the effect of SmartRub® on a randomized clinical trial aiming at improving the quality of HH and the compliance with the "5 moments".

As mentioned previously, SmartRub® monitors and provides real-time personalized feedback during daily clinical practices on the volume of ABHR and duration of hand friction, to individual HCWs. We expected this feedback to improve the quality of the HH action ("How to handrub"). Most importantly, we expected that the experience of having the HH continuously monitored would also lead to an improvement in the compliance with the "5 moments" ("When to handrub") [60].

FIGURE 2. SmartRub® device.

Aims

The **aims** of this doctoral project are:

1) To optimize the WHO recommended hand hygiene action ("How to handrub");
2) To improve the quality of the hand hygiene action ("How to handrub") performed by HCWs during daily clinical activities;
3) To improve compliance of HCWs with the "My 5 moments" of hand hygiene ("When to handrub").

This doctoral project aims to answer the following **research questions**:

1) What is an "adequate" hand hygiene action in terms of antimicrobial reduction on healthcare workers' hands?
2) Can the current WHO "How to handrub" hand hygiene action recommendation be optimized, maintaining an adequate antimicrobial action?
3) Does an optimized recommendation of hand hygiene action, delivered in real-time by a novel monitoring wearable device, improve the quality of the hand hygiene action performed by HCWs during daily clinical activities?
4) Does an optimized recommendation of hand hygiene action, delivered in real-time by a novel monitoring wearable device, improve the compliance with the "5 moments" for hand hygiene ("When to handrub")?

The **hypothesis** tested at this doctoral project are:

1) We hypothesize that the recommended WHO "How to handrub" hand hygiene action can be optimized, while maintaining its antimicrobial efficacy;
2) We hypothesize that in a population of healthcare workers performing clinical activities, an optimized hand hygiene action recommendation delivered in real-time by a monitoring wearable device, will improve the quality of the hand hygiene action ("How to handrub");
3) We hypothesize that in a population of healthcare workers performing clinical activities, an optimized hand hygiene action recommendation delivered in real-time by a monitoring wearable device, will improve hand hygiene compliance with WHO's "5 moments" ("When to handrub").

The **specific objectives** are:

1) To define a microbiological concept of "safe hands" by establishing the threshold of hand contamination that decreases the likelihood of bacterial cross-transmission between hands (Chapter 1);
2) To evaluate the effect on the antimicrobial efficacy of the hand hygiene action of:
    a. Modifying the sequence of the WHO 6-step technique – by rubbing the fingertips first (Chapter 2);
    b. Shortening the duration of hand rubbing from 30 seconds down to 15 seconds (Chapter 3);
    c. Combining the modified sequence (a.) and shorter duration of hand rubbing (b.) with a volume of ABHR customized for hand size (Chapter 4);
3) To evaluate if using a novel monitoring wearable device (SmartRub®) providing real-time customized feedback about the quality of hand hygiene action enhances the quality of the HH action (Chapter 5) and improves the compliance to the WHO 5 "moments" ("When to handrub") amongst HCWs performing patient-care activities (Chapter 5).

In order to achieve objectives 1 and 2 we performed a series of laboratorial studies with volunteers based on the EN 1500 [76] at the IPC laboratory of the University Hospitals of Geneva. Each study is described in detail in Chapters 1 to 4 (chapters adapted from peer-reviewed publications) [74,83–85].

In order to achieve objective 3 we performed a stepped wedge cluster randomized clinical trial at the Geriatric Hospital of the University Hospitals of Geneva. This trial is described in detail in chapter 5, adapted from a manuscript in preparation for submission.

Chapter 1

Assessing the Likelihood of Hand-to-Hand Cross-Transmission of Bacteria: An Experimental Study

Bellissimo-Rodrigues F*, Pires D*, Soule H, Gayet-Ageron A, Pittet D.

Infect Control Hosp Epidemiol 2017;38:553–8. https://doi.org/10.1017/ice.2017.9 [74]

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Introduction

An "adequate" HH action could be defined as one that turns contaminated hands into "safe hands". "Safe hands" are hands that do not serve as vehicles for cross transmission of microorganisms between patients or between the environment and patients. However, we do not know what is the threshold of hand contamination bellow which cross transmission is unlikely. Thus, it remains uncertain what is the magnitude of reduction of hand contamination required to consider a hand hygiene action as "adequate".

This information is essential for future research aiming at the improvement of the "How to handrub". Having a meaningful antimicrobial goal for a hand hygiene action could allow modifying the "How to handrub" technique while ensuring that the goal is met.

In addition, having an antimicrobial goal could help improving the international norms used for the development, regulation and comparison of products applied for hand hygiene (plain or antimicrobial soaps and alcohol-based handrubs) [86]. In fact, the most widely used tests to validate the microbiological efficacy of hand hygiene products, the European Norm 1500 (EN 1500) and the American Society for Testing and Materials recommendation E2755-10 (ASTM E2755-10) [76,87]), use arbitrary rather than evidence-based endpoints.

Specific objective #1

To evaluate the likelihood of *E. coli* cross-transmission from one artificially contaminated hand to another person's hand, as a function of the burden of contamination. We aimed to define a threshold in the level of hand contamination below which cross transmission is not expected in hand-to-hand contact.

Methods

Study setting, participants and eligibility criteria

We conducted a laboratory-based experimental study at the microbiology laboratory of the IPC Programme of HUG. Participants were HCWs with several years of training in infection control, with particular expertise in hand hygiene. All participants had short nails according to international recommendations [35,37]. Exclusion criteria included any skin disorder, current use of systemic or topical antimicrobial agents, use of immunosuppressive treatment, or presence of artificial fingernails.

The study was approved by the institutional review board of HUG and all participants signed written informed consent prior to the experiment.

Experimental design

All experiments were based on the European Norm 1500 standard [76]. *E. coli* was chosen because it is the bacteria used in the EN 1500 and it is also frequently associated with HAI.

In order to quantify hand contamination with *E. coli* associated with hand-to-hand transmission, we designed an experiment that consisted in 4 trials and included two HCWs. In each trial, one HCW (transmitter) had both hands contaminated with *E. coli* before holding with his right hand another HCW's right hand (host) for 1 minute (transmission procedure; Figure 3A). At the same time the transmitters' left hand was sampled (Figure 3A). After 1 minute, the host's hand that was being held was sampled to assess the degree of cross-transmission (Figure 3B). This trial was performed 4 times in each experiment with increasing concentrations of bacteria in the contamination suspensions. Subsequently, on the same experimental day, HCWs' roles (transmitter *versus* host) were swapped and the experiment repeated. In total, 6 HCWs participated in the study. All pairwise combinations of HCWs were performed, for a total of 30 experiments and 120 unique trials.

A. **B.**

FIGURE 3. Experimental study design. **A.** Hands position during the 1 minute transmission procedure: while the right hand of the transmitter holds the right hand of the host, the left hand is being sampled according to the fingertips method. **B.** After the one minute transmission procedure, the hand of the host that had been in contact with the hand of the transmitter is also sampled according to the fingertips method.

Contamination, transmission and sampling procedures

The reference strain *E. coli* ATCC 10536 was grown according to EN 1500 to obtain 4 homogeneous suspensions containing increasing concentrations of bacteria: 10^3 colony forming units (cfu)/mL, 10^4 cfu/mL, 10^5 cfu/mL, and 10^6 cfu/mL.

In brief, prior to each experiment and between each individual trial, participants were asked to wash their hands with 5 mL of a plain liquid soap for 1 minute and to handrub with 3 mL of 2-propanol 60%. All procedures were performed according to the 6 specific steps of the WHO technique. Participants were also instructed to avoid any accidental contamination during the entire study procedure.

The hands of the HCW acting as transmitter were artificially contaminated by being immersed into the bacterial suspension up to the mid-carpals for 5 seconds. Then, they were held up to dry for 3 minutes after which they were considered ready for a transmission episode.

The position to simulate a transmission episode was as follows: the dorsal side of the fingers of one participant (transmitter) are placed in the palm of the other participant (host) with fingers interlocked during 1 minute (Figure 3A). During the 1 minute-transmission trial, the left hand of the transmitter was sampled according to the procedure described in the EN

1500 (fingertips method). After the transmission trial, the hand of the host that had been in contact with the transmitter was sampled in the same way.

The fingertips method of microbiological sampling consists in having all 5 fingertips rubbed in a petri dish of 90 mm diameter containing 10 ml of tryptone soya broth (TSB) for one minute.

Dilution and plating of samples

Each sample was studied in 4 different dilutions to accurately estimate bacterial counts. While dilutions for the transmitter samples ranged from 10^{-4} to 10^{-7}, they ranged from 10^{-1} to 10^{-5} for the host samples. To maximize bacterial detection at lower levels of transmission, host samples were also filtered with EZ-Pak® Membrane Filters (Merck Milipore, France). The dilution and filtration figures derived from preliminary tests performed when setting the study design.

After dilution, 1 mL samples were distributed in tryptic soy agar plates within 30 minutes of recovery and incubated at 36±1ºC for 24 to 48h. After filtration, the filters were also incubated in tryptic soy agar plates at 36±1ºC for 24 to 48h.

Hand size calculation

Hand surface area of each participant was calculated and categorized as small (area ≤375 cm^2), medium (376–424 cm^2), or large (≥425 cm^2), as described elsewhere [81,88]. The median hand surface area was 434 cm^2 [standard deviation (SD) 97 cm^2]. According to hand size categorization, 2 participants had small hands, 2 medium, and 2 large.

Study outcomes and statistical analysis

E. coli cfu were counted by visual inspection of each plate, adjusted for the corresponding dilution factor, and converted to $\log_{10}$.

For each trial, we evaluated *E. coli* transmission qualitatively and quantitatively in relation to the contamination in the left hand of the transmitter. For qualitative evaluation, *E. coli* cfu counts detected on the hand of the host were transformed into a dichotomous variable (transmission was defined if at least 1 cfu was detected on the hand of the host; otherwise it was considered no-transmission). In the quantitative evaluation, we have described the median and interquartile range (IQR) cfu of *E. coli* retrieved in the hands of the host.

The primary outcome was the probability of transmission of at least 1 cfu of *E. coli* to the host's hand as a function of the *E. coli* counts on the transmitter's left hand.

We used a mixed logistic regression model with a random effect on the subject to assess the association between the transmission and the *E. coli* count on the hands of the transmitter. We adjusted the model for the hand size categories of the transmitter and sex. We also estimated the probability of cross-transmission predicted by the model and presented it as a function of the *E. coli* count on the hands of the transmitter.

Statistical analyses were performed using Stata® version 14 (StataCorp). Statistical significance was defined as $p<0.05$ (2-sided).

Results

The mean *E. coli* counts measured in the left hand of the transmitter after immersion in *E. coli* suspensions of 10^3 cfu/mL, 10^4 cfu/mL, 10^5 cfu/mL and 10^6 cfu/mL were 1.3 $\log_{10}$ (SD 0.57), 2 $\log_{10}$ (SD 0.63), 3.1 $\log_{10}$ (SD 0.76) and 4 $\log_{10}$ (SD 0.49), respectively. From the 120 transmission trials performed 8 (6.5%) were non-interpretable due to overgrowth of coagulase-negative staphylococci from the participants' own microbiota. Thus, study findings are based on 112 trials.

The total number of *E. coli* retrieved from the hand of the host in relation to the total bacteria count retrieved from the hand of the transmitter is shown in Table 1 and Figure 4. We found no episodes of cross transmission when the contamination on the hands of the transmitter was bellow 1 $\log_{10}$ cfu.

The probability of cross-transmission obtained from the model as a function of *E. coli* count on the hands of the transmitter is shown in Figure 5. By multivariate analysis, the transmission to the hands of the host was significantly ($p<0.001$) associated with the bacterial count on the hands of the transmitter after adjustment for sex and transmitter hand size (Table 2).

TABLE 1. Likelihood of *E. coli* transmission* and *E. coli* cfu retrieved from the hands of the host, according to the level of contamination on the hands of the transmitter.

E. coli (log_{10} cfu) counts on the hands of the transmitter (n)	Transmission events (≥ 1 *E. coli* cfu recovered from the hands of the host), n (%)	Number of *E. coli* (cfu) recovered from the hands of the host, median, min-max (IQR)
< 1 log_{10} (8)	0 (0%)	0, 0-0 (0-0)
≥ 1 log_{10} to < 2 log_{10} (29)	6 (23.1%)	0, 0-4 (0-0)
≥ 2 log_{10} to < 3 log_{10} (26)	8 (30.8%)	0, 0-13 (0-1)
≥ 3 log_{10} to < 4 log_{10} (27)	17 (63%)	1, 0-398 (0-6)
≥ 4 log_{10} (22)	20 (90.9%)	25, 0-1259 (8-200)

*Bacterial transmission is considered if at least 1 cfu of *E. coli* is found on the hand of the host.

cfu: colony-forming units; IQR: inter-quartile range; log: logarithmic.

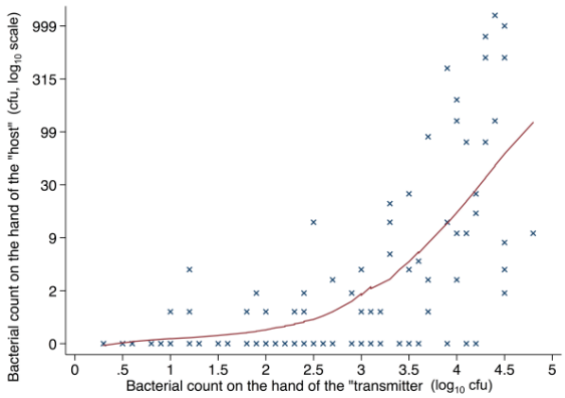

FIGURE 4. Cross-transmission of *E. coli* from the hands of transmitter to host. Bacterial count transmitted to the hand of the host*, according to the original level of contamination on the left hand of the transmitter; n=112 paired results.

cfu: colony-forming units; log: logarithmic.

*The logarithm of zero is not mathematically defined, but we consider important to show in the graph when the transmission experiment resulted in zero bacteria on the hands of host. Thus, we have added the value of 1 to all results of the y variable and corrected for this artefact in the shown y-axis labels.

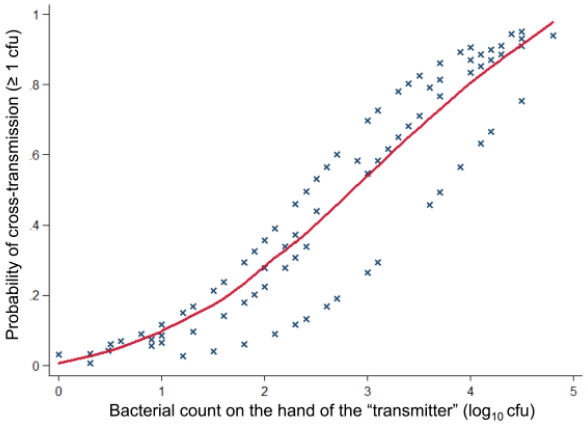

FIGURE 5. Probability of cross-transmission of *E. coli*. Likelihood of *E. coli* cross-transmission (at least 1 cfu) to the hand of the host, according to the original level of contamination on the left hand of the transmitter; n=112 paired results. The probability of cross-transmission predicted by the model is presented as a function of the bacterial count on the left hand of the transmitter.

cfu: colony-forming units; log: logarithmic.

TABLE 2. Likelihood of *E. coli* cross-transmission* (at least 1 cfu) according to *E. coli* cfu count on the transmitter hand after adjustment for sex and transmitter hand size; multivariate analysis.

	Odds ratio	**95% CI**	**p-value**
Bacterial count on the transmitter hand			<0.001
(ref. ≤ 1 $\log_{10}$)			
>1 & ≤ 3 $\log_{10}$	8.22	0.98-68.80	0.052
>3 & ≤ 3.5 $\log_{10}$	22.59	2.12-240.77	0.010
>3.5 & ≤ 4 $\log_{10}$	163.20	11.40-2335.52	<0.001
>4 $\log_{10}$	212.60	15.56-2905.16	<0.001
Hand size (ref. small)			0.219
Medium	0.22	0.04-1.22	0.083
Large	0.28	0.03-2.51	0.255
Women (ref. men)	0.18	0.03-1.19	0.075

[a] From a mixed logistic regression model with a random effect on the intercept.

*Bacterial transmission is considered if at least 1 cfu of *E. coli* is found on the host hand.

cfu: colony-forming units; CI: confidence interval; log: logarithmic; OR: odds ratio; ref.: reference.

Discussion

In the present study, we developed an experimental model to understand how *E. coli* load on hands affects the risk of cross-transmission following a single 1-minute hand-to-hand contact. We demonstrated a direct relationship between *E. coli* load on HCWs' hands and the likelihood of cross-transmission. We also observe that, under these experimental conditions, at least 1 $\log_{10}$ cfu of *E. coli* must be present on HCWs' hands in order to be transmitted to another individual.

Notably, an *E. coli* count in transmitter's hand of $> 1 \,\&\, \leq 3$ $\log_{10}$ was associated with an odds ratio of 8.22 of transmission, with a p-value of 0.052. The relatively small number of trials per *E. coli* concentration category probably explains the large confidence intervals and might have interfered with our ability to detect the magnitude of an increase in transmission for that contamination, as compared with <=1 $\log_{10}$. Therefore, we assumed the bacterial count of 1 $\log_{10}$ cfu of *E. coli* as the threshold below which cross transmission is not expected in hand-to-hand contact.

Importantly, our findings need to be further explored and confirmed by studies specifically addressing low concentration ranges, as well as other potential pathogens, like *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Enterococcus* spp, fungi or viruses. Should future studies replicate the findings presented here, 1 $\log_{10}$ cfu could be considered as a microbiological endpoint for the development and regulation of products intended to be used for hand hygiene in healthcare. This hypothetical scenario would call for a revision of the microbiological endpoints established by both the European and the American norms [EN 1500 [76] and ASTM E2755-10, [87] respectively] for evaluating the antimicrobial efficacy of hand hygiene products.

Our model could also be used to increase the understanding of the transmission dynamics of other pathogens outside the healthcare setting. It is considered that hand-to-hand contact plays a relevant role on Influenza virus transmission, leading WHO and the United States Centers for Disease Control and Prevention to recommend preventive measures such as hand hygiene and cough etiquette by any person to stop the spread of the virus during flu seasons [89,90].

Our study has several limitations, in addition to what was already discussed above. First, our results are based on the premise that both transmitter hands were contaminated with the same inoculum during the experimental procedure. This assumption has been used in prior studies [81,91]. We performed an additional experiment to compare right and left hand contamination with *E. coli* (10^8 cfu/mL) and found no significant differences

(unpublished data). Secondly, our results cannot be generalized to bacteria other than *E. coli* and should stimulate further research. However, we must point the fact that testing and validation of hand hygiene products antimicrobial efficacy usually relies on *E. coli* and *Serratia marcescens* [76,87]. Third, we used a hand-to-hand contact time of one minute, even though the average hand-to-skin contact in clinical practice is much shorter (i.e., in the order of a few seconds only). This choice aimed to account for the effect of repeated brief hand-to-skin contacts between HCWs and a patient [35,36,54]. Forth, our definition of transmission depended on bacterial quantification by culture, and we are therefore limited by sensitivity of culture-based technique for bacterial detection. Finally, we did not control for other variables known to influence transmission of pathogens, such as contact pressure and humidity.

Conclusions

In conclusion, we found that there is a direct relationship between the bacterial load present on hands and the risk of cross-transmission following a single hand-to-hand contact. Importantly, we observed that a minimum contamination of 1 $\log_{10}$ cfu of *E. coli* must be present for transmission to occur. This threshold might be useful for the development of an evidence-based "safe hands" microbiological concept, which can then be applied in research to investigate new techniques of infection prevention and control

Chapter 2

Revisiting the WHO "How to Handrub" Hand Hygiene Technique: Fingertips First?

Pires D*, Bellissimo-Rodrigues F*, Soule H, Gayet-Ageron A, Pittet D.

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Introduction

The "How to handrub" technique is one of the key components of the multimodal strategy to implement hand hygiene from the WHO. It includes 6 steps that healthcare workers should perform to clean their hands appropriately with alcohol-based handrub (Figure 1) [35]. However, recent studies recognized that few HCWs perform the complete technique [55,77]. Notably, rubbing the fingertips is the last step of the WHO 6-step technique (Figure 1), in spite of the fact that fingertips are the most heavily colonized hand area during clinical care [92].

Specific objective #2a

We aimed to evaluate whether modifying the sequence of the WHO technique, by performing "rubbing the fingertips" as the first step instead of the sixth, would result in greater bacterial reduction on HCWs' hands.

Methods

In an experimental study at the University of Geneva Hospitals, 16 HCWs with expertise in hand hygiene performed the standard WHO "How to handrub" technique (Figure 1) and a modified version (WHO "fingertips-first"). The WHO "fingertips-first" technique consisted of the same 6 steps of the standard WHO technique, but with the 6th step – rubbing of the fingertips- being performed first; the remaining sequence was unchanged. HCWs were supervised by 2 senior infection control experts.

Experiments were performed based on the European Norm 1500 standards [76]. Briefly, HCWs washed their hands with plain soap and water. Afterwards, hands were immersed until mid-metacarpals for 5 seconds in a bacterial suspension containing between 2.0×10^8 and 2.0×10^9 cfu/mL of the reference strain *E. coli* ATCC 10536 (contamination procedure). Hands were then left to dry for 3 minutes. Baseline microbiological sampling

consisted in rubbing the fingertips of the dominant hand in a petri dish containing 10 ml of tryptone soya broth for one minute.

After baseline sampling, the contamination procedure was repeated twice, each time followed by one of the 2 hand rubbing techniques under evaluation. All 16 volunteers performed both techniques, 8 started with the standard WHO technique and 8 with the WHO "fingertips-first". Regardless of the specific technique, hand rubbing was performed for 30 seconds using 3 mL of isopropanol 60% (v/v; i.e. EN 1500).

After each hand rubbing procedure, fingertips were sampled as described above. For each of the 4 dilutions studied (10^{-1} to 10^{-4}), a 1 mL sample was spread over the surface of a tryptone soy agar plate, subsequently incubated at 36°C ±1°C for 48 h. Bacterial cfu were quantified by visual inspection.

The primary outcome was the difference in cfu retrieved from hands at baseline and after each of the 2 different techniques ($\log_{10}$ reduction from baseline).

Hand surface areas were calculated and categorized as "small" (surface area ≤375 cm^2), "medium" (376–424 cm^2), or "large" (≥425 cm^2) [81,88].

A generalized linear mixed model with a random effect on the intercept was used to assess bacterial $\log_{10}$ reductions from baseline. The hand rubbing technique (WHO "fingertips-first" *vs*. standard WHO) was the main predictor. We assessed whether the effect of the technique differed according to hand size category by testing the interaction between the two variables. Finally, the model was adjusted for hand size and sex.

Statistical analyses were performed using Stata® version 14 (StataCorp). Statistical significance was defined as P <.05 (2-sided).

Results

Among the 16 participants, there were 7 nurses (43.8%) and 9 medical doctors/pharmacist/biologist (56.2%). Ten (62.5%) were women. Four had small (25.0%), 6 (37.5%) had medium and 6 (37.5%) had large sized hands.

Overall, *E. coli* cfu $\log_{10}$ reduction was significantly higher when performing the "WHO fingertips-first" compared to the standard WHO technique (Table 3). Findings were confirmed across the three hand size categories with no significant difference between the three groups (interaction term, p=0.587).

After adjustment for hand size and sex, the mean reduction of bacterial concentration was 0.77 $\log_{10}$ greater (95% Confidence interval (CI): 0.27-1.26, p=0.002) following the application of the "WHO fingertips-first" when compared to the standard WHO technique.

Bacterial reduction was not significantly associated with sex (p=0.142) or hand size category (p=0.199).

TABLE 3. Reduction of *E. coli* counts from mean baseline values depending on the sequence of the hand rubbing technique (log_{10} values; mean±SD (median)).

	Mean baseline count (n=16)	**Standard WHO technique (n=16)**	**WHO "fingertips-first" technique (n=16)**	**P-value**
Globally	6.18 (±0.86, 6.35)	2.68 (±1.48, 2.85)	3.44 (±1.33, 3.20)	<0.001[a]
By hand size				0.587
Small	5.30 (±0.85, 5.3)	3.40 (±1.83, 3.40)	3.95 (±1.84, 4.25)	<0.001[b]
Medium	6.22 (±0.80, 6.4)	2.57 (±1.62, 3.05)	3.10 (±1.59, 2.70)	<0.001
Large	6.73 (±0.42, 6.7)	2.30 (±1.17, 2.05)	3.45 (±0.60, 3.35)	0.001

[a] From a mixed linear model with a random effect on the intercept; [b] From a mixed linear model with a random effect on the intercept and an interaction between the sequence and hand size category.

Discussion

The WHO standard "How to handrub" technique was designed to ensure homogenous hand-surface coverage by applied hand hygiene agents. It was not developed to be user-friendly or to address the most contaminated parts of hands. Notably, when monitored, HCW compliance with all 6 steps is very low [55,77].

A recent study showed that the standard WHO 6-step technique reduced bacterial contamination on HCWs' hands more effectively than the Centers for Disease Control and Prevention's 3-step technique [93]. However, another evaluation performed in laboratory conditions showed that a 3-step technique was comparable to the standard WHO 6-step technique [94]. While the second study's 3-step technique included a fingertip rubbing step, the first did not, possibly accounting for the observed difference. Importantly, fingertips are significantly more contaminated than the thenar or hypothenar eminences or dorsum of hands after a standard clinical examination [92]. Indeed, in accordance with the relevance of fingertips contamination in clinical practice, the sampling of fingertips method was endorsed by the EN 1500 [76].

We investigated the effect of modifying WHO's "How to handrub" 6-step technique sequence, and showed that performing "fingertips rubbing" as the first step instead of last led to greater bacterial count reductions on HCWs' hands.

Several factors might explain these results. There might not be enough volume of ABHR left in HCWs hands' at the end of the standard WHO technique to adequately clean

fingertips. We observed a non-significant trend towards a greater difference between techniques among HCWs with large hands, which would support this hypothesis. Hand size was previously shown to influence the microbiological efficacy of hand hygiene action [81]. Importantly however, we observed greater reductions of bacterial counts with the "fingertips-first" technique across all hand size categories.

Our findings are particularly relevant considering the fact that the WHO technique is seldom preformed adequately [55,77]. Recommending HCWs to start the hand hygiene gesture by rubbing their fingertips focus their attention on the most contaminated hand area.

Moreover, the average volume of ABHR used in routine care is only approximately 1 mL. In this context, rubbing the fingertips first may ensure that the most contaminated area is exposed to enough ABHR.

Our study has several limitations. We only tested one strain and one type of ABHR, and the use of the fingertips sampling method might have favoured the "fingertips first" technique. However, evidence suggests that fingertips are strongly implicated in cross-transmission [92]. It would be worth trying to replicate our findings using other sampling techniques, including the American Society for Testing and Materials' "glove juice" method, which samples the entire hand. Finally, the clinical significance of the additional bacterial reduction achieved with the "fingertips-first" technique (i.e. mean 0.77 $\log_{10}$) remains unknown.

Conclusions

In conclusion, rubbing the fingertips first when performing hand hygiene is a simple measure that may lead to a greater reduction in bacterial hand contamination. We emphasize the importance of respecting the recommended steps of the WHO "How to handrub" technique, with particular attention to fingertips rubbing, which likely constitutes the most important step for reducing cross-transmission. Our findings need further validation but could potentially improve hand hygiene action, a gesture of utmost importance for patient safety.

Chapter 3

Hand Hygiene With Alcohol-Based Hand Rub: How Long Is Long Enough?

Pires D, Soule H, Bellissimo-Rodrigues F, Gayet-Ageron A, Pittet D.
Infect Control Hosp Epidemiol 2017;38:547–52. https://doi.org/10.1017/ice.2017.25 [84]

Introduction

According to recent studies, both the hand hygiene technique [73,93] and volume of ABHR [81] are major determinants of the antimicrobial efficacy of hand hygiene actions. However, the optimal duration of hand rubbing remains to be determined. Indeed, there is no strong evidence backing the WHO 20 to 30 seconds recommendation [35] of hand friction. This question is clinically relevant because lack of time is repeatedly identified as a major factor negatively influencing adherence to hand hygiene [32]. Therefore, shorter durations of hand rubbing could potentially lead to improved compliance.

Specific objective #2b

To evaluate the effect of hand rubbing duration on the antimicrobial efficacy of hand hygiene. We aimed to determine whether a shorter duration of hand friction could have an efficacy similar to the currently recommended 30 seconds standard.

Methods

Study setting, participants and eligibility criteria

Healthcare workers with extensive training and expertise in hand hygiene were enrolled in an experimental study at the laboratory of microbiology of the IPC Programme of University of Geneva Hospitals and Faculty of Medicine.

Participants were required to have short nails (< 1 mm). Exclusion criteria included the presence of artificial nails or skin disorders. All subjects gave informed consent to participate.

Study design

We performed a laboratory-based experimental study using the European Norm 1500 [76]. The study consisted in two experiments:

- In a first set of trials (experiment 1), we explored the effect of different durations of hand rubbing (10, 15, 20, 30, 45 and 60 seconds) in the reduction of bacterial counts on the hands of HCWs;
- In a second set of trials (experiment 2), we aimed to replicate the finding of non-inferiority of 15 seconds *versus* 30 seconds in terms of bacterial reduction on hands. We have chosen 15 seconds because it was the lowest hand rubbing duration (see bellow) that achieved the pre-specified non-inferiority margin in the first set of experiments.

Briefly, each trial consisted in a sequence of: contamination of HCWs' hands with *E. coli* (artificial contamination), hand rubbing for a pre-determined period of time and immediate microbiological sampling of hands. Baseline assessment was performed once in each set of trials (experimental session), by sampling the hands of HCWs immediately after contamination with *E. coli*. The microbiological sampling was performed according to the "fingertips method" [76].

Artificial contamination

The reference strain *E. coli* ATCC 10536 was used to prepare a homogeneous bacterial suspension of approximately 10^8 colonies forming units (cfu)/mL. Prior to each contamination procedure, participants were asked to thoroughly wash their hands with 5 mL of a non-antimicrobial liquid soap. Participants inserted their hands until metacarpals into the bacterial suspension for 5 seconds and then let their hands spontaneously dry during 3 minutes.

Hand rubbing for different lengths of time

Regardless of the specific duration of hand rubbing in test, it was performed according to the WHO "How to handrub" technique using 3 mL of isopropanol 60% (v/v; i.e. EN 1500 reference standard) [76]. We randomized the sequence of different hand rubbing durations in both experiments.

All HCWs were asked to follow the WHO "How to handrub" technique.

Microbiological sampling

At baseline and immediately after each hand rubbing sequence, recovery of bacteria from HCWs hands was performed using the "fingertips method". This consisted in rubbing the (five) fingertips of the dominant hand in a sterile dish containing tryptone soy broth

(TSB) for 1 minute. In trials of experiment 1, we used 10 mL of TSB as recommended by the EN 1500.

The EN 1500 [76] states that isopropanol 60% (v/v) is neutralised by dilution only and does not recommend the use of an inhibitor in the TSB sampling medium. However, we wanted to make sure that the additional isopropanol 60% (v/v) that could potentially remain on HCWs hands (in particular following shorter hand rubbing, i.e. 15 seconds) would not inhibit *E. coli* growth. Thus, in trials of experiment 2 we performed the "fingertips method" sampling in a larger sterile dish containing 100 mL of TSB in order to obtain a 10-times more diluted isopropanol on the TSB medium than that recommended by the EN 1500.

Plating of samples

In order to accurately estimate bacterial counts within each sample, samples were studied in 4 different dilutions (10^{-1} to 10^{-4}). A 1 mL sample of each of the dilutions was spread over the surface of a tryptone soy agar plate and subsequently incubated at 36°C ±1°C for 48 h. *E. coli* bacterial cfu were quantified by visual inspection, adjusted for the corresponding dilution factor, and converted to $\log_{10}$.

Study outcomes and statistical analysis

The primary outcome in both experiments was the difference in $\log_{10}$ cfu of *E. coli* recovered from HCWs' hands between baseline and after each hand rubbing durations (corresponding to a reduction of bacterial counts). The duration of hand rubbing (10, 15, 20, 30, 45 and 60 seconds) was the main predictor variable. We used a generalized linear mixed model with a random effect on the subject to analyse the results. We also assessed whether the effect of hand rubbing duration differed according to the hand size category by testing the interaction between these 2 variables. Hand surface areas were calculated and categorized as "small" (surface area ≤375 cm^2), "medium" (376–424 cm^2), or "large" (≥425 cm^2) [81,88]. The final model was adjusted for hand size and sex. The reduction of bacterial counts on HCWs hands between baseline and each of hand rubbing durations was also compared with the reduction achieved after 30 seconds of hand rubbing (considered as the reference for EN 1500).

Following the results of experiment 1 (see below), experiment 2 was designed to confirm whether the reduction of $\log_{10}$ cfu achieved after 15 seconds of hand rubbing was non-inferior to the reduction after 30 seconds. We pre-specified a 0.6 $\log_{10}$ difference between the two durations as the non-inferiority margin, based on EN 1500 [76].

Statistical analyses were performed using Stata® version 14 (StataCorp). Statistical significance was defined as P <0.05 (2-sided).

Results

Thirty-two HCWs participated in the experiments. Among participants, 22 were female (68.6%); they were medical doctors (n=10; 31.3%), nurses (n=17; 53.1%) and other health-care professionals (n=5; 15.6%). The mean hand surface area was 396.9 cm^2 [standard deviation (SD) 53.7, median 397]. Nine subjects had small hands (28.1%), 15 medium hands (46.7%) and 7 large hands (21.9%).

Twenty-three HCWs participated in 87 trials of experiment 1. The total *E. coli* counts on HCWs hands at baseline and after each hand rubbing duration are depicted in Table 4. After adjusting for sex and hand size, duration of hand rubbing was associated with bacterial counts on hands (p<0.001): compared to baseline values, the mean bacterial count reduction was -2.27 log_{10} [95% CI -2.99; -1.54] after 10 sec; -2.52 log_{10} (95% CI -2.93; -2.10) after 15 sec; -2.69 log_{10} (95% CI -3.16; -2.23) after 20 sec; -2.70 log_{10} (95% CI -3.11; -2.28) after 30 sec; -2.17 log_{10} (95% CI -2.71; -1.62) after 45 sec and -2.26 log_{10} (95% CI -2.80; -1.71) after 60 sec (Figure 6). The reduction of bacterial counts after hand rubbing for 30 seconds was not different from the reduction obtained after hand rubbing for 10, 15 or 20 seconds (Table 5). Importantly, only 15 and 20 seconds of hand friction achieve non-inferiority, according to the 0.6 log_{10} margin, when compared to 30 seconds (Table 4). The reduction in bacterial counts after 30 seconds of hand rubbing was larger than what was achieved after 45 or 60 seconds (Table 5). There was no interaction between hand size and hand rubbing duration (P=0.989).

Eighteen subjects were enrolled in 36 trials in experiment 2. All participants performed 15 and 30 seconds of hand friction. The mean total *E. coli* count on HCWs hands at baseline was 6.1 log_{10} (SD ±0.62, median 6.15); after 15 seconds of hand rubbing it was 3.28 log_{10} (SD ± 1.04, median 3.35) and after 30 seconds of hand rubbing it was 3.17 log_{10} (SD ± 1.07, median 3.2). After adjustment for sex and hand size, duration of hand rubbing was associated with bacterial counts on hands (P<0.001): compared to baseline values, the mean bacterial count reduction was - 2.85 log_{10} (95% CI - 3.25; - 2.45) after 15 seconds and - 2.96 log_{10} (95% CI -3.36; -2.56) after 30 seconds. The reduction in bacterial count was not significantly different between 30 and 15 seconds, after adjustment for sex and hand size (P= 0.532). The mean bacterial count reduction after 15 seconds of hand rubbing was 0.11 log_{10} (95% CI - 0.46; 0.24) lower than after 30 seconds of hand rubbing. Using the pre-

specified - 0.6 log_{10} non-inferiority margin, 15 seconds of hand rubbing was non-inferior to 30 seconds regarding bacterial count reductions.

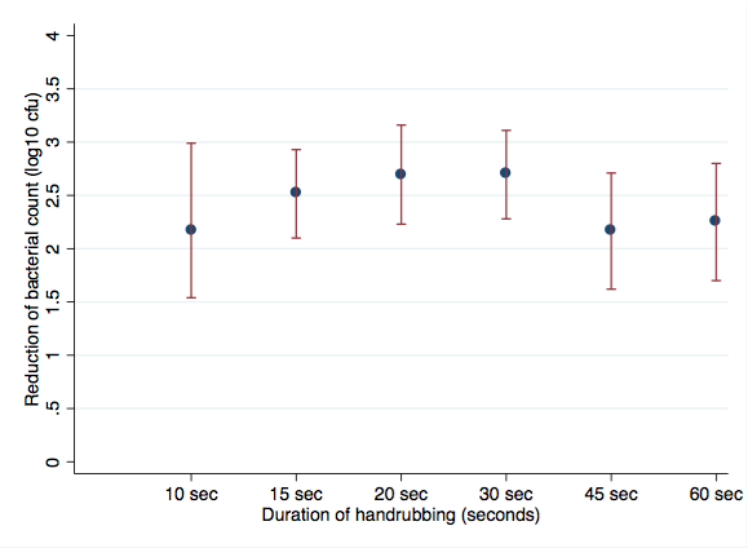

FIGURE 6. Bacterial count reduction (log_{10}) from baseline across 6 durations of hand friction* (mean and 95% CI).

*Alcohol-based handrub used was isopropanol 60% (v/v), 3 mL, according to EN 1500.

cfu: colony-forming units; CI: confidence interval.

TABLE 4. Bacterial counts (log_{10}) on HCWs' hands at baseline and following different durations of friction with alcohol-based handrub* (mean±SD, median).

	Baseline (n=23)	**10 sec (n=5)**	**15 sec (n=23)**	**20 sec (n=16)**	**30 sec (n=23)**	**45 sec (n=10)**	**60 sec (n=10)**
Globally	6.1 (±0.82, 6.4)	3.7 (±0.4, 3.8)	3.6 (±0.9, 3.5)	3.5 (±1.3, 3.3)	3.4 (±1.1, 3.1)	3.7 (±0.7, 3.8)	3.6 (±0.7, 3.9)
By hand size							
Small	5.7 (±1.1, 6.3)	3.9 (±0.2, 4.0)	3.1 (±0.7, 3.0)	2.8 (±0.7, 3.0)	2.9 (±0.8, 2.6)	3.1 (±0.6, 3.0)	3.1 (±0.6, 3.2)
Medium	6.2 (±0.7, 6.5)	3.4 (±0.6, 3.4)	3.7 (±0.9, 3.7)	3.6 (±1.7, 3.3)	3.4 (±1.1, 3.5)	3.8 (±0.6, 3.9)	3.7 (±0.8, 4.0)
Large	6.6 (±0.3, 6.6)	-	4.1 (±1.0, 4.2)	4.2 (±0.7, 4.3)	4.1 (±1.3, 4.4)	4.6 (±0.5, 4.6)	4.4 (±0.4, 4.4)

* Alcohol-based handrub used was isopropanol 60% (v/v), 3 mL, according to EN 1500.

HCWs: healthcare workers; SD: standard deviation.

TABLE 5. Bacterial count reduction (log_{10}) from baseline across 6 durations of hand friction* with 30 seconds as the reference (after adjustment for sex and hand size; multivariate analysis[#]).

	β coefficient	**95% confidence interval**	**p-value**
Duration of friction (ref. 30 seconds)			<0.001
10 sec	-0.45	-1.09; +0.19	0.174
15 sec	-0.18	-0.53; +0.17	0.312
20 sec	+0.07	-0.33; +0.47	0.720
45 sec	-0.71	-1.19; -0.23	0.004
60 sec	-0.62	-1.10; -014.	0.011

* Alcohol-based handrub used was isopropanol 60% (v/v), 3 mL, according to EN 1500.

[#] From a mixed linear model with a random effect on the intercept (SD ±0.98 around the intercept). SD: standard deviation.

Discussion

We investigated the influence of hand rubbing duration in the reduction of bacterial counts on HCWs hands. In the first experiment, we observed that the reduction of bacterial counts after hand rubbing for 10, 15 or 20 seconds was not significantly different from that achieved after 30 seconds and that 15 and 20 seconds achieved non-inferiority. In the second experiment, we confirmed that performing hand friction for 15 seconds was non-inferior to 30 seconds.

Our results expand and strengthen the findings of previous studies. Dharan *et al.* [91] showed that a 15-seconds application of different ABHRs on fingertips was not significantly different from a 30-seconds application in terms of bacterial reduction. Sickbert-Bennett *et al.* [80] studied the microbiological efficacy of 10-seconds hand friction with different hand hygiene agents according to the ASTM-E-1174-94 test method. However, the absence of comparison with other durations of hand friction made it difficult to draw conclusions. Our study compared different durations of hand rubbing and was performed in conditions closely mimicking clinical practice, controlling for the hand hygiene technique and the volume of ABHR used. We believe that this provides more meaningful data for the understanding of the effect of hand rubbing duration on the antimicrobial efficacy of hand hygiene action.

The recommendations of the WHO hand hygiene guidelines regarding the volume of ABHR to use in each hand hygiene action and the duration of hand friction are somewhat incomplete. They recommend: “Apply a palmful of ABHR and cover all surfaces of the hands. Rub hands until dry”. Additionally, the WHO “How to handrub” poster indicates that the duration of the hand hygiene procedure should be 20 to 30 seconds [35][36]. The Centers for Disease Control and Prevention guidelines for hand hygiene [37] are equally imprecise, mentioning that “if hands are dry before 10 to 15 seconds, an insufficient amount of ABHR has been used”. Furthermore, the European norm [76] to test hand products also includes 30 seconds of hand friction, but ABHRs can be tested with hand rubbing durations of up to 60 seconds to pass the norm [81,95,96]. These heterogeneous and imprecise recommendations reflect the overall poor level of evidence and lack of consensus.

As part of a randomized controlled trial, Reilly *et al.* [93] asked HCWs in clinical wards to perform the WHO 6-step hand hygiene technique using 3 mL of ABHR and found that the median hand rubbing duration was 43 seconds (95% CI 39; 45). The real duration of hand rubbing practiced by HCWs in routine care remains unknown, but it is almost certainly shorter (mean 11.6 seconds [SD ±0.7], according to Sickbert-Bennett *et al*.) [80].

HCWs and infection control practitioners would certainly welcome recommendations of shorter durations of hand rubbing, because lack of time remains one of the most important barriers to good practices [32]. In fact, a balance is needed between the optimal performance of the hand hygiene action and its feasibility in daily routine.

Our results demonstrate that hand rubbing for 15 seconds provides similar microbiological efficacy to that achieved after 30 seconds. These findings may allow the future endorsement of a shorter, more feasible and evidence-based duration of hand rubbing, moving forward from the concept of "rubbing hands until dry".

We are concerned with the rather vague concept of "rubbing hands until dry", recommended by both WHO and CDC guidelines, might lead to an utilization of smaller volumes of ABHR, which might interfere with the efficacy of hand hygiene. This is because "time to dry" depends on the volume, type and specific alcohol concentration on the ABHR [97]. Our group previously observed that the perception of dry hands at 30 seconds was only achieved in 1 out of 15 volunteers when using 2 mL of ABHR, but that this number increased to 13 out of 15 when using 1 mL [95]. Importantly, other authors have shown that small volumes that dry at 30 seconds fail to pass the EN 1500 norm [96,97]. One millilitre appears to be the average volume of ABHR used in routine care, [78] and this is clearly insufficient for an optimal hand hygiene procedure [81,96,97].

Hand rubbing for 45 or 60 seconds was associated with somewhat lower antimicrobial efficacy when compared to 30 seconds. We have no definite explanation for this finding. One possibility is that very prolonged hand rubbing might lead to the desquamation of the *stratum* layer of the skin during hand rubbing, leading to a loss of effectiveness in alcohol-induced bacterial killing.

In experiment 2 we confirmed the results obtained in experiment 1 with regards to the non-inferiority of 15 seconds' duration hand rubbing.

We are confident of the absence of any significant residual alcohol activity on hands after hand rubbing and of an effect on bacterial counts. According to Rotter, [86] the minimal bactericidal concentration (MBC) for isopropanol against *E. coli* at 1-minute in contact suspension tests is 26% (v/v). Even in the most extreme scenario where all 3 mL of isopropanol 60% (v/v) applied on HCW's hands for hand rubbing would be transferred to the TSB -which is very unlikely considering the volume of ABHR spread on hands during hand hygiene action, we would obtain maximal final isopropanol concentrations of 17% (v/v) in 10 mL of TSB and 1.7% (v/v) in 100 mL of TSB, respectively; both concentrations are thus below the MBC for *E. coli*.

Our study has several limitations. First, our observations were obtained in laboratory-based experimental conditions involving HCWs with extensive training and expertise in hand hygiene, who performed the WHO 6-step technique of hand rubbing using 3 mL of ABHR. This might limit the external validity of our findings. Second, we only tested the ABHR and bacterial strain used in the EN 1500. More studies with different bacteria and types of ABHRs would be required to further validate our results. Third, the methodology used in experiment 2 to standardize the WHO 6-step technique does not reflect daily clinical practice. Importantly, the jury is still out regarding the clinical significance of the observed

differences in bacterial reduction achieved with the different durations of hand rubbing. Forth, while bacterial reduction was not statistically different between 15, 20 and 30 seconds, we cannot rule out a type II error. Finally, it was not our intention to change the recommendations regarding the necessary duration of hand friction of individual ABHRs, as those need to be tested according to the appropriate norms. However, our results suggest that norm testing standards should be revisited and include evaluation times as short as 15 seconds.

Conclusions

Our results suggest that hand friction for 15 seconds is non-inferior to 30 seconds in reducing bacterial counts on HCWs hands, when performing the WHO "How to handrub" technique and using 3 ml of ABHR. In addition, no gain seems to result from performing hand rubbing for more than 30 seconds.

These results have important implications for hand hygiene practices and future research. Reducing the time needed to perform an optimal hand hygiene gesture could help improving hand hygiene compliance, because lack of time is a major factor driving non-compliance [32]. However, further studies are needed to assess the clinical significance of our findings.

Chapter 4

Antibacterial efficacy of hand rubbing for 15 versus 30 seconds: EN 1500-based randomized experimental study with different loads of *Staphylococcus aureus* and *Escherichia coli*

Pires D, Soule H, Bellissimo-Rodrigues F, de Kraker MEA, Pittet D.
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Introduction

A number of recent studies have suggested that the WHO-recommended hand hygiene action could be simplified while preserving antibacterial efficacy. This may include modifications in the technique ("6 steps fingertips-first" [83] or "3 steps" [94,98] instead of "6 steps"), shortening the duration of hand rubbing (15 seconds instead of "20 to 30 seconds or rubbing until dry" [84,99]) and customizing the volume of alcohol-based handrub (hand size customization instead of "palmful" [81]).

However, the jury is still out, as a recent systematic review on the efficacy of the HH technique highlights [82]. Moreover, most studies focused on just one aspect of the hand hygiene action, either technique, volume or duration, and the majority were performed at the laboratory using only a fixed concentration of *E. coli* to contaminate hands. Some studies were performed at the bedside, but those did not control for the type and concentration of bacteria on HCWs hands [93,98]. Therefore, it remains unknown how the proposed modifications interplay, particularly in terms of antimicrobial efficacy, and if results hold with different types and loads of transient bacteria.

Specific objective #2c

We tested the antibacterial efficacy of a hand size customized volume hand-hygiene action with different durations (15 seconds versus 30 seconds), bacteria (*S.s aureus* and *E. coli*) and bacterial contamination loads (10^8 or 10^6 cfu/mL).

Methods

We enrolled 18 HCWs with extensive hand hygiene training of the IPC Programme of the University of Geneva Hospitals. The study was part of the IPC quality assurance program approved by the local ethics committee and all HCWs agreed to participate. Participants were required to have short nails, no skin abrasion and to not use an ABHR

containing chlorhexidine for at least 24 hours. This cross-over, randomized experimental study was based on the European Norm 1500 [76].

All participants repeated the following sequence: a) artificial contamination of hands by immersing them down the metacarpals in a bacterial suspension; b) drying hands for 3 minutes; c) baseline microbiological sampling; d) hand rubbing with a hand size customized volume of isopropanol 60% (v/v); e) repetition of microbiological sampling.

The 10^8 cfu/mL bacterial suspension was prepared according to the EN 1500 [76] and the 10^6 cfu/mL was obtained by diluting the former with sterile water. The lower 10^6 cfu/mL concentration was chosen because it simulated the low-level contaminations that are thought to be encountered in clinical practice, while still allowing the recovery of bacteria from the hands of HCWs after hand rubbing in order to calculate the reduction factor. A slight modification of the EN 1500 "fingertips method" of sampling was used. It consisted of having all fingertips and thumbs of both hands rubbed in a sterile dish containing 100 mL of tryptone soy broth for 1 minute. Even if the EN 1500 does not recognize the use of an inhibitor of isopropanol 60% v/v in the TSB medium, we performed a 10-times higher dilution to ensure that there was no bacterial inhibition by the alcohol (in particular following the 15 seconds hand rubbing, where the HCWs hands could still be wet). Processing of samples and quantification of cfu were carried out by a blinded investigator and according to the EN 1500 [76].

All participants performed 12 experimental sequences with all possible combinations: type of bacteria (*E. coli* ATCC 10536 or *S. aureus* NC 10788), concentration of bacterial suspension (10^8 cfu/mL or 10^6 cfu/mL) and duration of hand rubbing (15, 30 seconds or no hand rubbing). The "no hand rubbing" experimental sequence intended to account for the bacterial reduction due to the sampling method and not to the hand rubbing itself ("correction factor").

The type of bacteria was not randomly allocated for practical reasons. For each type of bacteria, the experimental sequences started with the low concentration, to prevent carry-over of bacterial load. The order of 15 versus 30 seconds of hand rubbing was randomly allocated using a computer-generated sequence with a block randomization of size 2.

Participants were allowed to perform the WHO 6 steps technique according with their usual practice / preference, without specific advice or systematic assessment. The current expert consensus at the HUG favours the 6 steps technique starting with fingertips' rubbing [83].

Hand size customized volume of ABHR

The hand surface area (HSA) of participants was measured according to Hsu Y-W *et al.* [88]. In accordance with previous work [81,83,84], HSA was categorized as small (surface area ≤375 cm^2), medium (376–424 cm^2), or large (≥425 cm^2).

To calculate the volume of ABHR for each HCW, we used data from the paper by Bellissimo-Rodrigues *et al.* [81]. Our goal was to calculate the volume that achieved a 2 log_{10} reduction of bacterial counts on the hands of HCWs.

The 2 log_{10} reduction goal was based on the American Society for Testing and Materials requirements [87], since there is no proven threshold for transmissibility. The 2 log_{10} reduction was also in accordance to the threshold of non-inferiority of 1 log_{10} described in Chapter 1 [74], because several studies point to a mean of 3 log_{10} contamination in usual clinical activities [53].

In the paper of Bellissimo-Rodrigues *et al.* [81], a generalized linear mixed model assessed the log_{10} bacterial reduction depending on hand size category and volume of ABHR. We calculated the volume of ABHR required to achieve a 2 log_{10} bacterial reduction for each hand size category defined in that paper [81]. Then, we divided the volume obtained for each category by the mean hand surface area in each category. This resulted in a volume of ABHR per hand surface area (cm^2) per category: 0.0066 mL/cm^2 for the small hand category, 0.0057 mL/cm^2 for the medium hand category and 0.0070 mL/cm^2 for the large hand category. In order to calculate the volume of ABHR required for each HCW in our study, we multiplied those category-specific coefficients by their individual hand surface area.

Sample size calculation, study outcomes and statistical analysis

The EN 1500 specifies a sample size between 18 and 22 subjects to obtain the necessary precision to be able to perform a non-inferiority test. We followed this recommendation and recruited 18 volunteers.

We calculated the median and inter-quartile range (IQR) of bacterial counts (log_{10} cfu) at baseline and the corrected reduction factor (cRF) after hand rubbing for 30 or 15 seconds, for each bacteria and contamination fluid concentration tested. The reduction factor was obtained by calculating the difference in bacterial counts between baseline and the final sampling. The reduction factor obtained after no hand rubbing was designated «correction factor». The corrected reduction factor was then obtained by subtracting the correction factor from the reduction factor.

We performed a generalized linear mixed model with a random intercept for subject, corrected reduction factor as dependent variable and the following independent variables: type of bacteria (*E. coli* vs *S. aureus*), contamination fluid concentration (10^8 cfu/mL vs 10^6 cfu/mL) and hand rubbing duration (15 vs 30 sec). In order to assess whether the effects of hand rubbing duration and contamination fluid concentration differed according to the type of bacteria we included an interaction between each of those variables and type of bacteria. We performed a sub-group analysis per bacteria and per contamination fluid concentration, to ascertain whether the cRF achieved after 15 seconds of hand rubbing was non-inferior to that achieved after 30 seconds in all sub-groups.

Since there was inter-individual variation in the baseline bacterial counts after immersing hands in the contamination fluids with the same concentration, we performed a sensitivity analysis. We performed a categorization of the baseline bacterial counts into two categories: high (baseline bacterial counts >=6 $\log_{10}$ and <=8 $\log_{10}$) and low (>=3 $\log_{10}$ and <=5 $\log_{10}$). These categories were chosen based on the distribution of the baseline bacterial counts in order to allow for a maximal contrast. We excluded all values that did not fit in these intervals. We performed a generalized linear mixed model similar as described above but with categories of baseline bacterial counts instead of contamination fluid concentration as one of the dependent variables.

Statistical analyses were performed using Stata® version 13 (StataCorp). Statistical significance was defined as $P<0.05$ (2-sided). We pre-specified the NI margin at 0.6 $\log_{10}$ difference between the two durations based on EN 1500 [76] and 95% confidence intervals.

Results

Eighteen HCWs participated in all 12 experimental sequences (n=216). There were 12 women (67.0%), seven nurses (38.9%), two doctors (11.1%), two hospital pharmacists (11.1%), and two engineers (11.1%); five HCWs had other healthcare-related professions (27.8%).

Overall, 6 (33.3%) HCWs had small hands (median hand surface area 335.5 cm^2 IQR 331, 360), 7 (38.8%) had medium hands (395 cm^2 IQR 384, 397) and 5 (27.7%) had large hands (450 cm^2 IQR 431, 492). The median volume of ABHR used for each hand size group was 2.2 mL (IQR 2.2, 2.4) for small, 2.3 mL (IQR 2.2, 2.3) for medium and 3.2 mL (IQR 3.0, 3.4) for large hands.

The overall, median corrected reduction factor (cRF) was 2.10 $\log_{10}$ (IQR 1.50, 3.10). The corrected reduction factors per hand rubbing duration and contamination fluid

concentration are shown in Figure 7. The total bacterial counts at baseline, correction factors and corrected reduction factors are shown in Table 6.

After fitting the mixed effects model, the cRF was significantly higher for *S. aureus* compared to *E. coli* but there was no significant effect for duration of hand rubbing or contamination fluid concentration. There was a significant interaction between type of bacteria and contamination fluid concentration. There was no interaction between duration of hand rubbing and type of bacteria (Table 7). The 15 seconds of hand rubbing was non-inferior to 30 seconds in all pre-specified subgroups, regardless of bacterial type or contamination fluid concentration (Table 8).

In the sensitivity analysis for baseline bacterial counts, we excluded 31/144 values that did not fit in our pre-defined categories. The median $\log_{10}$ bacterial counts was 7.00 (IQR 6.60, 7.40) in the high baseline contamination category and 4.20 (IQR 3.60, 4.60) in the low baseline contamination category. Baseline bacterial counts on hands did influence cRF when hands were contaminated with *E. coli*, as a high baseline hand contamination resulted in larger bacterial reductions when compared to low baseline contamination. However, our main conclusion remains unchanged: 15 seconds of hand rubbing are NI to 30 seconds (Table 7).

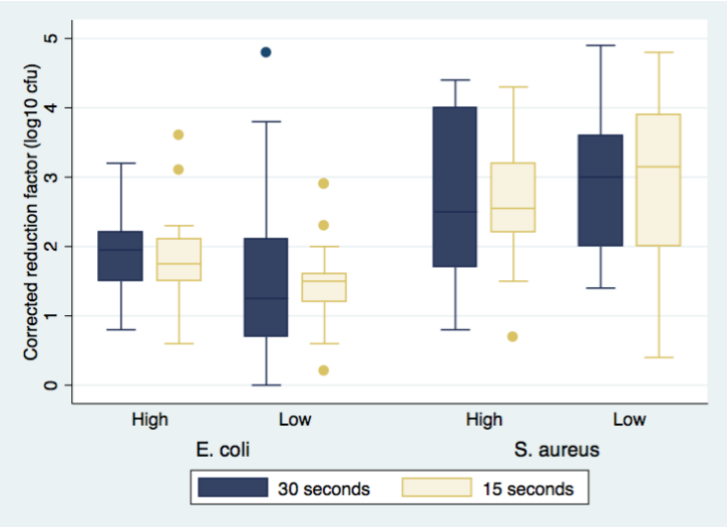

FIGURE 7. Box plot of the corrected reduction factor ($\log_{10}$ cfu) of bacterial counts on health-care workers' hands after hand rubbing with a hand size personalized volume of isopropanol 60% (v/v), according to duration of hand rubbing (15 and 30 seconds), type of bacteria (*Escherichia coli* and *Staphylococcus aureus*) and contamination fluid concentration (Low: 10^6 CFU/mL; High: 10^8 CFU/mL)

TABLE 6. Bacterial counts on healthcare workers' hands at baseline, correction factor[#], and the corrected reduction factor* after hand rubbing for 15 or 30 seconds, with a customized volume of alcohol-based handrub[+], according to type of bacteria and contamination fluid concentration.

	Bacterial counts at baseline [median (IQR); n=18]	Reduction of bacterial counts due to sampling[#] [**correction factor**; median (IQR); n=18]	Corrected reduction of bacterial counts after 15 sec of hand friction (**corrected reduction factor;** median (IQR); n=18]	Corrected reduction of bacterial counts after 30 sec of hand friction (**corrected reduction factor;** median (IQR); n=18]
Overall[$]	5.50 (4.20, 6.85)	0.65 (0.40, 0.95)	2.05 (1.50, 2.90)	2.10 (1.50, 3.20)
E. coli	5.50 (3.90, 7.00)	0.70 (0.50, 1.10)	1.50 (1.35, 1.95)	1.65 (1.15, 2.20)
10^6 cfu/mL contamination	3.90 (3.30, 5.00)	0.70 (0.30, 0.90)	1.50 (1.20, 1.60)	1.25 (0.70, 2.10)
10^8 cfu/mL contamination	7.00 (6.20, 7.40)	0.70 (0.60, 1.10)	1.75 (1.50, 2.10)	1.95 (1.50, 2.20)
S. aureus	5.60 (4.30, 6.80)	0.60 (0.40, 0.90)	2.70 (2.15, 3.65)	2.65 (1.95, 3.95)
10^6 cfu/mL contamination	4.30 (3.80, 4.80)	0.55 (0.30, 0.90)	3.15 (2.00, 3.90)	3.00 (2.00, 3.60)
10^8 cfu/mL contamination	6.80 (6.50, 7.20)	0.60 (0.50, 0.90)	2.55 (2.20, 3.20)	2.50 (1.70, 4.00)
E. coli and *S. aureus*[&] 10^6 cfu/mL contamination	4.20 (3.50, 4.90)	0.65 (0.30, 0.90)	1.80 (1.40, 3.15)	2.10 (1.25, 3.35)
E. coli and *S. aureus*[%] 10^8 cfu/mL contamination	6.85 (6.30, 7.40)	0.65 (0.50, 1.00)	2.15 (1.60, 2.80)	2.10 (1.55, 3.00)

[*] The corrected reduction factor was obtained by subtracting the correction factor to the reduction factor obtained with 15 and 30 seconds of hand rubbing by the same participant with the same type and concentration of bacteria. The reduction factor is the difference in $\log_{10}$ bacteria counts between baseline and the final sampling.

[#] The reduction factor obtained after "no hand rubbing" was called the correction factor. The "no hand rubbing" experimental sequence intended to account for the bacterial reduction due to the sampling method and not to the hand rubbing itself.

[+] Alcohol-based handrub used was isopropanol 60% (v/v).

$ Pooled results from all the experiences preformed (*S. aureus* with 10^6 cfu/mL and 10^8 cfu/mL contamination fluid concentration and *E. coli* with 10^6 cfu/mL and 10^8 cfu/mL contamination fluid concentration).

& Pooled results from the experiences with *S. aureus* and *E. coli* with 10^6 cfu/mL contamination fluid concentration.

% Pooled results from the experiences with *S. aureus* and *E. coli* with 10^8 cfu/mL contamination fluid concentration.

cfu: colony-forming units; IQR: inter-quartile range; sec: seconds.

TABLE 7. Corrected reduction factor (log_{10} cfu) of bacterial counts on HCWs hands after hand rubbing with hand size personalized volume of isopropanol 60% (v/v), depending on type of bacteria, hand rubbing duration and contamination fluid concentration[$] (original model) or type of bacteria, hand rubbing duration and baseline bacterial category* (sensitivity analysis), using a generalized linear mixed model with a random intercept for subject.

	Corrected reduction factor in log_{10} cfu (95% CI; P)	
	Original model (n =18) (*Contamination fluid concentration*[$])	**Sensitivity analysis (n=18)** (*Baseline bacterial category**)
15 seconds *versus* **30 seconds**	- 0.06 (-0.34, 0.22; P=0.659)	0.01 (-0.29, 0.30; P=0.966)
S. aureus *versus* ***E. coli***		
Low	1.41 (1.02, 1.81; P<0.001)	1.48 (1.04, 1.93; P<0.001)
High	0.79 (0.40, 1.19; P<0.001)	0.67 (0.26, 1.09; P=0.001)
Low *versus* **High**		
S. aureus	0.27 (-0.12, 0.66; P=0.179)	0.24 (-0.17, 0.64; P=0.250)
E. coli	- 0.35 (-0.74, 0.04; P=0.081)	-0.57 (-1.02, -0.13; P=0.012)

[$] Low contamination fluid concentration: 10^6 cfu/mL; High contamination fluid concentration: 10^8 cfu/mL
* Low baseline bacterial counts: >=3 log_{10} and <=5 log_{10}; High baseline bacterial counts: >=6 log_{10} and <=8 log_{10}
CI: Confidence Interval; HCW: healthcare workers.

TABLE 8. Corrected reduction factor (log_{10} cfu) of bacterial counts on HCWs hands after hand rubbing with hand size personalized volume of isopropanol 60% (v/v), using a generalized linear mixed model with a random intercept for subject and the following independent variables: type of bacteria, hand rubbing duration and contamination fluid concentration, for the pre-specified sub-groups in order to access the non-inferiority (NI) of 15 seconds vs 30 seconds of hand rubbing according to the EN 1500 0.6 NI margin.

Impact of 15 vs 30 seconds of hand rubbing for each subgroup	**Corrected reduction factor in log_{10} cfu**	**95% CI**	**P value**
E. coli	- 0.06	- 0.39, 0.27	0.717
S. aureus	- 0.06	- 0.46, 0.34	0.754
10^6 cfu/mL fluid concentration	-0.05	-0.49, 0.39	0.833
10^8 cfu/mL fluid concentration	-0.08	-0.42, 0.27	0.659
S. aureus 10^6 cfu/mL fluid concentration	-0.02	- 0.54, 0.50	0.950
S. aureus 10^8 cfu/mL fluid concentration	-0.11	-0.54, 0.32	0.613
E. coli 10^6 cfu/mL fluid concentration	-0.08	-0.53, 0.37	0.734
E. coli 10^8 cfu/mL fluid concentration	-0.04	-0.26, 0.18	0.692

CI: Confidence Interval; HCW: healthcare workers.

Discussion

In this laboratory-based study, we showed that 15 seconds of hand rubbing with a hand size customized volume of ABHR was non-inferior to 30 seconds of hand rubbing, both globally and for all pre-specified subgroups. Furthermore, our results show that the impact of hand rubbing depends on the pathogen: *S. aureus* was more easily removed from hands than *E. coli*. Level of hand contamination, as measured by baseline hands sampling, also influences *E. coli* eradication: high baseline hand contamination resulted in larger reductions.

These results significantly expand previous findings. We have previously demonstrated that 15 seconds of hand rubbing was non-inferior to 30 seconds for a fixed (3 mL) volume of isopropanol 60% (v/v), but this was tested only for *E. coli* at a concentration of 10^8 cfu/mL [84]. Kramer *et al.* [99] have also verified the non-inferiority of hand rubbing for 15 seconds with several commercial ABHR agents compared to 30 seconds with isopropanol 60% (v/v), again only using contamination with *E. coli* [10^8 cfu/mL]. Taken together, these findings provide further support for shortening the recommended duration of hand rubbing down to 15 seconds, in HCWs well trained in the "How to handrub" technique and using an appropriate volume of ABHR.

WHO recommendations regarding the volume of ABHR and duration of hand friction are relatively imprecise: "palmful" and "rubbing until dry or for 20 to 30 seconds", respectively [35]. Furthermore, studies have shown that practices in everyday clinical care are suboptimal (mean 1.69 mL of ABHR [100] and 11.6 seconds of hand rubbing [80]). Our findings are important and might be welcomed by HCWs, because a shortened hand hygiene action is more easily applicable to real-world clinical care. Indeed, it is known that time constraint is mentioned as a major barrier to compliance to hand hygiene [32]. Kramer *et al.* [99] performed a small-sized study in a Neonatal Intensive Care Unit where 14 nurses, well trained in the 6-step WHO technique, were randomly allocated to perform hand rubbing for either 15 or 30 seconds, during a 8 hour-shift. Interestingly, the group allocated to 15 seconds performed significantly more hand hygiene actions (7.9±4.3 standard deviation [SD] vs 5.8±2.9 SD per hour; P=0.05) [99]. Importantly, our study suggests that the duration of hand rubbing can be safely reduced without jeopardizing the antibacterial efficacy of the hand hygiene action. Further studies will be required to confirm whether compliance will indeed increase with a recommendation of hand rubbing for 15 seconds.

There are possible challenges in the implementation of a shorter hand rubbing action using hand size customized volumes of ABHR [101]. Hands were not completely dry after

15 seconds of hand rubbing with the hand sized ABHR volume used in our study. However, it has been shown that volumes of ABHR allowing for drying even after 30 seconds (1.7-2.1 mL [97]) are generally insufficient in terms of antimicrobial efficacy. Thus, there is currently a miss-match between the volume of ABHR and duration of friction required for an efficient handrub and the "time to dry" concept. There is a need for efficient ABHR products that dry faster. Additionally, there are still no commercially available ABHR dispensers capable of personalizing the volume of ABHR delivered to each HCW. However, with advancing technology, this could be overcome in the context of a high demand. Finally, the compliance of HCWs with the use of the personalized volume of ABHR also needs to be studied.

Interestingly, we have found a greater reduction of hand contamination for *S. aureus* than for *E. coli*. We are not aware of studies reporting a greater ABHR killing activity for *S. aureus*. Kramer *et al.* [102,103] did not find differences in ABHRs *in vitro* killing between *E. coli* and *S. aureus*. However, they did not perform direct comparisons. Another experiment showed no difference in $\log_{10}$ reduction between *E. coli* and *Micrococcus luteus*, used as a surrogate for Gram-positive pathogens [104]. More studies are required to confirm our results and, if confirmed, to ascertain whether the observed differential killing has any clinical relevance.

Our study has a couple of additional limitations. Participants were trained in the WHO 6 steps technique, but were left to perform it in accordance with their preference. We believe that this was an advantage, as it mimics more closely real-life clinical practice. However, we acknowledge that it also reduces standardization in the experiment. The expertise of this IPC team in hand hygiene might limit the external validity of the results, but this also highlights the need to train HCWs in the hand hygiene technique. We did not test a wide range of pathogens; more studies with other bacteria, strains and concentrations are needed. Finally, we used previous data from a small laboratory study to calculate the hand size customized volume of ABHR [81]. Other approaches to this customization are possible and we believe that further studies are needed to optimize it.

In addition, other variables need to be tested besides volume, hand size volume customization, hand rubbing duration, technique and bacterial type and load, such as ABHR alcohol concentration, contamination procedures [105], hands' moisture level, type of skin, and friction vigour [106].

Conclusions

In conclusion, our findings suggest that the WHO “How to handrub” technique can be safely shortened down to 15 seconds, without losing antimicrobial efficacy We invite other researchers to replicate our findings and help refining this proposed change in clinical practice. Importantly, the effect on compliance of a shorter “How to handrub” action needs to be tested in a clinical setting.

Chapter 5

Impact of wearing the SmartRub® device on hand hygiene compliance among healthcare workers: a stepped wedge cluster-randomized clinical trial

Pires D, Gayet-Ageron A, Guitart C, Robert YA, Fankhauser C, Tartari E, Peters A, Tymurkaynak F, Fourquier S, Soule H, Beuchat R, Bellissimo-Rodrigues F, Martin Y, Zingg W, Pittet D.

Advanced draft

Introduction

A number of studies suggest that the HH action as endorsed by the WHO could be simplified without compromising efficacy. Proposed changes include shortening the duration of hand rubbing to 15 seconds (instead of 30 seconds) [84,85,107,108] and changing the number or order of steps ("3 steps" or "fingertips-first" instead of the current "6 step" technique) [83,94,109]. Additionally, a recent study has suggested an approach to standardize the hand size customization of the volume of alcohol-based handrub (ABHR) recommended by the WHO guidelines ("palmful") [64,81]. It is hypothesized that the suggested simplification of the technique [109] and the shortening of the duration of hand rubbing [107] could lead to an increase in the quality of the HH action itself. In addition, some small studies have suggested that a simplified HH action could also improve compliance with the "5 moments" [107,109].

Monitoring and feedback are essential parts of the WHO multimodal strategy to improve HH [110]. However, direct observation and timely feedback are time-consuming, costly and prone to bias [60,61]. There has been a growing interest in the use of electronic monitoring, which can be applied during all shifts, increase the number of observed actions and remove the observation bias (Hawthorne effect) that has been observed with the WHO direct observation method [61].

SmartRub® is a novel electronic device, consisting of a wristband which communicates with a healthcare workers' (HCW's) personal ABHR bottle. It monitors and provides real-time, personalized feedback to HCWs, and is based on a simplified and customized HH action (15 seconds of hand rubbing and hand size customized volume of applied ABHR) [111]. This device is the result of an investigator-initiated partnership between three Swiss institutions: the University of Geneva Hospitals and Faculty of

Medicine, the "Haute école du paysage, d'ingénierie et d'architecture de Genève" (HEPIA) and iQati™, a start-up company.

Specific objective #3a and #3b

We aimed to test the impact of SmartRub® on HH compliance with the "5 moments" and on the quality of hand rubbing in daily patient-care activities.

Methods

Study design

This was a Swiss National Research Foundation (FNS)-funded, investigator-initiated-, single-centre-, stepped wedge-, cluster randomized-, and controlled open-label trial, conducted at the 300-bed geriatric hospital of HUG. Wards were considered clusters because each team has an individual organisational culture and behaviour change interventions affect team work as a whole. Randomisation followed a stepped-wedge design because although the sequence of interventions was fixed, secular trends could thus be taken into account.

Participants

All wards, including outpatient clinics and the emergency department of the geriatric hospital at HUG were eligible if a sufficient number (n=5) of HCWs working permanently in these wards volunteered to participate. HCWs were not eligible if they: a) planned to leave the ward during the study period; b) worked in more than one ward (to avoid contamination between wards); c) had more than three planned consecutive weeks of vacations during the study period; and d) used an ABHR agent other than the standard ABHR bottles provided at HUG due to skin allergies.

HCWs were recruited by twenty-six 10-minute information sessions at clinical nursing and medical staff meetings in the wards, as well as by posters and leaflets, distributed in all wards at the geriatric hospital. Video sessions were organized in participating wards to provide easy and coherent information to the HCWs.

The trial was approved by the Regional Research Ethics Committee (Geneva, Switzerland, 08/06/2016, ref: 2016-00714), and all participating HCWs provided written informed consent.

Settings

HUG is a 1900-bed, tertiary-care, university hospital, with approximately 50,000 admissions per year. The geriatric hospital is a free-standing building, located at a separate site in the city of Geneva, with a capacity of 300-beds and approximately 10,000 admissions per year.

Intervention

The intervention consisted on providing individual feedback to HCWs based on a shorter duration of hand rubbing (15 seconds) and a volume of ABHR customized to each HCW's hand size. Feedback was provided in real-time by the SmartRub® electronic device, which monitors these parameters each time the HCW performs a HH action (Figure 2).

The device was developed in collaboration between HUG, HEPIA and iQati™ [111]. SmartRub® consists of two elements, a bottle and a wristband. The bottle and wristband communicate with each other through wifi, and transfer data to a centralised database.

The pocket-size bottle of SmartRub® was integrated into the individual ABHR bottle that is widely used at HUG. Each of these bottles were equipped with a volumetric flow meter to measure the volume of ABHR poured onto hands [111]. Feedback was provided through the bottle vibrating as soon as the pre-defined ABHR volume had been applied. The volume was determined for each HCW by taking into account the surface area of each individual's hands [88], as described in previous laboratory studies [74,81,85]. The wristband contained an electronic device to identify spatial movements of hand rubbing, thus measuring the duration of HH action. Feedback for sufficient rubbing time was provided by the wristband, which vibrated after 15 seconds of hand rubbing [84,85].

HCWs were asked to disinfect the wristband and bottle and place them into a charging station at the end of each shift, and collect them upon starting the subsequent shift (Figure 2). Wristbands were made of silicone and could be easily disinfected using the detergent-disinfectant widely used at HUG (0,2% quaternary ammonium and 50% alcohol).

The device recorded the date, time of use, volume of applied ABHR, duration of hand rubbing, and whether feedback was provided to the HCW or not, for all of the HH actions. During the baseline period, HCWs did not wear the devices. During the transition period, the bottles and wristbands were worn, but the feedback mode (vibration) was not activated, though the device actively collected data on the volume of ABHR applied and the duration of hand rubbing. During the intervention period, both the feedback (vibration) and monitoring were activated.

The overall sensitivity and specificity of SmartRub® to correctly identify a HH action were 94.1% and 99.0% in laboratory conditions, and 96.8% and 98.3% in clinical conditions [111].

Study periods and randomisation

The study duration, including baseline, transition and intervention periods, was six months, followed by a 2-months wash-out- and a one-month follow-up period (Figure 8). At follow-up, HCWs did not use the device.

After an initial one month baseline period, wards were randomly assigned to start with the one-month transition period, followed by the intervention period at a rate of three wards per month. In total, 12 wards were randomized, following a computer-generated block randomization (1:1:1:1) performed by an independent statistician. Numbered opaque envelopes allowed allocation concealment.

Wards were informed the day before shifting from baseline to the transition period. Due to the nature of the study, it was not possible to mask study participants or observers after the baseline period.

Months	1	2	3	4	5	6	7	8	9
Group 1	baseline	transition	intervention	intervention	intervention	intervention			follow-up
Group 2	baseline	baseline	transition	intervention	intervention	intervention			follow-up
Group 3	baseline	baseline	baseline	transition	intervention	intervention			follow-up
Group 4	baseline	baseline	baseline	baseline	transition	intervention			follow-up

FIGURE 8. Design of the SmartRub® study, Geriatric Hospital, Geneva University Hospitals, June-December 2019.

Primary outcome

The primary outcome was overall HH compliance, monitored by direct observation of HH applying the WHO guidelines on direct HH observation [35]. Hand hygiene opportunities were identified, and actions, either by rubbing or washing, were recorded [33]. Hand hygiene compliance was calculated as a proportion of HH actions divided by the identified HH opportunities, with 95% confidence intervals (CI). Three validated observers performed HH observations during audit sessions on weekdays and day shifts. Each HCW was observed at least once per month, with the each observation period consisting of a minimum of 6 HH opportunities. HH compliance at the individual HCW level (closed cohort) between intervention and baseline was then compared.

Secondary outcomes

Secondary outcomes were: 1) volume of ABHR and duration of hand rubbing of each HH action in the transition and intervention periods, as automatically recorded by SmartRub®; 2) adherence to device use (hours of device use per day) and frequency of HH (HH events per hour), as automatically recorded by SmartRub®; 3) HH compliance of participants during follow-up by direct HH observation; 4) ABHR consumption per ward and study period, as per pharmacy dispensing; 5) adverse events spontaneously reported to the study team by HCWs; and 6) satisfaction and perception of device usefulness assessed by means of a post- study questionnaire and in focus group discussions.

Sample size estimation and statistical analysis

Sample size was estimated by hypothesizing a relative mean HH compliance increase by 20%, increasing from 69% (the 2015 HH compliance at HUG) to 83%, between the baseline and intervention period. This corresponds to a standardized difference in proportions of 0.35 [112]. Due to the clustered study design, we corrected for the correlation of compliance within wards by calculating a design effect [59]. We have used an intra-class correlation (ICC) coefficient of 0.015 (based on the 2013-2014 HH compliance data at HUG), leading to a design effect of 1.06. Considering a study power of 80%, and an alpha error fixed at 5% (two-sided), we estimated that 12 wards (3 per group, 4 groups) with at least 5 HCWs per ward would be needed to test our study hypothesis [113,114].

The study was registered with ISRCTN25430066 on the 22/05/2017 (http://www.isrctn.com/ISRCTN25430066).

Continuous variables were reported by means (±standard deviation, SD), medians (interquartile ranges, IQR), or means (95% confidence interval, 95% CI), where appropriate. Categorical variables were reported by numbers with relative frequency.

All analyses were conducted on an intention-to-treat (ITT) and a per-protocol (PP) basis, the latter excluding HCWs that did not have at least one HH observation in each study period or did not complete the intervention period. Pooled HH compliance between the intervention and the baseline periods was modelled using mixed-effects logistic regression. The effect of length of active device use on HH compliance was assessed by testing for interactions between the randomisation group and the study period (baseline versus intervention). The clustering of data was taken into account by including nested random

effects for HH opportunities within each audit session, HCWs within each ward, and the group of wards within each group (groups>wards>HCWs>sessions>opportunities).

All analyses were adjusted for covariates comprising HCWs' characteristics (age, sex, profession, years of work experience, years since last HH training, full/part-time work), ward specialty (internal medicine, geriatrics, emergency department, ambulatory care), workload (mean HH opportunities per minute) and the type of ABHR used (gel, rinse). The models also included time of exposure to the study (number of days between the start of the study and the individual HH session).

We compared the volume of applied ABHR and the duration of hand rubbing between the transition and intervention periods by performing a mixed linear regression model with each outcome as the dependent variable. We then assessed the effect of length of active device use on volume and duration of hand rubbing by testing for interaction between the randomisation group and the study period (transition versus intervention). Analysis used HCW-level data clustered within the ward level and group level (nested random effects: groups>wards>HCWs). All analyses were adjusted for the same covariates as the primary analysis. In addition, we calculated the proportion of correct HH actions, which were defined as a sufficient volume ABHR applied (as per hand size), and hand rubbing for at least 15 seconds. Finally, we calculated the HH compliance of all HCWs and per group for follow-up two months after the intervention period.

Two-sided P-values of less than 0.05 were considered statically significant. All analyses were performed using Stata IC version 16.0 (StataCorp, College Station, TX, USA).

Role of the funding source

This study was funded by the Swiss National Foundation (2003B_163262). The funder had no role in study design, data collection, data analysis, data interpretation, or writing of the manuscript. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit for publication.

Results

All 12 wards of the geriatric hospital at HUG were eligible and were recruited from 1 June 1 to 30 June, 2017. Of the 306 eligible HCWs in the 12 wards, 97 volunteered and were included in the study (Figure 9). Baseline characteristics of wards and HCWs per

randomisation group are presented in Table 9. Table 10 and Figure 10 summarise the starting and end dates of the study periods.

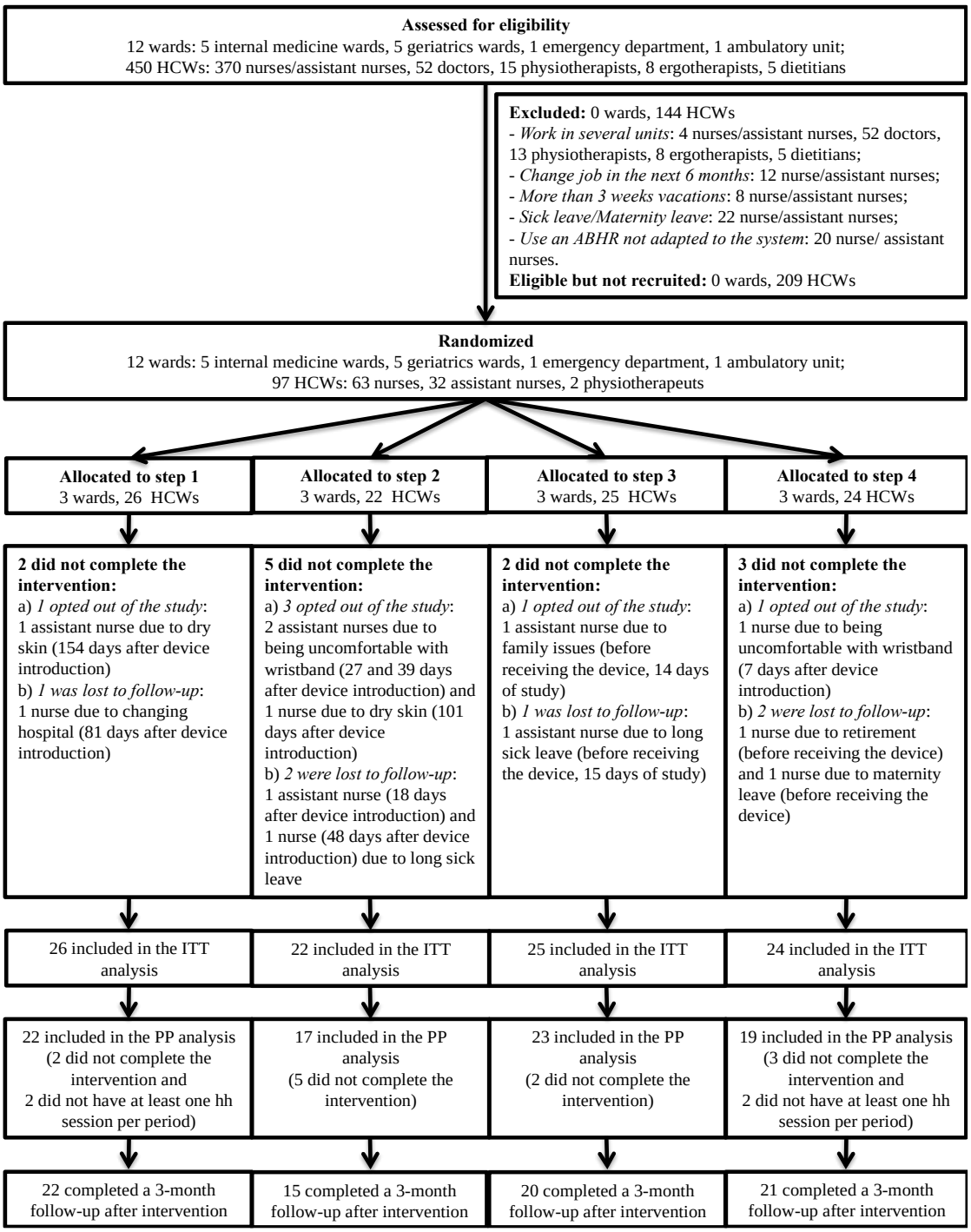

FIGURE 9. Flowchart of the SmartRub® study, Geriatric Hospital, Geneva University Hospitals, June-December 2019.

TABLE 9. Description of wards' and healthcare workers' characteristics by group at baseline; SmartRub® study, Geriatric Hospital, Geneva University Hospitals, June-December 2019.

	Group 1 (4 months of intervention)	**Group 2 (3 months of intervention)**	**Group 3 (2 months of intervention)**	**Group 4 (1 month of intervention)**
Wards	**(n=3)**	**(n=3)**	**(n=3)**	**(n=3)**
Type of ward, n (%)				
Ambulatory	1 (33.3)	0	0	0
Geriatric	1 (33.3)	2 (66.7)	1 (33.3)	2 (66.7)
Medical	1 (33.3)	1 (33.3)	1 (33.3)	1 (33.3)
Emergency	0	0	1 (33.3)	0
Number of inpatient beds	57	93	59	80
Number of HCWs working in patient care	77	107	93	93
Participating HCWs, n	**26**	**22**	**25**	**24**
Women, n (%)	19 (73.1)	18 (81.8)	23 (92.0)	20 (83.3)
Mean age (SD; median, IQR)	41.6 (11.2; 41.5, 35-52)	44.6 (9.2; 45, 36-53)	40.2 (12.6; 39, 27-52)	41.9 (12.2; 43, 30.5-53)
Professional categories, n (%)				
Nurses	19 (73.1)	12 (54.5)	14 (56.0)	18 (75.0)
Auxiliary Nurses	7 (26.9)	9 (40.9)	10 (40.0)	6 (25.0)
Physiotherapists	0 (0.0)	1 (4.6)	1 (4.0)	0 (0.0)
Part-time work, n (%)	12 (46.1)	14 (63.6)	15 (62.5)	14 (58.3)
Mean experience duration in years (SD; median, IQR)	17.2 (11.9; 18, 8-28)	20.2 (10.7; 20, 13-30)	13.6 (11.4; 11.5, 2.5-25)	17.9 (12.5; 22; 3-26)
Mean delay since last HH training (SD; median, IQR)	3.0 (4.2; 1, 0-2)	3.9 (5.2; 1, 1-5)	2.2 (2.5; 2, 1-2.5)	5.8 (5.4; 3, 1-10)
Category of hand size, n (%)*				
Small	11 (42.3)	11 (50.0)	19 (76.0)	13 (54.2)
Medium	6 (23.1)	6 (27.3)	5 (20.0)	5 (20.8)
Large	9 (34.6)	5 (22.7)	1 (4.0)	6 (25.0)
Type of ABHR, n (%)				
Rinse	16 (61.5)	11 (50.0)	11 (44.0)	17 (70.8)
Gel	10 (38.5)	11 (50.0)	14 (56.0)	7 (29.2)

* Category of hand size was defined in reference 18.

ABHR: alcohol-based handrub; IQR: inter-quartile range; HH: hand hygiene; HCW: healthcare workers; SD: standard deviation.

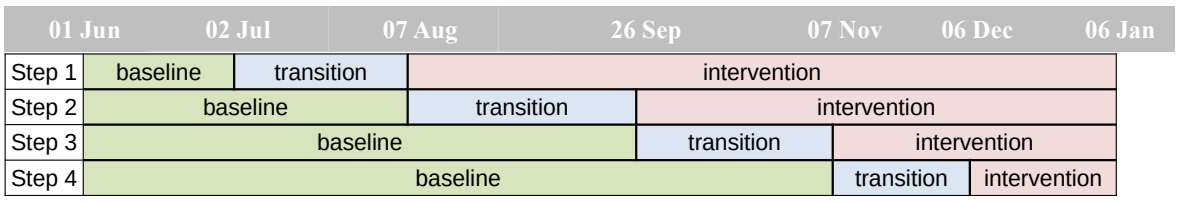

FIGURE 10. Staring dates and length of periods of the SmartRub® study, Geriatric Hospital, Geneva University Hospitals, June-December 2019.

TABLE 10. Length of the study, number of hand hygiene opportunities and actions observed and hand hygiene compliance in total, by group and period; SmartRub® study, Geriatric Hospital, Geneva University Hospitals, June-December 2019.

	Group 1 (4 months of intervention)	**Group 2 (3 months of intervention)**	**Group 3 (2 months of intervention)**	**Group 4 (1 month of intervention)**	**Total**
Number of study days, n					
Baseline	32	68	118	160	378
Transition	36	50	42	29	157
Intervention	146	96	60	31	333
Number of opportunities, n	1986	1229	1731	1932	6878
Number of opportunities per period, n					
Baseline	229	381	848	1210	2668
Transition	394	318	332	378	1422
Intervention	1363	530	551	344	2788
Number of actions, n	1362	847	1180	1107	4496
Number of actions per period, n					
Baseline	181	277	612	707	1777
Transition	299	229	224	213	965
Intervention	882	341	344	187	1754
Compliance with HH, % (95% CI)	68.6 (66.5-70.6)	68.9 (66.2-71.5)	68.2 (65.9-70.4)	57.3 (55.1-59.5)	65.4 (64.2-66.5)
Compliance with HH per period, % (95% CI)					
Baseline	79.0 (73.2-84.1)	72.7 (67.9-77.1)	72.2 (69.0-75.2)	58.4 (55.6-61.2)	66.6 (64.8-68.4)
Transition	75.9 (71.4-80.0)	72.0 (66.7-76.9)	67.5 (62.1-72.5)	56.3 (51.2-61.4)	67.9 (65.4-70.3)
Intervention	64.7 (62.1-67.2)	64.3 (60.1-68.4)	62.4 (58.2-66.5)	54.4 (48.9-59.7)	62.9 (61.1-64.7)
Follow-up	63.6 (57.2-69.9)	63.2 (55.0-71.4)	63.9 (57.6-70.2)	62.9 (56.2-69.6)	63.0 (60.1-66.8)
Compliance with HH per month, % (95%CI)					
Month 1	79.0 (73.2-84.1)	73.3 (66.0-79.7)	72.6 (66.3-78.4)	68.8 (62.2-75.0)	
Month 2	75.9 (71.4-80.0)	72.2 (65.7-78.2)	72.0 (67.0-76.6)	56.1 (50.9-61.3)	
Month 3	68.6 (64.0-72.9)	72.0 (66.7-76.9)	72.0 (66.2-77.2)	58.1 (52.9-63.2)	
Month 4	66.8 (61.7-71.5)	75.7 (68.8-81.7)	67.5 (62.1-72.5)	53.5 (47.2-59.7)	
Month 5	58.9 (53.3-64.3)	63.1 (57.0-69.0)	62.9 (58.0-67.7)	56.3 (51.2-61.4)	
Month 6	62.1 (55.5-68.4)	42.7 (31.8-54.1)	61.2 (53.0-69.0)	54.4 (48.9-59.7)	

HH: hand hygiene.

Overall, 750 direct HH observation sessions were performed, resulting in a total of 6'878 HH opportunities. The median duration of each HH session was 18 minutes (IQR 15-23). We observed a median of 72 HH opportunities (IQR 61-84) per HCW during a median of 8 (IQR 7-9) observation sessions.

Hand hygiene compliance (95% CI) during baseline, transition and intervention periods was 66.6% (64.8-68.4), 67.9% (65.4-70.3) and 62.9% (61.1-64.7), respectively. Compliance by group, study period and month are summarised in Table 10 and Figure 11.

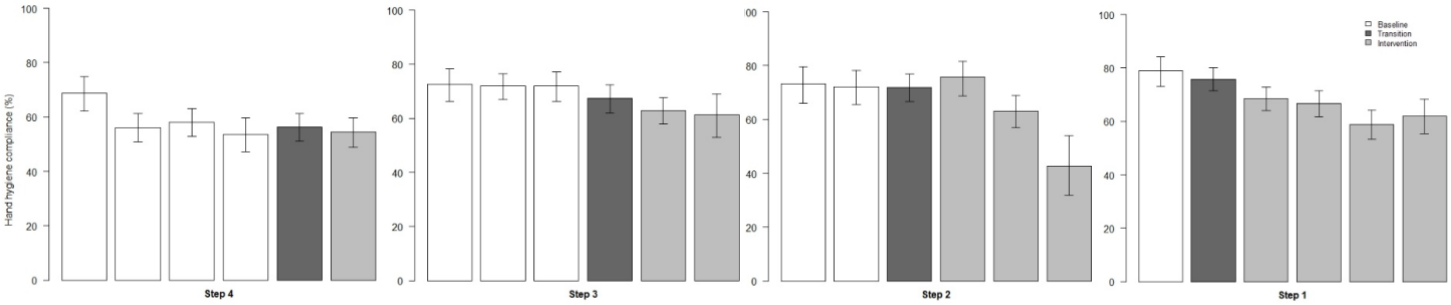

FIGURE 11. Hand hygiene compliance by periods, months and group of the SmartRub® study, Geriatric Hospital, Geneva University Hospitals, June-December 2019.

After adjusting for covariates, HH compliance was not significantly different between baseline and intervention periods (p=0.235) in the ITT analysis. The randomisation group which took the different lengths of intervention into account was not associated with a change in HH compliance (interaction term between study period and group p=0.387). Thus the interaction term was not included in the final model. However, overall HH compliance was significantly higher in groups 1, 2 and 3 than in group 4 (p=0.015). HH compliance was inversely correlated to study duration (study days before individual HH observation), age, and workload (Table 11).

In the per-protocol analysis, 81 (83.5%) HCWs were analysed: 22 (91.7%) in group 1, 17 (77.3%) in group 2, 23 (92%) in group 3, and 19 (79.2%) in group 4. The fitted model confirmed that overall HH compliance to the "5 moments" was not significantly different between intervention and baseline periods (odds ratio 0.99, 95% CI 0.71-1.40 p=0.999).

TABLE 11. Effect of real-time feedback provided by SmartRub® on compliance with hand hygiene across calendar time and exposure to the intervention[1]; SmartRub® study, Geriatric Hospital, Geneva University Hospitals, June-December 2019.

	OR	95%CI	p-value
Reference: Baseline			
Intervention	1.03	0.75-1.42	0.854
Reference: Group 4 (1 month of intervention)			
Group 3 (2 months of intervention)	**1.59**	**1.14-2.23**	**0.006**
Group 2 (3 months of intervention)	**1.61**	**1.15-2.26**	**0.005**
Group 1 (4 months of intervention)	**1.47**	**1.03-2.12**	**0.035**
Days since the start of the study until the HH observation	**0.997**	**0.994-0.998**	**<0.001**
Age in years at baseline	**0.97**	**0.95-0.99**	**0.015**
Male sex	0.77	0.57-1.04	0.084
Aux. Nurse (vs. nurses)	1.10	0.84-1.44	0.488
Experience in years	1.02	0.99-1.05	0.061
Years since last hand hygiene training	0.99	0.96-1.02	0.483
Medical/emergency ward (vs. ambulatory/geriatric)	1.20	0.82-1.74	0.343
Part-time (vs. full-time)	0.81	0.63-1.02	0.077
Gel (vs. rinse)	1.07	0.84-1.35	0.590
Work load (mean opp. per minute)	**0.29**	**0.20-0.41**	**<0.001**

[1]Mixed logistic regression model with nested random effect: group>ward>healthcare worker>session

Ninety-three of the included 97 HCWs (95.9%) wore SmartRub® and contributed electronic data. The bottles and bracelets recorded 65'818 and 45'813 actions in total (Table 12). The individually applied ABHR volume increased during the intervention period (feedback on) [median (IQR): 1.71 ml (1.01-2.76)] as compared to the transition period (feedback off) [1.12 mL (0.76-1.68)]. Correct HH actions (95% CI), as per correct application of volume of ABHR only, increased from 10.2% (9.8-10.6) during the transition period to 30.5% (30.0-30.9) during the intervention period (Table 12).

After adjusting for covariates, the effect of the intervention on higher applied ABHR volume was significant in every group (see below), but the magnitude of effect differed by group (interaction term, p<0.001; Table 13). The applied ABHR volume increased by a mean of 0.92 mL/per action (95% CI 0.88-0.95, p<0.001) in group 1 (4 months of intervention), 0.51mL/per action (95% CI 0.47-0.55, p<0.001) in group 2 (3 months), 0.64 mL/per action (95% CI 0.60-0.69, p<0.001) in group 3 (2 months), and 0.62 mL/per action (95% CI 0.55-0.68, p <0.001) in group 4 (1 month).

TABLE 12. Hand hygiene practices: description of number of actions, volume of applied alcohoç-based handrub and duration of hand rubbing, as well as overall correctness of each action per group and period as recorded by SmartRub®; SmartRub® study, Geriatric Hospital, Geneva University Hospitals, June-December 2019.

	Group 1 (4 months of intervention)	**Group 2 (3 months of intervention)**	**Group 3 (2 months of intervention)**	**Group 4 (1 month of intervention)**
Bottle actions (n)				
Transition	10'139	7'319	6'317	2'747
Intervention	22'747	8'573	5'503	2'473
Mean bottle actions per HCW (SD; median, IQR)				
Transition	389.9 (246.0; 340.5, 20.02-488.0)	365.9 (292.1; 268.5, 165.5-524.5)	287.1 (215.2; 218, 150.0-417.0) 250.1	130.8 (109.8; 109, 44-206)
Intervention	874.9 (555.8; 725.0, 497.0-1067.0)	476.3 (290.7; 463, 236-624)	(179.9; 282, 53-398)	117.8 (71.7; 97, 78.0-157)
Mean volume of applied ABHR per hand rubbing action, mL (SD; median, IQR)				
Transition	1.20 (0.75; 1.02, 0.72-1.49)	1.84 (1.75; 1.44, 0.92-2.22)	1.26 (0.79; 1.05, 0.78-1.55)	1.29 (0.88; 1.03, 0.69-1.56)
Intervention	1.99 (1.15; 1.73, 1.03-2.81)	2.17 (1.5; 1.86, 1.10-2.93)	1.69 (1.37; 1.32, 0.87-2.34)	1.97 (1.21; 1.82, 0.87-2.79)
Overall correctness of applied AHBR volume* (95%CI, %)				
Transition				
Intervention	3.7 (3.3-4.1) 30.6 (30-31.2)	18.1 (17.3-19) 32.6 (31.6-33.6)	11.5 (10.7-12.3) 24.6 (23.4-25.7)	10.2 (9.1-11.3) 35.3 (33.4-37.2)
Wristband actions (n)				
Transition	8'101	3'160	4'103	1484
Intervention	17'742	5'798	3'560	1865
Mean wristband actions per HCW (SD; median, IQR)				
Transition	311.6 (166; 310.0, 188-414)	175.6 (125.8; 119.5, 93-248.0)	178.4 (130.6; 142, 83-286)	82.4 (63.6; 60.5, 24-150)
Intervention	682.4 (455.9; 571.5, 353-826)	322.1 (163.1; 337.5, 189-445)	169.5 (138.5; 138, 48-264)	88.8 (57.1; 77, 56-102)
Mean duration of hand rubbing performed per HCWs, seconds (SD; median, IQR)				
Transition	8.67 (4.87; 7, 4.5-11)	8.88 (5.85; 6.5, 4.5-11)	7.21 (4.21; 5, 4.5-8.5)	9.1 (5.72; 7, 4.5-11.5)
Intervention	10.24 (5.96; 8, 5-15)	9.87 (6.23; 7, 4.5-15)	10.41 (6.71; 8, 4.5-15.5)	12.39 (6.67; 12, 5.5-17)
Overall correctness of HH duration#, seconds (95%CI, %)				
Transition	12.2 (11.5-13)	15.1 (13.9-16.4)	6 (5.3-6.8)	14.4 (12.6-16.2)
Intervention	25.6 (25-26.3)	25.6 (24.5-26.7)	27.8 (26.4-29.3)	41.1 (38.8-43.3)
Mean calculated hand size volume of ABHR per HCW, mL (SD; median, IQR)	2.58 (0.45; 2.38, 2.20-3.06)	2.47 (0.48; 2.29, 2.17-2.46)	2.26 (0.22; 2.26, 2.14-2.37)	2.50 (0.55; 2.31, 2.18-3.04)

*Defined by a volume of ABHR above the estimated hand sized volume.

Defined as at least 15 seconds of hand friction.

ABHR: alcohol-based handrub; CI: confidence interval; IQR: inter-quartile range; HCW: healthcare workers; HH: hand hygiene; SD: standard deviation.

TABLE 13. Effect of the real-time feedback provided by SmartRub® on the individual volume of applied alcohol-based handrub across calendar time and exposure to the intervention[1]; SmartRub® study, Geriatric Hospital, Geneva University Hospitals, June-December 2019.

	Coeficient	95%CI	p-value
Reference: volume used in transition period of each group			
Volume used in intervention period in group 4	0.62	0.55-0.68	**<0.001**
Volume used in intervention period in group 3	0.64	0.60-0.69	**<0.001**
Volume used in intervention period in group 2	0.51	0.47-0.55	**<0.001**
Volume used in intervention period in group 1	0.92	0.88-0.95	**<0.001**
Days since the start of the transition period	**-0.0007**	**-0.001--0.0004**	**<0.001**
Age at baseline	-0.02	-0.05-0.01	0.261
Male sex	-0.77	-0.49-0.33	0.710
Assistant nurse (vs. nurses)	0.27	-0.06-0.59	0.106
Work experience in years	0.01	-0.02-0.05	0.463
Years since last hand hygiene training	-0.02	-0.05-0.02	0.335
Medical/emergency ward (vs. ambulatory/geriatric)	-0.33	-0.66-0.007	0.055
Part-time (vs. full-time)	-0.20	-0.505-0.10	0.190
Gel (vs. rinse)	0.19	-0.14-0.52	0.253

[1]Mixed linear regression model with nested random effect: group>wards>healthcare workers.

CI: confidence interval.

Notably, the duration of hand rubbing also increased during the intervention period [median (IQR), 8 sec (4.5-15.5) as compared to the transition period (6.5 sec, 4.5-10.5). Correct HH actions (95% CI), as per correct duration only, increased from 11.4% (10.6-11.9) in the transition period to 26.9% (26.4-27.4; Table 12) during the intervention period. After adjusting for covariates, the effect of the device on the duration of hand rubbing was significant in every group, but the magnitude of effect differed by group (interaction term, p<0.001; Table 14). Mean duration of hand rubbing increased by 2.96 seconds/per action (95% CI 1.43-1.69 p<0.001) in group 1, 1.36 seconds/per action (95% CI 1.10-1.61 p < 0.001) in group 2, 2.89 seconds/per action (95% CI 2.64-3.13, p<0.001) in group 3, and 2.65 seconds/per action (95% CI 2.29-3.02, p<0.001) in group 4.

TABLE 14. Effect of the real-time feedback provided by SmartRub® on duration of hand rubbing across calendar time and exposure to the intervention[1]; SmartRub® study, Geriatric Hospital, Geneva University Hospitals, June-December 2019.

	Coeficient	95%CI	p-value
Reference: duration of hand rubbing in transition period of each group			
Duration of hand rubbing in intervention period in group 4	3.18	2.82-3.54	**<0.001**
Duration of hand rubbing in intervention period in group 3	3.63	3.38-3.88	**<0.001**
Duration of hand rubbing in intervention period in group 2	2.55	2.28-2.83	**<0.001**
Duration of hand rubbing in intervention period in group 1	2.96	2.78-3.14	**<0.001**
Days since the start of the transition period	**-0.018**	**-0.02--0.017**	**<0.001**
Age at baseline	-0.05	-0.22-0.11	0.547
Male sex	-0.45	-2.54-1.63	0.669
Assistant nurse (vs. nurses)	1.33	-0.25-2.90	0.099
Work experience in years	0.05	-0.12-0.22	0.546
Years since hand hygiene training	0.013	-0.14-0.17	0.864
Medical/emergency ward (vs. ambulatory/geriatric)	-0.44	-1.89-1.01	0.553
Part-time (vs. full-time)	-0.45	-1.92-1.04	0.559
Gel (vs. rinse)	-0.30	-1.93-1.32	0.715

[1]Mixed linear regression model with nested random effect: group>wards>healthcare worker.

CI: confidence interval.

Follow-up HH observations, at 3 months after the end of the intervention, were performed on 78 (80.4%) HCWs. A total of 790 opportunities were observed. The overall HH compliance was 63.4% (95% CI 60.1-66.8). Compliance at follow-up was not different from baseline (p=0.097) or intervention periods (p=0.795). Compliance per group was: 63.6% (57.2-69.9) in group 1, 63.2% (55.0-71.4) in group 2, 63.9% (57.6-70.2) in group 3 and 62.9% (56.2-69.6) in group 4 (p=0.997).

The proportion of participants experiencing any adverse event was 19 (19.6%). All events were skin related: 6 reports on dry skin due to changing from gel to rinse (which subsided after changing back to gel), 3 reports on itching from the bracelet (which subsided after temporarily stopping bracelet use), and 9 reports of dry skin due to using more ABHR than previously. These adverse events directly motivated two drop-outs (1 related to bracelet itching and 1 to dry skin). As judged by investigators, no serious adverse event was reported.

Device satisfaction and perception was evaluated by questionnaires and focus group discussions. Results are reported elsewhere (P 374 ICPIC 2019). [111]

Discussion

This stepped wedge cluster randomized clinical trial tested the effect of a wearable electronic device to improve HH (SmartRub®) by providing feedback on appropriately applied ABHR volume and sufficient duration of hand rubbing on both compliance and quality of hand hygiene practices.

Our study did not show significant effects of SmartRub® on HH compliance, after adjusting for various confounders and time-dependent variables. However, wearing SmartRub® did improve the quality of the hand hygiene action, with a significant increase in both ABHR volume applied and in the duration of hand rubbing.

Intriguingly, we observed a gradual decline in HH compliance throughout the study, from 73.5% (95% CI 70.5-76.5) during the first month to 56.6% (95% CI 53.2-60.1) during the last month. This time trend was unexpected and, in all likelihood, hampered the assessment of the effect of SmartRub® on HH compliance.

We hypothesise a significant Hawthorne effect when HCWs were observed for the first time during the baseline period, which resulted in overestimated HH compliance. The same auditors performed repeated observations of the same HCWs, and HCWs may have become "used" to them over time. Thus, the presence of the auditors may not have sparked immediate behaviour change in subsequent observations. This would be associated with regression of an initial Hawthorne effect, resulting in HH compliance falling back to routine behaviour. This phenomenon of "habituation" has been previously described [63].

In contrast, monitoring of HCWs behaviour with a wearable device might be less prone to be affected by those variables. This advantage probably allowed SmartRub® to detect the true effect of the device feedback on HH quality. Notably, we observed a significant relative increase of around 50% in the mean volume of applied ABHR per action, and prolonged hand rubbing by 20%. It has been widely shown that volume of ABHR and duration of hand rubbing are major determinants of the microbial efficacy of the HH action [64,81,84,85,107].

Although we did manage to improve the quality of the HH action, we did not achieve full adherence to the optimized parameters that we have set. SmartRub® is a novel device and, not surprisingly, we faced a series of technical challenges throughout the study. Problems included recurrent, random inactivation of the devices (more frequent with the wristband) and loss of data due to errors in data transfer to the server. These technical problems prevented us from calculating pre-determined secondary outcomes, such as the frequency of hand hygiene events, adherence to device use and ward level ABHR

consumption. The lack of such data did not allow analysis if trends of observed HH compliance were different from trends of performed HH events.

The introduction of a wearable device to improve HH during daily clinical practice is challenging [115]. In order to implement such systems, issues related with transparency and confidentiality in the use of the data, [116] accuracy, adaptation to clinician workflow, budget and organisational culture need to be taken into account [115]. In spite of the technical challenges, we observed good buy-in from HCWs, as the proportion of drop-outs related to the use of SmartRub® was small (only three HCWs dropped out because they did not feel comfortable wearing the bracelet). In a questionnaire performed after the study, [111] the majority (78.6% 55/70) of HCWs providing feedback agreed that SmartRub® was a helpful reminder for correct HH, and 57% (40/70) would continue to use it after the trial. However, 27% (19/70) were concerned about confidentiality issues (due to electronic surveillance and data transfer), and 23% (16/70) reported that using the wristband could interfere with their clinical activities [111].

The most recent systematic review on the effect of monitoring technologies on HH adherence suggested that electronic devices have the potential to change HH behaviour [116]. However, the overall quality of studies on this topic was poor [116–118]. Indeed, there is a need to address uncertainties about the accuracy and cost-effectiveness of such systems, [117] and to conduct clinical trials with solid controlled study designs that include system-independent and relevant outcomes [118]. We believe that our stepped wedge, cluster-randomized clinical trial contributed to raise the standards of methodological quality of studies on this topic. Importantly, we tested a device that has gone through an in-depth sensitivity and specificity analysis [111].

We are confident that our study is a proof of concept of the benefits of a wearable device on improving the quality of HH in clinical practice, and that it opens perspectives for new strategies on HH improvement. To the best of our knowledge, and based on published literature, other HH monitoring devices focus on dispensing events or proxies for HH indications (i.e., entry and exit of rooms, proximity of a HCW to a patient bed) [61]. SmartRub® is unique in its potential to interact directly with the HCW during HH, in real time and during daily routine.

Conclusions

Wearing SmartRub® led to an improvement of the quality of the HH action, as measured by an increase in the volume of ABHR per hand action and in the duration of hand rubbing. However, we cannot evaluate the effect of SmartRub® on HH compliance because of a gradual decline in HH compliance unrelated to the use of the device and probably associated with a high magnitude Hawthorne effect at baseline. In order to reach its full potential, this innovative device merits further development and adequate testing in future studies.

Discussion

The first part of this project aimed to optimize the current WHO "How to handrub" recommendation. We have shown, in 4 laboratory-based experiments with 74 HCWs, that it is possible to simplify and customize the hand hygiene action while maintaining its antimicrobial efficacy. Indeed, we propose a shorter hand hygiene action, with 15 seconds of hand friction [84], starting by rubbing the fingertips in a modified WHO 6 steps technique [83] and using a hand size customized volume of ABHR [81,85]. This modified hand hygiene action was also tested with different types of bacteria and concentration ranges, therefore adding a layer of reassurance in support of the proposed changes [85].

In the second part of this project, we investigated whether using a wearable electronic device (SmartRub®) to deliver our new recommendation on "How to handrub" would increase HCWs' compliance with the "5 moments" of HH and the quality of the HH action itself. We performed a stepped wedge, cluster randomized clinical trial including 97 HCWs and with a duration of 7 months. Wearing SmartRub® improved the quality of hand hygiene action ("How to handrub"), with HCWs performing more prolonged hand friction and using higher volumes of ABHR. However, the device failed to show any effect on the compliance with the "5 moments" ("When to handrub").

As mentioned before, we started by developing a more evidence-based and optimized HH action. In our first experiment, we aimed to determine the threshold of hand contamination that makes bacterial cross-transmission between hands unlikely [74]. This had to be the first step in this project, considering the current lack of scientific consensus on the bacterial reduction required by an adequate hand hygiene action, i.e. one that prevents microorganism cross-transmission.

We built a model to study the transmission of bacteria between hands of HCWs and showed that hands with less than 1 $\log_{10}$ of *E. coli* contamination do not seem to be vehicles for cross-transmission between patients [74]. However, this model was only tested using one species of Gram-negative bacteria (*E. coli*) and with a limited number of experiments at the lower concentration range.

A few previous studies have addressed this topic [119–121]. Ehrenkranz and Alonso [119] performed a study where HCWs' hands were contaminated by palpating the femoral artery of patients known to be carriers of Gram negative bacteria. HCWs then performed HH using soap and water or ABHR, and finally manipulated a sterile urinary catheter. Handwash

with soap failed to prevent catheter contamination by Gram negative bacteria in 11 out of 12 experiments, whereas ABHR failed in only 2 of 12. Importantly, there was a correlation between the concentration of bacteria in the skin of the patient and the probability of transmission to the hands of HCWs: bacterial concentration > 5.5 x 10^3/ml established transient colonization in 23 of 30 exposures following soap handwash while a concentration of < 3.5 x 10^3/ml established colonization in only 1 of 22 similar exposures ($p<0.001$). The authors did not seek to determine the threshold of "no transmission" but they nicely demonstrated a correlation between level of contamination and likelihood of cross-transmission. In addition, the advantages of ABHR in relation to soap and water handwash were also clearly demonstrated. However, the differences in microbiological methods prevent a meaningful comparison with our results.

Interestingly, we have recently performed an experiment on the same topic, [111] which consisted in performing the following sequence: 1. artificial hand contamination with *S. aureus*; 2. optimized HH (based on the parameters developed in this PhD project) or no HH; 3. manipulation of an aseptic infusion set; 4. sampling of the infusion set. The median amount of *S. aureus* on the fingertips after contamination was $10^{6.8}$ cfu (or 6.8 $\log_{10}$). Without HH, bacteria were detected in the final infusion set after all 20 trials (range: 25 to 25 800 cfu), whereas with HH, bacteria were not detected after any of the 20 trials ($p<0.001$). We demonstrated that even with a heavy contamination of the fingertips with *S. aureus*, our optimized HH method prevented transmission of bacteria to the infusion set. However, we did not sample the hands of the volunteers after HH and before touching the infusion set to assert their level of contamination. Therefore, we do not know threshold of hand contamination with *S. aureus* resulting in no-transmission to the infusion set.

Humidity of hands or environment was not taken into account in our cross-transmission model but could have affected the transmission threshold, and therefore should be further explored in future studies [74]. Indeed, studies performed in the 70's and 80's highlighted the importance of moisture in the transmission of bacteria from contaminated fabrics to HCWs hands and vice-versa [120,121]. More recent studies confirmed and expanded these findings to include other types of surfaces such as skin and food [122,123].

As we have seen, there are other possible models of cross-transmission to investigate the microbiological concept of safe hands. We do believe that it is useful to continue developing the microbiological concept of safe hands, as this is essential to improve the HH action.

As a first stage in the optimization of the recommended HH action in this PhD project, we investigated the effect of "rubbing the fingertips" as the first instead of last step in the traditional 6-step sequence endorsed by WHO (Figure 1). We observed that "rubbing the fingertips" first led to greater reductions of hand contamination in a laboratory experiment [83]. Rubbing the fingertips first ensures that there is enough ABHR to adequately cleanse this area, which is arguably the most contaminated hand part in clinical practice [92]. In addition, instructing HCWs to start HH by rubbing the fingertips might help remembering that step, ensuring that it is performed during daily clinical activities. This is particularly important because the adherence to the overall 6 steps and the fingertips step is very low (respectively 8.5% and 19.5% in a prospective study in Basel, Switzerland) [77]. Our study already led to a local change in clinical practice at the HUG, where it is now recommended to start the HH action by rubbing the fingertips.

This proposed change in the sequence of the WHO technique is part of a wider discussion about the simplification of the HH technique and its effect on the antimicrobial efficacy. A recent systematic review investigated the available evidence on this topic, and showed a lack of consensus [82]. Two studies compared the antimicrobial efficacy of the WHO technique versus the CDC technique (hand rubbing with alcohol covering all hand surfaces in no particular order [37]) during clinical activities [93,98]. Chow et al. [98] reported no difference in the effectiveness of the WHO 6-step technique compared to the CDC technique, whereas Reilly et al. [93] found the WHO 6-step technique to be more effective. Tschudin-Sutter *et al.* [94,109] tested the antimicrobial efficacy of the WHO technique against a new 3-step action (covering all hand surface with ABHR, rotational rubbing of fingertips in the palm of the alternate hand and rotational rubbing of both thumbs) in a laboratory-based study. The authors found that the new simplified 3-step technique was more effective in reducing bacteria in the hands of HCWs than the WHO 6-step technique [94,109]. More recently, the same authors published a clinical trial reporting no significant differences in antimicrobial efficacy between the 2 techniques, during clinical care [109].

The importance of the contamination of fingertips and thumbs in clinical care might help explaining why a simplified 3-step technique including the fingertips appears to be at least equivalent to the original WHO technique [94,109], whereas one simplified technique not explicitly including fingertips' rubbing was inferior in one of two studies [93,98].

All studies we have just mentioned used the "glove juice" method to sample the hands of HCWs. This method samples the entire hand, and is recommended by the American norm to introduce ABHR products in the market [87]. As we have previously discussed, our

laboratorial studies used the fingertips sampling method. The fact that these studies, using a different sampling method, also point to the importance of the fingertips, reinforces our results.

One criticism regarding the CDC technique and the new simplified 3-step technique, is the lack of specific guidance on how to perform the step "covering all surfaces of the hands" with ABHR. Indeed, it is difficult to understand how this action can be made with only one "step". This guidance might create heterogeneity in practices and confusion among HCWs.

Furthermore, several other studies [72,73,93] have shown that the adherence to the correct WHO 6-step technique was significantly related to its microbiological efficacy. This evidence highlights the importance of training HCWs in the correct performance of the HH technique, in order to achieve best results. All participants in our laboratory-based studies were experts in IPC and hand hygiene. This constitutes an advantage when testing variations in the HH technique but might simultaneously also limit the generalizability of our findings.

In the following stage of our journey to optimize the HH action, we investigated whether a shorter duration of hand friction could have an efficacy similar to the currently recommended standard of 30 seconds. Importantly, we demonstrated that performing hand friction for 15 seconds was non-inferior to 30 seconds. We believe that this finding might have important implications for the practice of hand hygiene. Reducing the duration of an optimal hand hygiene gesture could help improving compliance, because lack of time appears to be a major factor driving non-compliance [32].

Our findings on the feasibility of shortening the duration of hand friction without sacrificing antimicrobial efficacy are supported by a growing body of literature. Kampf and Hollingsworth [103] have shown that a 15 seconds contact time obtained more than 5 $\log_{10}$ bactericidal activity in a great sample of clinically meaningful bacteria. However, they did not compare this reduction with that obtained with 30 seconds. Darhan *et al*. [91] showed that a contact time of 15 seconds was equivalent to 30 seconds in terms of bacterial reduction in the hands of HCWs. However, this experiment did not mimic the usual application of ABHR in clinical practice. Bacteria were spread only in the fingertips of HCWs and 0.4 mL of ABHR was used for rubbing the thumbs against the fingertips.

Following the publication of our article, Kramer *et al*. [99] confirmed the antimicrobial equivalence of 15 seconds as compared to 30 seconds of hand friction, for

different types of ABHRs. This replicated and expanded our findings, as we only used the standard product of the EN 1500 (60% v/v isopropanol) as ABHR.

Harnoss *et al.* [108] recently published a cross-over trial where nurses working at a Gynaecology Department were individually randomized to a recommendation of hand rubbing for 15 or 30 seconds. The authors evaluated what they called the "accumulated bioburden", by sampling the hands every hour during a day of clinical practice. They found no differences in the bioburden between the groups, which further supports the equivalence of recommending HCWs to rub hands for only 15 seconds.

Some studies investigated the efficacy of hand rubbing from a perspective of hand surface coverage by ABHR, instead of microbiological reduction [124,125]. Reilly *et al.*[93] did not find a correlation between hand surface coverage and microbiological outcomes, but overall this has not been sufficiently studied. Nevertheless, Paula *et al.* [125] showed that there were no differences in hand surface coverage by 3 mL of ABHR after 15 or 30 seconds of hand friction according to the WHO 6-step technique. Importantly, they observed an improvement of hand surface coverage in both groups after training the participants in the technique. Again, this highlights the important of training HCWs in the technique. However, Kampf *et al.* [124] reported more uncovered hand areas following 12 to 17 seconds of hand rubbing as compared 30 seconds, while using the same 3 mL of ABHR and the WHO 6 steps technique. However, do not know whether these differences were significant because the authors did not report statistical analysis of their results.

When hand rubbing using the volumes of ABHR recommended by either the international norms or our hand size customized approach, hands will not dry out after 15 seconds [101,126]. Indeed, volumes of ABHR that allow drying out in 15 seconds are for the most part insufficient for an adequate hand hygiene action [97]. This is a limitation to the proposed 15 seconds of hand rubbing. However, it is important to highlight that even volumes of ABHR that dry out in 30 seconds appear to be too small, as they fail to pass the EN 1500 norm of antimicrobial efficacy [97]. As we have discussed, we believe that it will only be possible to solve this issue if faster drying ABHR products arrive on the market.

There is widespread consensus on the importance of volume of ABHR on the antimicrobial efficacy of the hand hygiene gesture [35,127,128]. In addition, our group previously demonstrated that the hand size of a HCW influences the antimicrobial efficacy of a given volume of ABHR [64,81]. This is a concept we have used in our laboratory [85] and clinical studies. The personalized volume of ABHR used in our last laboratorial

experiment and on our clinical trial was calculated based in previous studies investigating the influence of hand size on the efficacy of a given volume of ABHR [81] and on cross-transmission of *E. coli* between the hands of HCWs [74]

However, other groups have failed to confirm an association between hand size, volume of ABHR and antimicrobial reduction [129,130]. Wilkinson *et al.*[130] commented that the range of hand sizes of the participants in their study was smaller than in ours, and that they have only tested the effect of hand size in the efficacy of more than 3 mL of ABHR, and not for smaller volumes. Goroncy-Bermes *et al.*[129] acknowledged the considerable variation in the $\log_{10}$ reduction in their study, and that the range of hand sizes was so limited that this might have prevented the detection of a possible correlation between hand size and antimicrobial reduction. Both studies might have suffered from a type II error.

Previous studies on the hand hygiene action have focused either on the technique, duration of hand friction or on the volume of ABHR. This allowed the researcher to investigate one component, while controlling for the other elements that were kept uniform across groups. However, this approach is obviously very artificial. In real life, the hand hygiene action is much more complex, with a permanent interaction between its multiple dimensions.

Against this background, we performed an additional laboratorial experiment to investigate the combined effect of the proposed changes to the "How to handrub" (rubbing the fingertips first on a modified WHO 6-step technique, with a volume of ABHR customized for hand size and a shorter duration of hand friction) when cleansing hands contaminated by different bacteria (*S. aureus* and *E. coli*) and with different contamination loads (10^8 or 10^6 cfu/mL) [85]. Indeed, in real-life HCWs are also exposed to many different types of bacteria and contamination levels. To the best of our knowledge, this is the first experiment of this kind to be published.

We found that 15 seconds of hand rubbing was non-inferior to 30 seconds regardless of the type of bacteria and level of contamination. This provided additional support to the proposed reformulation of WHO's "How to handrub", which could be shortened down to 15 seconds without losing antimicrobial efficacy. Our results also offered further validation to the use of a volume of ABHR customized for hand size.

We believe that this kind of integrative approach at the laboratory bench should be used to test future proposed modifications of the "How to handrub". However, laboratory-based research is logically followed by studies performed at the bedside, such as those

published by Reilly *et al*. [93], Chow *et al*. [98] and Tschudin *et al*. [109] where the microbiological efficacy of the HH action was assessed directly during clinical practice.

The findings presented on this PhD thesis, together with the work of other research groups producing new evidence on the same topic, helped igniting a debate on the reformulation of WHO hand hygiene guidelines regarding the "How to handrub". I presented a review of the available evidence on this topic, including the studies in this doctoral project, in an international WHO expert-meeting held in February 2019. The final draft report of this meeting is still under discussion and its publication has been delayed due to the Covid-19 pandemic.

We used a consistent methodology throughout our laboratory-based studies, largely based on the EN 1500 norm, which reinforces the internal and external validity of our findings. Indeed, this consistency allowed us to build upon previous results to design subsequent experiments. In addition, it also allows comparing our results with others in the literature.

However, the EN 1500 [76] norm does also have limitations deserving further discussion. First, its outcomes and non-inferiority margin were developed to compare ABHR products but do not necessarily correspond to clinically relevant endpoints. Second, it endorses the fingertips sampling method, which leaves unsampled other hand areas. However, and has mentioned before, the fingertips are the most contaminated hand area and for this reason also the most important area to sample. In contrast, the ASTM norm [87], which is used in the United States of America, is based on the sampling of the entire hand. In our opinion this approach has the disadvantage of not taking into account the relative importance of different hand parts in cross transmission. Indeed, the back of the hands is not as important as the fingertips, as the last contact more closely with contaminated surfaces and patients. Third, EN 1500 relies in the reduction achieved for *E. coli*, and does not take into account other bacteria or microorganisms such as viruses and fungi.

A Swiss consortium including our group developed a wearable electronic device (SmartRub®) capable of monitoring and providing real-time feedback to HCW about the quality of their HH action (volume of applied ABHR and duration of hand friction). We programmed SmartRub® according with our optimized version of the "How to handrub".

The final step in my PhD project was to investigate the effect of SmartRub® in the quality of HH action ("How to handrub") and on the compliance of HCWs to the "5 moments" of HH ("When to handrub").

Our choice of primary outcome reflected the importance in IPC of finding new strategies to improve compliance with the "5 moments". In addition, and in spite of its limitations, WHO's direct observation method is the most extensively studied method to evaluate the HH "5 moments" [131]. However, as discussed in the draft manuscript of SmartRub®, our study design might have been inadequate to measure the effect of wearing SmartRub® on HH compliance.

The assessment of the compliance with the "5 moments" of HH is a challenging issue. The WHO direct observation method [36], used in our clinical trial, is the gold standard for auditing HH. The most common critique to this method is exactly the overestimation of compliance rates due to the Hawthorne effect, which has an uncertain magnitude [63]. However, if HH observations are performed under the same conditions, the Hawthorne effect is believed to be fairly constant. Thus, we know that HH compliance is being overestimated but we can safely compare results between units or within the same unit over time [35,36,63]. However, in our study we observed the same HCWs over time, which might have led to an "habituation" of HCWs to observers and thus to a gradual fading of the Hawthorne effect. In fact, the repeated observation of the same HCW over time is an unusual design in studies assessing HH compliance. Most studies perform group level (i.e. ward level) HH observations over time, and are therefore less susceptible to this "habituation" effect [60].

Notably, SmartRub® did succeed in improving the quality of the HH action ("How to handrub"). Overall, the proportion of HH actions performed using the correct volume of ABHR and hand rubbing for the correct length of time increase from a baseline of 10% up to around 30%. As we have discussed, these parameters are very important for the reduction of bacterial loads in the hands of HCWs [74,81,83–85]. Importantly, these figures demonstrate the potential of SmartRub® to change the behaviour of HCWs during daily clinical practice, improving the quality of the HH action.

Although we did manage to improve the quality of the HH action, we were far from achieving full adherence to the optimized parameters that we have set. We believe that this was at least partially a result of technical problems and limitations in the SmartRub® device. Further technical improvements are warranted in order to take fully advantage of this tool.

We run focus groups after the conclusion of the trial, and these revealed that 15 seconds duration of hand rubbing and the proposed hand size customized volume of ABHR are still regarded by many HCWs as excessive and cumbersome during daily clinical activities. HCWs explained that they assess the risk of clinical tasks and adapt the volume

of ABHR and duration of hand rubbing to the perceived risk of contamination and transmission. Importantly, HCWs were keener to comply to the recommendations and feedback delivered by SmartRub® in situations with a higher perceived risk of transmission. Overall, SmartRub® was regarded as a useful tool by participants in the trial [111]. In addition, the efforts of our IPC team to optimize the hand hygiene action were highly appreciated by HCWs, and they reported more willingness to comply to the recommendations after understanding their rationale.

The introduction of a volume of ABHR customized for hand size requires the development of novel devices like SmartRub®. Based on our experience, HCWs often underestimate the volume of ABHR required for an adequate hand hygiene action, and this might explain the pervasive utilization of insufficient volumes. While it is still not possible to benefit from precise customization of the volume of ABHR to the hand size in routine clinical practice, we consider that it would be worth introducing this concept in training sessions on HH. Importantly, based on our unpublished qualitative study, HCWs appear to be open to the concept of hand customization and keen to know what would be the ideal volume of ABHR for their hand size.

Other recent studies investigating the impact of a simplification of HH action on HH compliance reported encouraging results. Those studies tested simplified [109] and shorter [99] versions of the HH technique. However, they did not use a device to monitor and deliver their HH instructions and relied on direct observation of the HH action to measure their outcomes. Tschudin-Sutter *et al.*[109] performed a 2-month cluster randomized trial testing the effect of training HCWs on a 3-step HH technique (*versus* the WHO 6-step technique) on HH compliance. They found that the compliance with the 3-step technique was significantly higher than with the 6-step (51.7% *versus* 12.7% $p<0.001$) and that the compliance with the "5 moments" was also higher in the 3-step group (75.9% *versus* 65.0% $p<0.001$). Kramer *et al.*[107] randomly allocated nurses from a Neonatal Intensive Care Unit to a 15 or 30 seconds hand rubbing recommendation. Nurses allocated to the 15 seconds group performed significantly more HH actions than those in the 30 seconds group (7.9±4.3 standard deviation [SD] vs 5.8±2.9 SD per hour; P=0.05). However, there was no difference in the duration of hand rubbing between the groups. These two studies suggested that recommending a simplified HH action might improve the quality of the HH action and the compliance with the "5 moments", or at least increase the frequency of hand rubbing.

Interestingly, both Kramer *et al.*[99] and our SmartRub® study recommended *shorter* durations of hand rubbing (15 seconds instead of 30 seconds), but resulted in HCWs

performing HH more prolonged hand rubbing as compared to the baseline. This suggests that “lowering the bar” in HH recommendations in this context might lead to better performances. This observation has potentially important implications for behavioural change in healthcare and needs to be better understood. Importantly, it helps dispelling the fears that any “lowering” of the current standards would lead to even further deteriorations of HCWs’ performance.

Conclusion and future perspectives

Hand hygiene is the cornerstone of Infection prevention and control, possibly constituting the single most effective measure to avoid healthcare associated infections [35]. However, the implementation of this apparently simple action is still a major challenge in healthcare institutions worldwide.

We worked towards the development of a shorter, customized, microbiologically equivalent version of the current HH action endorsed by the WHO. A growing body of literature suggests that optimizing the recommended "How to handrub" is a promising strategy to improve real-world practices regarding the HH action [107,109].

An ideal "How to handrub" is one that prevents cross-transmission of microorganisms and is easy to remember and perform. The modifications proposed in this PhD project are in accordance with those goals. A shorter duration of hand friction (15 seconds) is better adapted to the workflow in clinical practice. Recommending to rub the fingertips first ensures that the most contaminated part of the hand is not forgotten, and is properly cleaned. Finally, the customization of the volume of ABHR to hand size acknowledges the impact of different hand sizes in the requirements of ABHR.

However, there are still many challenges ahead. In terms of methodology, there is no international consensus about efficacy "cut-offs" based on transmission thresholds. In addition, it is difficult to design controlled, laboratory-based studies closely mimicking conditions of clinical practice. Broadly speaking, it is still uncertain how much the HH action can be simplified without sacrificing efficacy standards. As discussed previously, there is an unmet need for hand hygiene products that dry faster. Finally, implementing hand size customized volume of ABHR to routine clinical practice in different contexts will likely require the development of multiple, novel approaches. These are some of the gaps that remain to be addressed and that therefore deserve further research.

In our cluster randomized clinical trial, SmartRub® led to an improvement in the quality of the HH action, with an increment in both the duration of hand friction and volume of ABHR. However, we failed to detect any effect of SmartRub® on the compliance with the "5 moments". Our study design involved repeated direct observations of HCWs over time. A resulting Hawthorne effect and its fading over time likely affected the measurement of our primary outcome, preventing us from drawing any definitive conclusions. Future clinical trials on this topic could account for the impact of the Hawthorne effect by adding a control arm including HCWs without SmartRub® that would be observed repeatedly over time. This

would allow quantifying the magnitude of change in HH compliance attributable to the fading of the Hawthorne effect and therefore ascertaining the true effect of SmartRub® on HH compliance. Another option would be programming SmartRub® to quantify the frequency of HH actions, which would then be used as a "proxy" for HH compliance. An increase in the frequency of HH actions while wearing the SmartRub® with feedback would suggest a possible increment in the compliance with the "5 moments".

SmartRub® could have a role as a "training tool", to help HCWs improving the quality of their HH action. However, the optimal duration of this training needs to be determined. In this context, it would be necessary to investigate the existence of a "carry over" effect on HH quality, persisting even after HCWs stopped wearing SmartRub®.

Several novel applications of SmartRub® deserve to be further tested in clinical trials. Indeed, a new software module can be added to the wristband to monitor the sequence of gestures performed by HCWs in a HH action. This would allow for the evaluation of the technique of HH. In addition, it is now possible to provide the HCWs with a regular aggregated feedback on their individual performance with regards to the HH action. This feedback could be delivered by email (e.g. on a weekly basis) or through a purposely-built mobile phone App.

It would also be interesting to try integrating SmartRub® with other available technologies that use crude movements of HCWs as proxies for the "5 moments" of HH. However, those technologies still need to be further developed as they currently mainly evaluate room "entrance and exit", instead of real HH moments.

In my perspective, the future of hand hygiene lies in the complementation of the classic and well-established WHO multimodal strategy by the introduction of new technologies aiding in behaviour modification. New technologies can be useful to monitor and provide feedback on HH but are also most welcomed to boost other components of the WHO's multimodal strategy, such as training and education. However, the use of these new technologies does also raise problems of confidentiality, data protection and workplace surveillance, which need to be carefully considered, in order to define appropriate limits and safeguards.

The foundation of IPC lies in a relation of trust between IPC teams and frontline HCWs, overseen by a supportive leadership. The confidence that all parts do their best to provide effective and safe care for both patients and providers is a key element of this collaboration. This is why we keep working to improve hand hygiene. Outstanding care for

patients and the safety of HCWs are at the centre of IPC. These goals should keep fuelling research to improve every aspect of IPC, because *Clean Care is Safer Care*.

Bibliography

1. Suetens C, Latour K, Kärki T, et al. Prevalence of healthcare-associated infections, estimated incidence and composite antimicrobial resistance index in acute care hospitals and long-term care facilities: results from two European point prevalence surveys, 2016 to 2017. Eurosurveillance **2018**; 23.

2. Cassini A, Plachouras D, Eckmanns T, et al. Burden of Six Healthcare-Associated Infections on European Population Health: Estimating Incidence-Based Disability-Adjusted Life Years through a Population Prevalence-Based Modelling Study. PLoS Med **2016**; 13:e1002150.

3. Allegranzi B, Nejad SB, Combescure C, et al. Burden of endemic health-care-associated infection in developing countries: systematic review and meta-analysis. The Lancet **2011**; 377:228–241.

4. Jim O'Neill. Tackling Drug-Resistant Infections Globally: final report and recommendations. 2016.

5. Teillant A, Gandra S, Barter D, Morgan DJ, Laxminarayan R. Potential burden of antibiotic resistance on surgery and cancer chemotherapy antibiotic prophylaxis in the USA: a literature review and modelling study. Lancet Infect Dis **2015**; 15:1429–1437.

6. Pires D, de Kraker MEA, Tartari E, Abbas M, Pittet D. 'Fight Antibiotic Resistance—It's in Your Hands': Call From the World Health Organization for 5th May 2017. Clin Infect Dis **2017**; 64:1780–1783.

7. Tacconelli E, Carrara E, Savoldi A, et al. Discovery, research, and development of new antibiotics: the WHO priority list of antibiotic-resistant bacteria and tuberculosis. Lancet Infect Dis **2018**; 18:318–327.

8. World Health Organization. Antimicrobial resistance: global report on surveillance. 2014.

9. World Health Organization. Global action plan on antimicrobial resistance. 2015. Available at: http://apps.who.int/iris/handle/10665/193736. Accessed 30 January 2017.

10. Draft political declaration of the high-level meeting of the General Assembly on antimicrobial resistance. 2016. Available at: https://www.un.org/pga/71/wp-content/uploads/sites/40/2016/09/DGACM_GAEAD_ESCAB-AMR-Draft-Political-Declaration-1616108E.pdf. Accessed 30 January 2017.

11. World Health Organization. Guidelines on core components of infection prevention and control programmes at the national and acute health care facility level. 2016. Available at: http://apps.who.int/iris/handle/10665/251730. Accessed 17 December 2016.

12. Umscheid CA, Mitchell MD, Doshi JA, Agarwal R, Williams K, Brennan PJ. Estimating the proportion of healthcare-associated infections that are reasonably preventable and the related mortality and costs. Infect Control Hosp Epidemiol **2011**; 32:101–114.

13. Zingg W, Holmes A, Dettenkofer M, et al. Hospital organisation, management, and structure for prevention of health-care-associated infection: a systematic review and expert consensus. Lancet Infect Dis **2015**; 15:212–224.

14. Tomczyk S, Zanichelli V, Grayson ML, et al. Control of Carbapenem-resistant Enterobacteriaceae, Acinetobacter baumannii, and Pseudomonas aeruginosa in Healthcare Facilities: A Systematic Review and Reanalysis of Quasi-experimental Studies. Clin Infect Dis Off Publ Infect Dis Soc Am **2018**;

15. World Health Organization. Guidelines for the prevention and control of carbapenem-resistant Enterobacteriaceae, Acinetobacter baumannii, and Pseudomonas aeruginosa in health care facilities. 2017. Available at: http://www.ncbi.nlm.nih.gov/books/NBK493061/. Accessed 10 December 2018.

16. Gammon J, Morgan-Samuel H, Gould D. A review of the evidence for suboptimal compliance of healthcare practitioners to standard/universal infection control precautions. J Clin Nurs **2008**; 17:157–167.

17. Otter JA, Mutters NT, Tacconelli E, Gikas A, Holmes AH. Controversies in guidelines for the control of multidrug-resistant Gram-negative bacteria in EU countries. Clin Microbiol Infect Off Publ Eur Soc Clin Microbiol Infect Dis **2015**; 21:1057–1066.

18. European Centre for Disease Prevention and Control. Point prevalence survey of healthcare-associated infections and antimicrobial use in European acute care hospitals: 2011-2012. 2013.

19. European Centre for Disease Prevention and Control. Surveillance of antimicrobial resistance in Europe – Annual report of the European Antimicrobial Resistance Surveillance Network (EARS-Net) 2017. 2018.

20. Pires D, Zagalo A, Santos C, et al. Evolving epidemiology of carbapenemase-producing Enterobacteriaceae in Portugal: 2012 retrospective cohort at a tertiary hospital in Lisbon. J Hosp Infect **2016**; 92:82–85.

21. Clack L, Zingg W, Saint S, et al. Implementing infection prevention practices across European hospitals: an in-depth qualitative assessment. BMJ Qual Saf **2018**; 27:771–780.

22. Giguère A, Légaré F, Grimshaw J, et al. Printed educational materials: effects on professional practice and healthcare outcomes. Cochrane Database Syst Rev **2012**; 10:CD004398.

23. Mutters NT, Tacconelli E. Infection prevention and control in Europe – the picture in the mosaic. Clin Microbiol Infect **2015**; 21:1045–1046.

24. Birgand G, Castro-Sánchez E, Hansen S, et al. Comparison of governance approaches for the control of antimicrobial resistance: Analysis of three European countries. Antimicrob Resist Infect Control **2018**; 7. Available at: https://aricjournal.biomedcentral.com/articles/10.1186/s13756-018-0321-5. Accessed 11 December 2018.

25. Hansen S, Zingg W, Ahmad R, et al. Organization of infection control in European hospitals. J Hosp Infect **2015**; 91:338–345.

26. Pires D, Tartari E, Bellissimo-Rodrigues F, Pittet D. Why language matters: a tour through hand hygiene literature. Antimicrob Resist Infect Control **2017**; 6:65.

27. Nuland SB. The doctor's plague: germs, childbed fever, and the strange story of Ignác Semmelweis. New York: WW Norton & Company, 2003.

28. Stewardson A, Pittet D. Ignác Semmelweis-celebrating a flawed pioneer of patient safety. Lancet Lond Engl **2011**; 378:22–23.

29. Mortimer EA, Lipsitz PJ, Wolinsky E, Gonzaga AJ, Rammelkamp CH. Transmission of staphylococci between newborns. Importance of the hands to personnel. Am J Dis Child 1960 **1962**; 104:289–295.

30. Garner JS, Favero MS. CDC guideline for handwashing and hospital environmental control, 1985. Infect Control **1986**; :231–243.

31. Larson EL. APIC guideline for handwashing and hand antisepsis in health care settings. Am J Infect Control **1995**; 23:251–269.

32. Pittet D, Mourouga P, Perneger TV. Compliance with handwashing in a teaching hospital. Ann Intern Med **1999**; 130:126–130.

33. Pittet D, Hugonnet S, Harbarth S, et al. Effectiveness of a hospital-wide programme to improve compliance with hand hygiene. The Lancet **2000**; 356:1307–1312.

34. Allegranzi B, Gayet-Ageron A, Damani N, et al. Global implementation of WHO's multimodal strategy for improvement of hand hygiene: a quasi-experimental study. Lancet Infect Dis **2013**; 13:843–851.

35. World Health Organization. Guidelines on hand hygiene in health care. First global patient safety challenge: Clean Care is Safer Care. 2009.

36. World Health Organization. A Guide to the Implementation of the World Health Organization Multimodal Hand Hygiene Improvement Strategy. 2009.

37. Boyce JM, Pittet D. Guideline for hand hygiene in health-care settings: recommendations of the Healthcare Infection Control Practices Advisory Committee and the HICPAC/SHEA/APIC/IDSA Hand Hygiene Task Force. Am J Infect Control **2002**; 30:S1–S46.

38. Allegranzi B, Storr J, Dziekan G, Leotsakos A, Donaldson L, Pittet D. The First Global Patient Safety Challenge 'Clean Care is Safer Care': from launch to current progress and achievements. J Hosp Infect **2007**; 65 Suppl 2:115–123.

39. Kilpatrick C. Save lives: clean your hands. A global call for action at the point of care. Am J Infect Control **2009**; 37:261–262.

40. Saito H, Allegranzi B, Pittet D. 2018 WHO hand hygiene campaign: preventing sepsis in health care and the path to universal health coverage. Lancet Infect Dis **2018**; 18:490–492.

41. Peters A, Borzykowski T, Tartari E, et al. 'Clean care for all - It's in your hands': The May 5th, 2019 World Health Organization SAVE LIVES: Clean Your Hands campaign. Int J Infect Dis IJID Off Publ Int Soc Infect Dis **2019**; 82:135–136.

42. Peters A, Lotfinejad N, Guitart C, Simniceanu A, Allegranzi B, Pittet D. 'Nurses and midwives: clean care is in your hands': the 5th May 2020 World Health Organization SAVE LIVES: Clean Your Hands campaign. Clin Microbiol Infect Off Publ Eur Soc Clin Microbiol Infect Dis **2020**;

43. World Health Organization. Guide to local production: WHO-recommended Handrub Formulations. 2010.

44. Ahmed QA, Memish ZA, Allegranzi B, Pittet D, WHO Global Patient Safety Challenge. Muslim health-care workers and alcohol-based handrubs. Lancet Lond Engl **2006**; 367:1025–1027.

45. Longtin Y, Sax H, Allegranzi B, Schneider F, Pittet D. Hand hygiene. N Engl J Med **2011**; 364:e24.

46. Campion EW, Scott L, Graham A, Prince JM, Morrissey S, Drazen JM. NEJM.org - 20 Years on the Web. N Engl J Med **2016**; 375:993–994.

47. Luangasanatip N, Hongsuwan M, Limmathurotsakul D, et al. Comparative efficacy of interventions to promote hand hygiene in hospital: systematic review and network meta-analysis. BMJ **2015**; 351:h3728.

48. Johnson PDR, Martin R, Burrell LJ, et al. Efficacy of an alcohol/chlorhexidine hand hygiene program in a hospital with high rates of nosocomial methicillin-resistant Staphylococcus aureus (MRSA) infection. Med J Aust **2005**; 183:509–514.

49. Vernaz N, Sax H, Pittet D, Bonnabry P, Schrenzel J, Harbarth S. Temporal effects of antibiotic use and hand rub consumption on the incidence of MRSA and Clostridium difficile. J Antimicrob Chemother **2008**; 62:601–607.

50. Lee Y-T, Chen S-C, Lee M-C, et al. Time-series analysis of the relationship of antimicrobial use and hand hygiene promotion with the incidence of healthcare-associated infections. J Antibiot (Tokyo) **2012**; 65:311–316.

51. Pelat C, Kardaś-Słoma L, Birgand G, et al. Hand Hygiene, Cohorting, or Antibiotic Restriction to Control Outbreaks of Multidrug-Resistant Enterobacteriaceae. Infect Control Hosp Epidemiol **2016**; 37:272–280.

52. World Health Organization. Evidence of hand hygiene to reduce transmission and infections by multi- drug resistant organisms in health-care settin. 2014; Available at: (http://www.who.int/gpsc/5may/MDRO_literature-review.pdf).

53. Pittet D, Allegranzi B, Sax H, et al. Evidence-based model for hand transmission during patient care and the role of improved practices. Lancet Infect Dis **2006**; 6:641–652.

54. Sax H, Allegranzi B, Uçkay I, Larson E, Boyce J, Pittet D. 'My five moments for hand hygiene': a user-centred design approach to understand, train, monitor and report hand hygiene. J Hosp Infect **2007**; 67:9–21.

55. Stewardson AJ, Iten A, Camus V, et al. Efficacy of a New Educational Tool to Improve Handrubbing Technique amongst Healthcare Workers: A Controlled, Before-After Study. PLoS ONE **2014**; 9:e105866.

56. Tartari E, Fankhauser C, Peters A, et al. Scenario-based simulation training for the WHO hand hygiene self-assessment framework. Antimicrob Resist Infect Control **2019**; 8:58.

57. Kilpatrick C, Tartari E, Gayet-Ageron A, et al. Global hand hygiene improvement progress: two surveys using the WHO Hand Hygiene Self-Assessment Framework. J Hosp Infect **2018**; 100:202–206.

58. Pires D, Pittet D. Hand hygiene electronic monitoring: are we there yet? Am J Infect Control 2017;45(5):464-465.

59. World Health Organization. Hand hygiene technical reference manual: to be used by health-care workers, trainers and observers of hand hygiene practices. 2009.

60. Jeanes A, Coen PG, Gould DJ, Drey NS. Validity of hand hygiene compliance measurement by observation: A systematic review. Am J Infect Control **2019**; 47:313–322.

61. Boyce JM. Electronic monitoring in combination with direct observation as a means to significantly improve hand hygiene compliance. Am J Infect Control **2017**; 45:528–535.

62. McCambridge J, Witton J, Elbourne DR. Systematic review of the Hawthorne effect: new concepts are needed to study research participation effects. J Clin Epidemiol **2014**; 67:267–277.

63. Chen LF, Vander Weg MW, Hofmann DA, Reisinger HS. The Hawthorne Effect in Infection Prevention and Epidemiology. Infect Control Hosp Epidemiol **2015**; 36:1444–1450.

64. Zingg W, Haidegger T, Pittet D. Hand coverage by alcohol-based handrub varies: Volume and hand size matter. Am J Infect Control **2016**; 44:1689–1691.

65. Stewardson AJ, Iten A, Camus V, et al. Efficacy of a new educational tool to improve Handrubbing technique amongst healthcare workers: a controlled, before-after study. PloS One **2014**; 9:e105866.

66. Erasmus V, Daha TJ, Brug H, et al. Systematic review of studies on compliance with hand hygiene guidelines in hospital care. Infect Control Hosp Epidemiol **2010**; 31:283–294.

67. Tartari E, Pires D, Bellissimo-Rodrigues F, et al. The global hand-sanitizing relay: promoting hand hygiene through innovation. J Hosp Infect **2017**; 95:189–193.

68. Seto WH, Cowling BJ, Cheung CWY, et al. Impact of the first hand sanitizing relay world record on compliance with hand hygiene in a hospital. Am J Infect Control **2015**; 43:295–297.

69. Tartari E, Fankhauser C, Masson-Roy S, et al. Train-the-Trainers in hand hygiene: a standardized approach to guide education in infection prevention and control. Antimicrob Resist Infect Control **2019**; 8:206.

70. Seto WH, Li K-H, Cheung CWY, Ching PTY, Cowling BJ. Breaking a Guinness World Record on Hand Sanitizing Relay, initiating a call for vital research in overcoming campaign fatigue for hand hygiene. F1000Research **2014**; 3:234.

71. Pittet D. Commentary: hand hygiene: it's all about when and how. Infect Control Hosp Epidemiol **2008**; 29:957–959.

72. Widmer AF, Conzelmann M, Tomic M, Frei R, Stranden AM. Introducing alcohol-based hand rub for hand hygiene: the critical need for training. Infect Control Hosp Epidemiol **2007**; 28:50–54.

73. Laustsen S, Lund E, Bibby BM, Kristensen B, Thulstrup AM, Mller JK. Effect of correctly using alcohol-based hand rub in a clinical setting. Infect Control **2008**; 29:954–956.

74. Bellissimo-Rodrigues F, Pires D, Soule H, Gayet-Ageron A, Pittet D. Assessing the Likelihood of Hand-to-Hand Cross-Transmission of Bacteria: An Experimental Study. Infect Control Hosp Epidemiol **2017**; 38:553–558.

75. Ayliffe G, Babb J, Quoraishi A. A test for 'hygienic' hand disinfection. J CLIN PATHOL **1978**; 31:923–928.

76. European Committee for Standardization. European Norm 1500: chemical disinfectants and antiseptics. Hygienic handrub. Test method and requirements (phase 2/step 2). Version 2013.

77. Tschudin-Sutter S, Sepulcri D, Dangel M, Schuhmacher H, Widmer AF. Compliance with the World Health Organization Hand Hygiene Technique: A Prospective Observational Study. Infect Control Hosp Epidemiol **2015**; 36:482–483.

78. Leslie L, Donskey C, Zabarsky T, Parker A, Macinga D, Assadian O. Measuring alcohol-based hand rub volume used by healthcare workers in practice. Antimicrob Resist Infect Control **2015**;

79. Dalziel C, McIntyre J, Chand AG, McWilliam S, Ritchie L. Validation of a national hand hygiene proxy measure in NHS Scotland. J Hosp Infect **2018**; 98:375–377.

80. Sickbert-Bennett EE, Weber DJ, Gergen-Teague MF, Sobsey MD, Samsa GP, Rutala WA. Comparative efficacy of hand hygiene agents in the reduction of bacteria and viruses. Am J Infect Control **2005**; 33:67–77.

81. Bellissimo-Rodrigues F, Soule H, Gayet-Ageron A, Martin Y, Pittet D. Should Alcohol-Based Handrub Use Be Customized to Healthcare Workers' Hand Size? Infect Control Hosp Epidemiol **2016**; 37:219–221.

82. Price L, Melone L, McLarnon N, et al. A systematic review to evaluate the evidence base for the World Health Organization's adopted hand hygiene technique for reducing the microbial load on the hands of healthcare workers. Am J Infect Control **2018**; Available at: http://linkinghub.elsevier.com/retrieve/pii/S0196655318300658. Accessed 31 March 2018.

83. Pires D, Bellissimo-Rodrigues F, Soule H, Gayet-Ageron A, Pittet D. Revisiting the WHO 'How to Handrub' Hand Hygiene Technique: Fingertips First? Infect Control Hosp Epidemiol **2017**; 38:230–233.

84. Pires D, Soule H, Bellissimo-Rodrigues F, Gayet-Ageron A, Pittet D. Hand Hygiene With Alcohol-Based Hand Rub: How Long Is Long Enough? Infect Control Hosp Epidemiol **2017**; 38:547–552.

85. Pires D, Soule H, Bellissimo-Rodrigues F, de Kraker MEA, Pittet D. Antibacterial efficacy of handrubbing for 15 versus 30 seconds: EN 1500-based randomized experimental study with different loads of Staphylococcus aureus and Escherichia coli. Clin Microbiol Infect **2019**; 25:851–856.

86. Rotter M, Sattar S, Dharan S, Allegranzi B, Mathai E, Pittet D. Methods to evaluate the microbicidal activities of hand-rub and hand-wash agents. J Hosp Infect **2009**; 73:191–199.

87. American Society for Testing and Materials (ASTM) International. Standard test method for evaluation of the effectiveness of health care personnel or consumer handwash formulations. 1999 (designation: E 1174).

88. Hsu Y-W, Yu C-Y. Hand Surface Area Estimation Formula Using 3D Anthropometry. J Occup Environ Hyg **2010**; 7:633–639.

89. Brankston G, Gitterman L, Hirji Z, Lemieux C, Gardam M. Transmission of influenza A in human beings. Lancet Infect Dis **2007**; 7:257–265.

90. Bridges CB, Kuehnert MJ, Hall CB. Transmission of influenza: implications for control in health care settings. Clin Infect Dis Off Publ Infect Dis Soc Am **2003**; 37:1094–1101.

91. Dharan S, Hugonnet S, Sax H, Pittet D. Comparison of Waterless Hand Antisepsis Agents at Short Application Times: Raising the Flag of Concern •. Infect Control Hosp Epidemiol **2003**; 24:160–164.

92. Longtin Y, Schneider A, Tschopp C, et al. Contamination of Stethoscopes and Physicians' Hands After a Physical Examination. Mayo Clin Proc **2014**; 89:291–299.

93. Reilly JS, Price L, Lang S, et al. A Pragmatic Randomized Controlled Trial of 6-Step vs 3-Step Hand Hygiene Technique in Acute Hospital Care in the United Kingdom. Infect Control Hosp Epidemiol **2016**; :1–6.

94. Tschudin-Sutter S, Rotter ML, Frei R, et al. Simplifying the WHO 'how to hand rub' technique: three steps are as effective as six-results from an experimental randomized crossover trial. Clin Microbiol Infect Off Publ Eur Soc Clin Microbiol Infect Dis **2017**; 23:409.e1-409.e4.

95. Gayet-Ageron A, Bellissimo-Rodrigues F, Soule H, Martin Y, Pittet D. Relationship between hand size, volume of alcohol-based handrub and time needed to dry hands. An experimental laboratory-based study. Antimicrob Resist Infect Control **2015**; 4:P303.

96. Kampf G, Marschall S, Eggerstedt S, Ostermeyer C. Efficacy of ethanol-based hand foams using clinically relevant amounts: a cross-over controlled study among healthy volunteers. BMC Infect Dis **2010**; 10:1.

97. Macinga DR, Shumaker DJ, Werner H-P, et al. The relative influences of product volume, delivery format and alcohol concentration on dry-time and efficacy of alcohol-based hand rubs. BMC Infect Dis **2014**; 14:1.

98. Chow A, Arah OA, Chan S-P, et al. Alcohol handrubbing and chlorhexidine handwashing protocols for routine hospital practice: A randomized clinical trial of protocol efficacy and time effectiveness. Am J Infect Control **2012**; 40:800–805.

99. Kramer A, Pittet D, Klasinc R, et al. Shortening the Application Time of Alcohol-Based Hand Rubs to 15 Seconds May Improve the Frequency of Hand Antisepsis Actions in a Neonatal Intensive Care Unit. Infect Control Hosp Epidemiol **2017**; :1–5.

100. Scheithauer S, Bickenbach J, Heisel H, Fehling P, Marx G, Lemmen S. Do WiFi-based hand hygiene dispenser systems increase hand hygiene compliance? Am J Infect Control **2018**; Available at: https://linkinghub.elsevier.com/retrieve/pii/S0196655318302578. Accessed 17 June 2018.

101. Kampf G. The Puzzle of Volume, Coverage, and Application Time in Hand Disinfection. Infect Control Hosp Epidemiol **2017**; :1–2.

102. Kampf G, Meyer B, Goroncy-Bermes P. Comparison of two test methods for the determination of sufficient antimicrobial activity of three commonly used alcohol-based hand rubs for hygienic hand disinfection. J Hosp Infect **2003**; 55:220–225.

103. Kampf G, Hollingsworth A. Comprehensive bactericidal activity of an ethanol-based hand gel in 15 seconds. Ann Clin Microbiol Antimicrob **2008**; 7:2.

104. Goroncy-Bermes P. Hand disinfection according to the European Standard EN 1500 (hygienic handrub): a study with Gram-negative and Gram-positive test organisms. Int J Hyg Environ Health **2001**; 204:123–126.

105. Macinga DR, Beausoleil CM, Campbell E, et al. Quest for a Realistic *In Vivo* Test Method for Antimicrobial Hand-Rub Agents: Introduction of a Low-Volume Hand Contamination Procedure. Appl Environ Microbiol **2011**; 77:8588–8594.

106. Sickbert-Bennett EE, Weber DJ, Gergen-Teague MF, Rutala WA. The effects of test variables on the efficacy of hand hygiene agents. Am J Infect Control **2004**; 32:69–83.

107. Kramer A, Pittet D, Klasinc R, et al. Shortening the Application Time of Alcohol-Based Hand Rubs to 15 Seconds May Improve the Frequency of Hand Antisepsis Actions in a Neonatal Intensive Care Unit. Infect Control Hosp Epidemiol **2017**; 38:1430–1434.

108. Harnoss JC, Dancer SJ, Kaden CF, et al. Hand antisepsis without decreasing efficacy by shortening the rub-in time of alcohol-based handrubs to 15 seconds. J Hosp Infect **2019**;

109. Tschudin-Sutter S, Sepulcri D, Dangel M, Ulrich A, Frei R, Widmer AF. Simplifying the World Health Organization Protocol: 3 Steps Versus 6 Steps for Performance of Hand Hygiene in a Cluster-randomized Trial. Clin Infect Dis Off Publ Infect Dis Soc Am **2019**; 69:614–620.

110. World Health Organization. A Guide to the Implementation of the World Health Organization Multimodal Hand Hygiene Improvement Strategy. 2009.

111. Abstracts from the 5th International Conference on Prevention & Infection Control (ICPIC 2019). Antimicrob Resist Infect Control **2019**; 8. Available at: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6737605/. Accessed 19 December 2019.

112. Allegranzi B, Sax H, Bengaly L, et al. Successful implementation of the World Health Organization hand hygiene improvement strategy in a referral hospital in Mali, Africa. Infect Control Hosp Epidemiol **2010**; 31:133–141.

113. Hemming K, Taljaard M. Sample size calculations for stepped wedge and cluster randomised trials: a unified approach. J Clin Epidemiol **2016**; 69:137–146.

114. Baio G, Copas A, Ambler G, Hargreaves J, Beard E, Omar RZ. Sample size calculation for a stepped wedge trial. Trials **2015**; 16:354.

115. Conway LJ. Challenges in implementing electronic hand hygiene monitoring systems. Am J Infect Control **2016**; 44:e7–e12.

116. Meng M, Sorber M, Herzog A, Igel C, Kugler C. Technological innovations in infection control: A rapid review of the acceptance of behavior monitoring systems and their contribution to the improvement of hand hygiene. Am J Infect Control **2019**; 47:439–447.

117. Ward MA, Schweizer ML, Polgreen PM, Gupta K, Reisinger HS, Perencevich EN. Automated and electronically assisted hand hygiene monitoring systems: a systematic review. Am J Infect Control **2014**; 42:472–478.

118. Srigley JA, Gardam M, Fernie G, Lightfoot D, Lebovic G, Muller MP. Hand hygiene monitoring technology: a systematic review of efficacy. J Hosp Infect **2015**; 89:51–60.

119. Ehrenkranz N, Alfonso B. Failure of bland soap handwash to prevent hand transfer of patient bacteria to urethral catheters. Infect Control Hosp Epidemiol **1991**; 12:654–62.

120. Mackintosh CA, Hoffman PN. An extended model for transfer of micro-organisms via the hands: differences between organisms and the effect of alcohol disinfection. J Hyg (Lond) **1984**; 92:345–355.

121. Marples RR, Towers AG. A laboratory model for the investigation of contact transfer of micro-organisms. J Hyg (Lond) **1979**; 82:237–248.

122. Sattar SA, Springthorpe S, Mani S, et al. Transfer of bacteria from fabrics to hands and other fabrics: development and application of a quantitative method using Staphylococcus aureus as a model. J Appl Microbiol **2001**; 90:962–970.

123. Patrick DR, Findon G, Miller TE. Residual moisture determines the level of touch-contact-associated bacterial transfer following hand washing. Epidemiol Infect **1997**; 119:319–325.

124. Kampf G, Reichel M, Feil Y, Eggerstedt S, Kaulfers P-M. Influence of rub-in technique on required application time and hand coverage in hygienic hand disinfection. BMC Infect Dis **2008**; 8:149.

125. Paula H, Becker R, Assadian O, Heidecke C-D, Kramer A. Wettability of hands during 15-second and 30-second handrub time intervals: A prospective, randomized crossover study. Am J Infect Control **2018**; Available at: https://linkinghub.elsevier.com/retrieve/pii/S0196655318301378. Accessed 11 July 2018.

126. Suchomel M, Leslie RA, Parker AE, Macinga DR. How long is enough? Identification of product dry-time as a primary driver of alcohol-based hand rub efficacy. Antimicrob Resist Infect Control **2018**; 7:65.

127. Larson EL, Eke PI, Wilder MP, Laughon BE. Quantity of soap as a variable in handwashing. Infect Control IC **1987**; 8:371–375.

128. Kampf G, Ruselack S, Eggerstedt S, Nowak N, Bashir M. Less and less-influence of volume on hand coverage and bactericidal efficacy in hand disinfection. BMC Infect Dis **2013**; 13:472.

129. Goroncy-Bermes P, Koburger T, Meyer B. Impact of the amount of hand rub applied in hygienic hand disinfection on the reduction of microbial counts on hands. J Hosp Infect **2010**; 74:212–218.

130. Wilkinson MAC, Ormandy K, Bradley CR, Fraise AP, Hines J. Dose considerations for alcohol-based hand rubs. J Hosp Infect **2017**; 95:175–182.

131. Luangasanatip N, Hongsuwan M, Limmathurotsakul D, et al. Comparative efficacy of interventions to promote hand hygiene in hospital: systematic review and network meta-analysis. BMJ **2015**; 351:h3728.