Original article

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COLONIZATION WITH MULTIDRUG-RESISTANT BACTERIA – ON THE EFFICIENCY OF LOCAL DECOLONIZATION PROCEDURES

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The effectiveness of a disinfectant-based decolonization strategy for multidrug-resistant bacteria like extended spectrum β-lactamase (ESBL)-positive Gram-negative bacteria with or without additional fluoroquinolon and carbapenem resistance as well as vancomycin-resistant enterococci and methicillin-resistant *Staphylococcus aureus* was assessed.

Between 2011 and 2015, 25 patients from Libya, Syria, and the Ukraine with war traumata were treated at the Bundeswehr hospital Hamburg. The patients were heavily colonized and infected with multidrug-resistant bacteria, altogether comprising 371 distinct combinations of pathogens and isolation sites. Local disinfection was assessed for effectiveness regarding successful decolonization of multidrug-resistant bacteria.

Altogether, 170 cases of successful decolonization were observed, comprising 95 (55.8%) such events at sampling sites that were accessible to disinfecting procedures. The remaining 75 (44.2%) decolonization events had to be considered as spontaneous. In contrast, 95 out of 172 (55.2%) colonized isolation sites that were accessible to disinfection procedures were successfully decolonized. Patient compliance with the enforced hygiene procedures was associated with decolonization success. Systemic antibiotic therapy did not relevantly affect isolation time.

Disinfecting washing moderately supports local decolonization of multidrug-resistant pathogens in comparison with spontaneous decolonization rates if the patients' compliance with the applied hygiene procedures is ensured.

Keywords: decolonization, multidrug-resistant bacteria, ESBL, MRSA, VRE, carbapenem resistance, war injuries

Introduction

International travel to high endemicity settings is associated with the risk of colonization with multidrug-resistant bacteria and with a potential spread of these agents in the home country [1, 2]. This leads to an increasingly homogeneous global distribution of genetic resistance determinants as recently observed for fecal carriage of resistant Enterobacteriaceae [3].

Influx of resistant pathogens in the course of international migration of refugees can contribute to this phenomenon. As impressively demonstrated by the colleagues in Frankfurt/Main, Germany [4, 5], considerably increased detection rates of colonization with multidrug-resistant bacteria were observed in refugees that were admitted to their hospital. Similar observations were made by Israeli physicians treating Syrian civilians in Israeli hospitals [6]. Also, in Germany, medical tourism from international

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countries of origin contributes to the influx of resistant bacteria [7].

In German military hospitals (Bundeswehr hospitals), similar experience was made with war-traumatized patients from international war and crisis zones. The patients were brought to Germany for the treatment of injuries that were too complex for local management in the crisis settings [8].

Colonization with up to six different multidrug-resistant bacteria was reported but showed excellent response to local disinfection-based decolonization approaches without the use of systemic antibiotic drugs. In a small cohort of four severely colonized patients from Libya, a 100% local decolonization success was described just by hygiene and disinfection procedures at the Bundeswehr Hospital Hamburg [8].

During later treatment approaches of injured patients from war zones in Syria and the Ukraine, however, such excellent response rates have never again been observed. Therefore, this study was conducted to assess factors that are associated with local decolonization success in case of colonization with multidrug-resistant bacteria.

Materials and methods

Patients

Patients from Libya, Syria, or the Ukraine with war injuries that were treated between October 2011 and September 2015 at the Bundeswehr hospital Hamburg with proven colonization or infection with multidrug-resistant bacteria were included in the study. Multidrug-resistant bacteria were defined as methicillin-resistant *Staphylococcus aureus* (MRSA), vancomycin-resistant enterococci (VRE) as well as Enterobacteriaceae and non-fermentative Gram-negative rod-shaped bacteria with resistance against penicillins and cephalosporins (later referred to as 2MRGN – multidrug-resistant Gram-negative bacteria with resistance against two bactericidal antibiotic groups), mostly of the extended spectrum β-lactamase (ESBL)- or the ampC-type, against penicillins, cephalosporins, and fluorquinolones (later referred to as 3MRGN – multidrug-resistant Gram-negative bacteria with resistance against three bactericidal antibiotic groups) and against penicillins, cephalosporins, fluorquinolones, and carbapenems (later referred to as 4MRGN – multidrug-resistant Gram-negative bacteria with resistance against four bactericidal antibiotic groups). There were no exclusion criteria. Data on the patients were retrospectively assessed in an anonymous way.

Anonymously assessed patient-related data

The assessed patient-related data comprised country of origin, age at the time of admission, sex, main diagnoses, and outcome as well as application or non-application of a systemic antibiotic therapy. Furthermore, the rooms and wards where the patients were treated were recorded to get hints for potential nosocomial transmission.

Case definition

A case was defined as a unique combination of an isolation site (pharynx, nostril, etc.) and a certain colonizing or infecting pathogen (MRSA, VRE, etc.) of a patient. Several pieces of information per case were assessed, comprising isolation site, bacterial species, resistance type, and clinical relevance, i.e., presumed colonization or infection, number of days between hospital admission and first isolation of the strain, decolonization success, number of days between first isolation and decolonization in case of successful decolonization, and recolonization events of the same localizations after initial decolonization success as well as performed or not-performed confirmation of decolonization by at least three different swabbing attempts at the decolonized site. Decolonization was defined as any negative swabbing result without later documented reoccurring of the strain at the respective swabbing site. If, in contrast, the resistant strain was still detectable during the last assessable screening of the isolation site, the decolonization was considered as not successful. Recolonization was defined as later recurrence of the same pathogen at the same location after intermittent negative screening results at the respective observed assessment site.

Hygiene procedures

As previously detailed [8], various procedures were enforced to prevent nosocomial spread of resistant bacteria from the patients and to achieve local decolonization. Infrastructural measures comprised the use of sufficiently dimensioned wards as isolation area for cohort isolation. Personnel for the management of the multiply colonized patients were identified and specifically trained regarding the enforced advanced hygiene procedures. Additional personnel were recruited from nearby other military departments to ensure sufficient manpower. Personal protective equipment for the personnel and antiseptic wound dressing were ordered in sufficient quantities via the hospital's pharmacy. Furthermore, sufficient supply of clothing, bed linen, and everyday wear for the patients was ensured. Food and beverages were provided using one-way dishes.

Surface disinfection on the wards was performed using the disinfectant Terralin PAA (Schülke & Mayr Ltd., Norderstedt, Germany). Furthermore, a special sewage sink was purchased which could be used with one-way products made of cellulose to remove excretions of immobilized patients without the risk of contamination and pathogen spread by urine bottles and bedpans.

The patients were treated in cohort isolation on room level. Each patient room was equipped with an own bathroom. The nursing staff had to change the personal protection equipment if another patient in the same room was

cared for. Weekly hygiene screening for multidrug-resistant bacteria by swabbing and cultural assessment at the Central Institute of the Bundeswehr Medical Service Kiel, Department Berlin, was performed from the nostrils, the throat, the groins, and the perianal region as well as from wounds of all patients.

Patient-related hygiene procedures had to be performed either by the patients themselves or by the nursing staff in charge, usually supported by an interpreter. Daily washing with disinfecting liquids based on octenidin for the intact skin and octenidol for mucous membranes like in the oral cavity and for superficial wounds (Schülke & Mayr Ltd.) was performed in line with a daily showering and washing plan. The nose and the pharynx were disinfected even three times a day. After the washing procedure, the complete cloths, towels, and bed linen were exchanged. The patients' property and patient-specifically applied medical equipment were disinfected several times a day.

Based on these hygiene procedures, localizations that were accessible to local disinfection procedures were defined. They comprised the nostrils, the oral cavity, the skin, and superficial wounds. All other sites of detection were considered as inaccessible to such procedures.

Assessment of patient compliance with the enforced hygiene procedures

As patient compliance had early been suspected to interfere with decolonization efficiency [8], compliance of the patients was assessed by the hygiene nurses in charge based on a visual analogous scale ranging from 1 to 10. While ten described excellent patient compliance, 1 described the poorest possible way of adherence to the hygiene schemes.

Statistics

The collected and analyzed data were mainly descriptively assessed. Non-parametric Mann–Whitney *U*-testing was used to assess effects of patient compliance on decolonization success and to calculate two-tailed *P*-values. Effects of applied or not applied systemic antibiotic therapy on the time of detection of multidrug-resistant pathogens during screening assessments were calculated using Fischer's two-sided exact test. The applied statistics software was GraphPad Instat®, version 3.06 (GraphPad Software Inc., San Diego, USA).

Ethics

Ethical clearance was obtained from the ethics committee of the medical association of Hamburg (WF-053/15).

Results

Patients

Altogether, 25 patients could be included in the assessment, comprising six injured from Libya, nine from Syria, and ten from the Ukraine. The patients were male without exemption. All patients suffered from relevant war-related traumata. The mean age was under 30 years with the Libyan patients being slightly older. While antibiotic therapy could be completely avoided in the Libyan and Syrian patients, 70% of the Ukrainians had to be treated with antibiotics due to systemic infection signs. While most of the patients were routinely dismissed after completing their therapy, individual patients vanished to avoid the retransfer back to their home country or had to be dismissed due to inappropriate behavior towards the hospital staff *(Table 1)*. Accordingly, the assessment period was shortened in the respective way for these individuals.

Cases

Altogether, 371 cases were defined *(Table 2)*. They comprised colonization and infections with 15 MRSA, 2 VRE,

Table 1. Characterization of the study population

Country	Number (*n*)	Gender	Age (years) ± SD	Main diagnoses	Percentage of patients with systemic antibiotic therapy (%)	Observed clinical outcomes (%)
Libya	6	Male	32.2 ± 3.8	Gun shot injury (*n* = 3) Limb amputation (*n* = 1) Splinter injury (*n* = 1) Wound infection (*n* = 1)	0	100 dismissal
Syria	9	Male	25.1 ± 4.9	Gun shot injury (*n* = 6) Blast injury (*n* = 3)	0	77.7 dismissal, 22.2 other*1
Ukraine	10	Male	27.7 ± 7.8	Gun shot injury (*n* = 5) Splinter injury (*n* = 4) Blast injury (*n* = 1)	70	70.0 dismissal, 10.0 transfer, 20.0 other*2
Total	25	Male	27.8 ± 6.4	---------- ----------	28	80.0 dismissal, 4.0 transfer, 16.0 other*3

Explanation of observed clinical outcomes: *1: *n* = 1 got transferred to another hospital and escaped before dismissal, *n* = 1 disappeared somewhere in Germany without an official dismissal; *2: *n* = 1 was sent back home because of aggressive behavior towards nursing staff, *n* = 1 was still hospitalized at the time-point of data acquisition; *3: referring to *1 and *2 (*n* = 4)

Table 2. Characterization of isolation events of multidrug resistant (MDR) bacteria

ESBL/2MRGN (*n* = 47)					
Species	Isolation site(s)	Percentage of presumed infections	Days in hospital until first isolation (mean ± SD)	Percentage of successful decolonization	Percentage of successful decolonization proven by 3 negative swabs
Klebsiella pneumoniae (*n* = 22)	Inguinal (*n* = 9) Perianal (*n* = 5) Nose (*n* = 3) Rectal (*n* = 2) Pharynx (*n* = 1) Gluteal (*n* = 1) Wound (*n* = 1)	4.6	21.2 ± 17.3	68.2	18.2
Escherichia coli (*n* = 19)	Perianal (*n* = 9) Abdomen (*n* = 2) Urine (*n* = 2) Wound (*n* = 1) Inguinal (*n* = 1) Gluteal (*n* = 1) Rectal (*n* = 1) Artificial anus (*n* = 1) Suprapubic catheter (*n* = 1)	47.4	10.7 ± 22.4	63.2	5.3
Enterobacter cloacae (*n* = 4)	Perianal (*n* = 1) Wound (*n* = 1) Elbow (*n* = 1) Heel (*n* = 1)	75.0	15.5 ± 10.4	75.0	0
Citrobacter freundii (*n* = 2)	Perianal (*n* = 1) Inguinal (*n* = 1)	0	4.0 ± 4.2	0	0
3MRGN (*n* = 168)					
Species	Isolation site(s)	Percentage of presumed infections	Days in hospital until first isolation (mean ± SD)	Percentage of successful decolonization	Percentage of successful decolonization proven by 3 negative swabs
Klebsiella pneumoniae (*n* = 69)	Perianal (*n* = 15) Inguinal (*n* = 9) Rectal (*n* = 8) Nose (*n* = 5) Upper leg (*n* = 5) Pharynx (*n* = 4) Armpit (*n* = 3) Urine (*n* = 3) Abdominal deep (*n* = 2) Deep shoulder (*n* = 2) Gluteal (*n* = 1) Artificial anus (*n* = 1) Wound (*n* = 1) Lower leg (*n* = 1) Chest (*n* = 1) Forehead (*n* = 1) External nose (*n* = 1) Abdominal (*n* = 1) Shoulder (*n* = 1) Mesh graft (*n* = 1) Blood culture (*n* = 1) Bowel (*n* = 1) Tracheal secretion (*n* = 1) Central venous catheter (*n* = 1)	36.2	26.1 ± 25.7	46.4	10.1

Table 2. *(cont'd)*

Escherichia coli (*n* = 32)	Perianal (*n* = 7) Urine (*n* = 4) Rectal (*n* = 4) Gluteal (*n* = 3) Inguinal (*n* = 2) Abdomen (*n* = 2) Suprapubic catheter (*n* = 2) Artificial anus (*n* = 2) Pharynx (*n* = 1) Nose (*n* = 1) Forehead (*n* = 1) Fistula (*n* = 1) Tibia (*n* = 1) Lower leg (*n* = 1)	53.1	34.2 ± 39.6	46.9	6.3
Enterobacter cloacae (*n* = 29)	Perianal (*n* = 7) Inguinal (*n* = 3) Rectal (*n* = 3) Nose (*n* = 2) Wound (*n* = 2) Knee (*n* = 2) Upper leg (*n* = 2) Pharynx (*n* = 1) Lower leg (*n* = 1) Tibia (*n* = 1) Gluteal (*n* = 1) Heel deep (*n* = 1) Knee deep (*n* = 1) Elbow deep (*n* = 1) Left eye (*n* = 1)	82.8	18.3 ± 19.6	62.1	0
Morganella morganii (*n* = 9)	Upper leg (*n* = 3), Rectal (*n* = 2) Perianal (*n* = 1) Forehead (*n* = 1) Artificial anus (*n* = 1) Suprapubic catheter (*n* = 1)	55.6	23.0 ± 27.0	77.8	11.1
Pseudomonas aeruginosa (*n* = 6)	Wound (*n* = 3) Perianal (*n* = 1) Superficial shoulder (*n* = 1) Humerus intra-operative (*n* = 1)	66.7	10.8 ± 11.0	83.3	0
Citrobacter freundii (*n* = 6)	Perianal (*n* = 2) Superficial shoulder (*n* = 1) Deep shoulder (*n* = 1) Inguinal (*n* = 1) Humerus intra-operative (*n* = 1)	50.0	13.5 ± 10.9	100	0
Enterobacter aerogenes (*n* = 6)	Shoulder (*n* = 1) Deep shoulder (*n* = 1) Upper leg (*n* = 1) Lower leg (*n* = 1) Perianal (*n* = 1) Rectal (*n* = 1)	33.3	27.3 ± 15.9	83.3	3.4
Proteus mirabilis (*n* = 5)	Inguinal (*n* = 2) Nose (*n* = 1) Rectal (*n* = 1) Perianal (*n* = 1)	0	35.6 ± 20.6	60.0	0
Acinetobacter baumannii (*n* = 2)	Heel deep (*n* = 1) Upper leg (*n* = 1)	50.0	18.5 ± 24.7	50.0	0
Serratia marcescens (*n* = 2)	Rectal (*n* = 1) Artificial anus (*n* = 1)	0	5.0 ± 0	100	0
Proteus vulgaris (*n* = 1)	Inguinal (*n* = 1)	0	10.0 ± (−)	100	0

Table 2. *(cont'd)*

Klebsiella oxytoca (*n* = 1)	Nose (*n* = 1)	0	253.0 ± (−)	100	0
		4MRGN (*n* = 139)			
Species	Isolation site(s)	Percentage of presumed infections	Days in hospital until first isolation (mean ± SD)	Percentage of successful decolonization	Percentage of successful decolonization proven by 3 negative swabs
Acinetobacter baumannii (*n* = 53)	Inguinal (*n* = 7) Perianal (*n* = 5) Upper leg (*n* = 4) Forehead (*n* = 4) Armpit (*n* = 4) Elbow (*n* = 3) Lower leg (*n* = 3) Nose (*n* = 3) Pharynx (*n* = 3) Gluteal (*n* = 3) External fixator (*n* = 2) Rectal (*n* = 2) Wound (*n* = 2) Chest (*n* = 1) Knee (*n* = 1) Tibia (*n* = 1) Forearm (*n* = 1) Heel (*n* = 1) Lower abdomen (*n* = 1) Suprapubic catheter (*n* = 1) Tracheal secretion (*n* = 1)	54.7	8.5 ± 13.0	64.2	3.8
Klebsiella pneumoniae (*n* = 40)	Inguinal (*n* = 7) Perianal (*n* = 6) Upper leg (*n* = 4) Nose (*n* = 3) Pharynx (*n* = 3) Rectal (*n* = 3) Urine (*n* = 2) Blood culture (*n* = 2) Lower leg (*n* = 1) Abdominal (*n* = 1) Abdominal deep (*n* = 1) Tracheal secretion (*n* = 1) Shoulder deep (*n* = 1) Gluteal (*n* = 1) Intra-operative swab (*n* = 1) Tracheostomy (*n* = 1) Epidural catheter (*n* = 1) Artificial anus (*n* = 1)	37.5	23.2 ± 30.7	50.0	10.0
Pseudomonas aeruginosa (*n* = 40)	Upper leg (*n* = 8) Pharynx (*n* = 3) Inguinal (*n* = 3) Perianal (*n* = 3) Urine (*n* = 3) Armpit (*n* = 2) Nose (*n* = 2) Gluteal (*n* = 2) Forearm (*n* = 1) Wrist (*n* = 1) Hip (*n* = 1) Lower leg (*n* = 1)	57.5	14.7 ± 24.9	45.0	10.0

Table 2. *(cont'd)*

Pseudomonas aeruginosa (*n* = 40) *(cont'd)*	Chest (*n* = 1) Forehead (*n* = 1) External nose (*n* = 1) Abdomen (*n* = 1) Heel (*n* = 1) Abdominal deep (*n* = 1) Femoral shaft (*n* = 1) Rectal (*n* = 1) Artificial anus (*n* = 1) Central venous catheter (*n* = 1)	57.5	14.7 ± 24.9	45.0	10.0
Escherichia coli (*n* = 3)	Inguinal (*n* = 1) Perianal (*n* = 1) Urine (*n* = 1)	33.3	9.0 ± 7.0	33.0	0
Enterobacter cloacae (*n* = 2)	Upper leg (*n* = 1) External fixator (*n* = 1)	100	18.5 ± 24.7	0	0
Enterobacter aerogenes (*n* = 1)	Nose (*n* = 1)	0	27.0 ± (−)	100	0
		MRSA (*n* = 15)			
Species	Isolation site(s)	Percentage of presumed infections	Days in hospital until first isolation (mean ± SD)	Percentage of successful decolonization	Percentage of successful decolonization proven by 3 negative swabs
Staphylococcus aureus (*n* = 15)	Nose (*n* = 2) Pharynx (*n* = 2) Knee (*n* = 1) Tibia right (*n* = 1) Lower leg (*n* = 1) Upper leg (*n* = 1) Forearm (*n* = 1) Elbow deep (*n* = 1) Shoulder (*n* = 1) Shoulder deep (*n* = 1) Perianal (*n* = 1) rectal (*n* = 1) intra-operative swab (*n* = 1)	60.0	37 ± 48.9	26.6	13.3
		VRE (*n* = 2)			
Species	Isolation site(s)	Percentage of presumed infections	Days in hospital until first isolation (mean ± SD)	Percentage of successful decolonization	Percentage of successful decolonization proven by 3 negative swabs
Enterococcus faecium (*n* = 2)	Pharynx (*n* = 1) Rectal (*n* = 1)	0	32.5 ± 4.9	0	0

47 2MRGN, 168 3MRGN, and 139 4MRGN. The localization of the pathogens comprised sites both accessible and inaccessible to hygienic decolonization procedures. The distribution of presumed colonization and infection widely varied without any obvious difference between Gram-positive and Gram-negative agents. About all assessed species, the mean time between hospital admission and pathogen detection exceeded 72 h by far, formally fulfilling the criteria of nosocomial transmission. However, partly very big standard deviations of the respective mean values primarily explained this phenomenon. Some isolates were detected very late in the course of medical treatment while others were isolated directly after admission. Focusing on the percentage of successful decolonization events, the decolonization rate of many Gram-negative species was higher than for MRSA. Low percentages of cases, in which decolonization was confirmed by at least three negative swabs, however, weaken the interpretability of this finding.

From the total of 371 cases, successful decolonization was documented for 170 (45.8%) *(Table 3),* comprising both hygiene associated and spontaneous decolonization

Table 3. Characterization of successful decolonization events

Number of successful colonization events per species	Recolonization with the strain after initial successful decolonization (defined as intermitting negative screening results) (%)	Number of days between first isolation and successful decolonization (negative swabbing result without later reoccurring), mean ± standard deviation (SD)
Acinetobacter baumannii (*n* = 34)	5.9	35.5 ± 28.7
Citrobacter freundii (*n* = 6)	0	36.2 ± 23.5
Enterobacter aerogenes (*n* = 6)	0	32.5 ± 20.3
Enterobacter cloacae (*n* = 18)	5.6	41.6 ± 33.9
Escherichia coli (*n* = 20)	10	37.1 ± 41.9
Klebsiella oxytoca (*n* = 1)	0	28.0 ± 0
Klebsiella pneumoniae (*n* = 45)	17.8	48.8 ± 73.8
Morganella morganii (*n* = 7)	14.3	54.5 ± 51.4
Proteus mirabilis (*n* = 3)	0	29.7 ± 13.8
Proteus vulgaris (*n* = 1)	0	13.0 ± 0
Pseudomonas aeruginosa (*n* = 23)	17.4	82.9 ± 100.4
Serratia marcescens (*n* = 2)	0	31.5 ± 4.9
Staphylococcus aureus (*n* = 4)	50.0	39.3 ± 49.2
Total (*n* = 170)	11.8	46.3 ± 59.2

events. The mean time from first detection to documented decolonization was 46.3 days, ranging from 13.0 days for *Proteus vulgaris* to 82.9 days for *Pseudomonas aeruginosa*. The mean recolonization rate prior to final decolonization was 11.8%, with *Klebsiella pneumoniae*, *Morganella morganii*, *P. aeruginosa*, and MRSA being more frequently associated with recolonization after initial decolonization success.

Table 4. Association between decolonization success and patient compliance with decolonization procedures. Only isolates from the skin, pharyngeal mucous membrane, and superficial wounds that were affected by the decolonization procedures were included in the assessment

Species and groups	Compliance (mean ± standard deviation (SD)) in cases of successful decolonization	Compliance (mean ± standard deviation (SD)) in cases without decolonization success	Significance level (*P*)	Percent of successful decolonization (%)
Acinetobacter baumannii	6.4 ± 1.8 (*n* = 22)	4.8 ± 2.6 (*n* = 16)	0.06	57.9 (22/38)
Citrobacter freundii	6.0 ± 1.4 (*n* = 2)	– (*n* = 0)	Not applicable	100 (2/2)
Enterobacter aerogenes	6.3 ± 2.9 (*n* = 3)	– (*n* = 0)	Not applicable	100 (3/3)
Enterobacter cloacae	4.8 ± 1.6 (*n* = 8)	3.7 ± 2.2 (*n* = 6)	0.21	57.1 (8/14)
Enterococcus faecium	– (*n* = 0)	7.0 (*n* = 1)	Not applicable	0 (0/1)
Escherichia coli	6.1 ± 1.6 (*n* = 8)	6.2 ± 1.8 (*n* = 6)	0.95	57.1 (8/14)
Klebsiella oxytoca	8.0 (*n* = 1)	– (*n* = 0)	Not applicable	100 (1/1)
Klebsiella pneumoniae	6.7 ± 1.5 (*n* = 28)	6.0 ± 1.9 (*n* = 21)	0.20	57.1 (28/49)
Morganella morganii	8.0 (*n* = 3)	7.0 (*n* = 1)	Not applicable	75.0 (1/4)
Proteus mirabilis	7.0 (*n* = 2)	7.0 (*n* = 1)	Not applicable	66.7 (2/3)
Proteus vulgaris	7.0 (*n* = 1)	– (*n* = 0)	Not applicable	100 (1/1)
Pseudomonas aeruginosa	7.3 ± 1.5 (*n* = 16)	5.6 ± 2.9 (*n* = 16)	0.18	50 (16/32)
Serratia marcescens	– (*n* = 0)	– (*n* = 0)	Not applicable	Not applicable
Staphylococcus aureus	7.0 (*n* = 1)	4.2 ± 1.9 (*n* = 9)	Not applicable	10 (1/10)
MRGN 2-4	6.6 ± 1.7 (*n* = 94)	5.5 ± 2.4 (*n* = 67)	*P* = 0.007	58.4 (94/161)
Gram-positive bacteria (*S. aureus* and *Enterococcus* spp.)	7.0 (*n* = 1)	4.2 ± 2.0 (*n* = 10)	Not applicable	9.1 (1/11)
Total	6.6 ± 1.7 (*n* = 95)	5.3 ± 2.3 (*n* = 77)	*P* = 0.001	55.2 (95/172)

Of note, the 170 decolonization events *(Table 3)* comprised 95 (55.8%) events *(Table 4)* at sampling sites that were accessible to disinfecting procedures. The remaining 75 out of 170 (44.2%) events *(Table 2)* had to be considered as spontaneous decolonization or response to systemic antibiotic therapy. In total, 172 *(Table 4)* out of the altogether 371 cases (46.4%) comprised locations that were accessible to local disinfection. From these 172 cases, disinfection was successful in 95 events (55.2%) and not successful in 77 events (44.8%) *(Table 4)*.

Effect of patient compliance on decolonization success

Significant effect of patient compliance with local decolonization procedures on the efficiency of decolonization was proven for all 172 cases that were accessible to decolonization and for the cases of colonization with Gram-negative bacteria *(Table 4)*. On the visual analogues scale, slightly better compliance was shown for cases with successful decolonization approaches in these instances. Poor decolonization success was observed for Gram-positive bacteria including MRSA. Any statistical significance got lost in case of assessments on species level with comparably low numbers of accessible cases.

Association of recolonization events with patients in the same room or on the same ward showing identical colonization

As recolonization may occur both due to auto-inoculation from other colonization sites of the same patient or from the patient's environment as well as due to nosocomial transmission events from other patients, e.g., via the hands of the staff, it was assessed whether or not other patients with identical colonizing bacteria were either in the same room or at least on the same ward for all 46 documented recolonization events. Identical colonization of different patients with recolonization events in the same room were just observed for one instance of three cases of colonization or infection with *K. pneumoniae* comprising perianal and pharyngeal colonization as well as blood stream infection in two patients *(Table 5)*. Nosocomial transmission was formally not excluded here. Regarding treatment on the same ward, patients with identical colonization were observed for the most assessed pathogens for which more than one assessed case was defined *(Table 5)*.

Assessment of factors associated with decolonization success

From the 172 cases that were accessible to local disinfection procedures, an assessment of the effects of the factors kind of pathogen, localization site, and systemic antibiotic therapy was performed *(Table 6)*. While the decolonization percentages with regard to the most factors did not relevantly differ from the total decolonization rate of 55.2% (95/172), particularly good decolonization effects were observed for the nostrils with 71.4% success while poor results were observed for Gram-positive pathogens with only 10% decolonization success for MRSA. Focusing on the factor time to decolonization, especially high rates of early decolonization were observed for the phar-

Table 5. Association of recolonization events with cohort isolation with patients carrying the same species

Species	Resistance type	Number of recolonization events (n = 46)	Percentage of cases with a patient colonized with the same pathogen in the same room (%)	Percentage of cases with a patient colonized with the same pathogen on the same ward (%)
Acinetobacter baumannii	4MRGN	2	0	100
Enterobacter cloacae	3MRGN	2	0	100
Enterobacter cloacae	3MRGN ESBL	1	Not applicable	Not applicable
Escherichia coli	3MRGN	4	0	75.0
Escherichia coli	3MRGN ESBL	2	0	100
Escherichia coli	ESBL	1	Not applicable	Not applicable
Klebsiella pneumoniae	3MRGN	2	0	100
Klebsiella pneumoniae	3MRGN ESBL	4	0	100
Klebsiella pneumoniae	4MRGN	7	42.9	85.7
Klebsiella pneumoniae	4MRGN ESBL	1	Not applicable	Not applicable
Morganella morganii	3MRGN	1	Not applicable	Not applicable
Pseudomonas aeruginosa	3MRGN	1	Not applicable	Not applicable
Pseudomonas aeruginosa	4MRGN	10	0	100
Staphylococcus aureus	MRSA	8	0	87.5

Table 6. Factors affecting the decolonization success

	Failed decolonization	Successful decolonization	Early decolonization (number of days until decolonization ≤ median (28) of the assessed cases)	Late decolonization (number of days until decolonization > median (28) of the assessed cases)	Cases with recolonization events
Total (*n* = 172)	77/172 (44.8 %)	95/172 (55.2%)	54/172 (31.4%)	41/172 (23.8%)	26/172 (15.1%)
MRSA (*n* = 10)	9/10 (90.0%)	1/10 (10.0%)	0/10 (0%)	1/10 (50.0%)	6/10 (60.0%)
VRE (*n* = 1)	1/1 (100%)	0/1 (0%)	0/1 (0%)	0/1 (0%)	0/1 (0%)
Gram-negative bacteria (*n* = 161)	67 /161 (41.6%)	94/161 (58.4%)	54/161 (33.5%)	41/161 (25.5%)	20/161 (12.4%)
Pharynx (*n* = 18)	9/18 (50.0%)	9/18 (50.0%)	7/18 (38.9%)	2/18 (11.1%)	8/18 (44.4%)
Nostrils (*n* = 21)	6/21 (28.6%)	15/21 (71.4%)	6/21 (28.6%)	9/21 (42.9%)	5/21 (23.8%)
Skin (*n* = 68)	36/68 (52.9%)	32/68 (47.1%)	21/68 (30.9%)	11/68 (16.2%)	5/68 (7.4%)
Superficial wounds (*n* = 65)	26/65 (40.0%)	39/65 (60.0%)	20/65 (30.8%)	19/65 (29.2%)	8/65 (12.3%)
Systemic antibiotic therapy applied (*n* = 113)	59/113 (52.2%)	54/113 (47.8%)	22/113 (19.5%)	32/113 (28.3%)	20/113 (17.7%)

ynx while decolonization of the nostrils usually required more time *(Table 6)*. Recolonization events were particularly frequent for pharyngeal colonization and for colonization with MRSA. Of note, decolonization success in patients with systemic antibiotic therapy was with 47.8% slightly worse than the average decolonization rate of 55.2% *(Table 6)*.

Effect of systemic antibiotic therapy on the time of initial detection of multidrug-resistant pathogens

To assess selecting effects of systemic antibiotic therapy as a potential reason for the late detection of many assessed cases, cases being detected within the first 72 h after admission to hospital were compared with cases being detected later regarding their association with systemic antibiotic therapy. The results were virtually identical, with 37.8% of cases (48/127) without systemic antibiotic therapy being detected within the first 72 h and 36.1% (66/183) of cases with systemic antibiotic therapy (P = 0.81, not significant) *(Table 7)*.

Discussion

Our present study assessed the efficiency of local decolonization procedures against multidrug-resistant bacteria including factors associated with success or failure. Therefore, heavily colonized patients from international war and crisis zones were assessed, comprising 371 colonization or infection events for as few as 25 individuals.

Although this quantity is strikingly high, it is well in line with previous reports on traumatized patients from war and crisis zones. In a recent report on multidrug-resistant *Acinetobacter* spp. isolates from war victims from the Eastern Ukraine [9], as many as 32 carbapenem-resistant *Acinetobacter* spp. were isolated from 21 patients. This report underlines the tremendous importance of multidrug-resistant bacteria in international war and crisis zones. Of note, not only harmless colonization was observed but one Ukrainian patient also died from a severe infection caused by such a strain [9].

The assessment of nosocomial transmissions was not a major focus of this study as no molecular typing was performed, and it would by far have been beyond the scope

Table 7. Association of antibiotic therapy and first isolation >72 h after hospital admission

(Total = 310)	Number of isolation events without antibiotic therapy	Number of isolation events with antibiotic therapy
First isolation ≤72 h after hospital admission	48	66
First isolation >72 h after hospital admission	79	117

P = 0.81 (Fischer's exact test, two-sided)

of this work. Of note, however, nosocomial transmission from another colonized patient in the same room was likely in no more than a single assessed recolonization event. This finding suggests effectiveness of the applied hygiene procedures. The other recolonization events were either due to auto-inoculation or, theoretically, due to transmissions from other patients on the same ward.

Another interesting finding is the dense colonization of the patients' skin with multidrug-resistant Gram-negative pathogens. The increase of Gram-negative persistence on human skin under conditions of high temperatures and humidity, as found in subtropical and tropical settings, was described as early as in 1975 [10]. Comparable phenomena of severe colonization of the skin of Iraqi patients with Gram-negative bacteria were described by the US American military medical service during the Iraqi war [11]. In the highlands of Madagascar, nasal colonization with Gram-negative bacteria of students and health care workers was 53.0% and even 74.6% for hospital patients in a recent assessment [12]. Accordingly, Gram-negative bacteria play a considerable role as skin colonizers in other regions. This has to be considered if respective patients are treated.

Beneficial effects of disinfecting washing on the reduction of local multidrug-resistant colonization flora are considered as well established [13, 14], also leading to a reduced number of systemic infections [15]. In line with previous considerations [8], patient compliance was shown in our investigation to play a significant role in terms of the effectiveness of disinfection-based local decolonization procedures. Although species-specific epidemiological features are likely to play a role as well [16], the case numbers of the here described assessment were too small to allow for significant results on species level. Anyhow, the compliance effect should not be overestimated. Of note, 44.2% of the observed decolonization events comprised spontaneous decolonization at sites which were not accessible to the applied decolonization procedures. In comparison, 55.2% successful decolonization events were observed at sites being accessible to the applied disinfecting schemes. Accordingly, the quantitative dimension of the additional decolonizing effects of the applied disinfecting procedures has to be considered as moderate.

The phenomenon of relatively rapid spontaneous decolonization of especially Gram-negative multidrug-resistant bacteria has impressively been described for enterically colonized travelers returning from high endemicity settings. Within as little time as three months, a spontaneous tremendous decrease of detectable ESBL-positive colonization flora in the gut was recorded [17]. In a similar way, low enteric colonization with ESBL-positive Enterobacteriaceae was observed in stool samples of German soldiers after deployments to subtropical and tropical settings 2 to 3 months after their return to the home country [18], while, in contrast, considerably higher colonization rates could be found in European soldiers at tropical deployment sites [19].

Accordingly, spontaneous displacement of multidrug-resistant bacteria by endemic colonization flora has to be expected. This assumption is in line with previous observations focusing on facultatively pathogenic bacteria. As early as in 1962, it was known that an intact gut microbiome provides a certain infection resistance against *Shigella flexneri* [20]. Similar results were shown for *Salmonella* spp. in 1964, associated with an increased risk of symptomatic infection in case of disruption of the gut flora by the application of streptomycin [21]. However, as recently shown, this colonization resistance can be restored by the transfer of defined microbiota at least in the mouse model [22]. It is likely that the restoration of the gut microbiome of the travelers after returning home might have led to a restoration of colonization resistance against imported ESBL-positive Enterobacteriaceae [17, 18]. In contrast, attempts of antibiotic application for the decolonization of resistant Gram-negative bacteria in the gut have usually failed so far [23]. This suggests that restoration of the microbial diversity of the gut microbiome might be superior to further depletion of this diversity by antibiotic drug-based approaches.

In line with this, as particularly well studied for infections due to *Clostridium difficile,* dysbiosis as a consequence of a depletion of the microbiome due to antibiotic therapy is likely to support compensatory colonization by undesired microorganisms [24]. Therefore, a higher proportion of late detections of colonization with multidrug-resistant bacteria could have been expected in patients who received systemic antibiotic therapy. Interestingly, this phenomenon was not observed in this study but the high number of interfering factors like new environmental influences after the transfer of the patients to Germany as well as unaccustomed food composition and hygiene regiments make any consideration about potential reasons highly speculative.

Conclusion

In conclusion, patient compliance with enforced hygiene protocols was identified as a determinant of success of disinfection-based decolonization strategies. The quantitative effect in comparison with spontaneous decolonization at other colonization sites that were not accessible to decolonizing hygiene procedures was, however, moderate. Decolonizing washing strategies can therefore contribute to quantitative reduction of colonization with multidrug-resistant bacteria in patients but are not necessarily associated with reliable decolonization success.

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Declaration of interest

The authors declare that there are no conflicts of interest.

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