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Anatomic Sites of Patient Colonization and Environmental Contamination with *Klebsiella pneumoniae* Carbapenemase–Producing Enterobacteriaceae at Long-Term Acute Care Hospitals

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Abstract

OBJECTIVE.—To determine anatomic sites of colonization in patients and to assess environmental contamination with *Klebsiella pneumoniae* carbapenemase (KPC)–producing Enterobacteriaceae.

DESIGN, SETTING, AND PATIENTS.—We conducted a cross-sectional microbiologic survey of 33 patients and their environments at 6 long-term acute care hospitals (LTACHs) in metropolitan Chicago. Swab samples of anatomic sites and inanimate surfaces in patients' rooms and common areas were cultured. *bla*_{KPC} was verified by polymerase chain reaction. Patient charts were reviewed for covariates known to be associated with colonization and environmental contamination.

RESULTS.—Mean age was 66 years. Median length of stay prior to surveillance was 50 days. Thirty (91%) patients were mechanically ventilated, 32 (97%) were bedbound, and 27 (82%) had fecal incontinence. Of the 24 patients with KPC-producing Enterobacteriaceae recovered from 1 or more anatomic sites, 23 (96%) had KPC-producing Enterobacteriaceae detected at 1 or more skin sites. Skin colonization was more common in patients with positive rectal/stool swab cultures or positive clinical cultures ($P < .001$). Rectal/stool swab was the single most sensitive specimen for detecting KPC-producing Enterobacteriaceae colonization (sensitivity, 88%; 95% confidence interval [CI], 68%–97%); addition of inguinal skin swab culture resulted in detection of all colonized patients (sensitivity, 100%; 95% CI, 86%–100%). Only 2 (0.5%) of 371 environmental specimens grew KPC-producing Enterobacteriaceae.

CONCLUSIONS.—Culture of more than 1 anatomic site was required to detect all KPC-producing Enterobacteriaceae-colonized patients. Skin colonization was common, but

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environmental contamination was rare. These results can guide development of multimodal interventions for control of KPC-producing Enterobacteriaceae in LTACHs.

Long-term acute care hospitals (LTACHs) serve patients with multiple comorbidities and complex medical requirements.¹ These features place LTACH patients at high risk of colonization and infection with multidrug-resistant organisms (MDROs), such as methicillin-resistant *Staphylococcus aureus* (MRSA), vancomycin-resistant enterococci (VRE), and extended-spectrum beta-lactamase (ESBL)-producing gram-negative bacteria.² Recently, LTACHs have been found to play an important role in the regional dissemination of an emerging MDRO, *Klebsiella pneumoniae* carbapenemase (KPC)-producing Enterobacteriaceae.^{3,4} Halting the spread of KPC-producing Enterobacteriaceae is a public health priority in the United States and elsewhere.⁵⁻⁷

The most effective measures to prevent transmission of KPC-producing Enterobacteriaceae in LTACHs and other healthcare facilities are unknown. One strategy uses active surveillance to identify and isolate carriers,⁵ but the optimal testing protocol, including which anatomic sites are most sensitive for detecting colonization, is not well defined.⁸ Antiseptic skin cleansing with chlorhexidine gluconate and enhanced environmental cleaning have both been used in successful multimodal interventions to control outbreaks of KPC-producing Enterobacteriaceae, but there are few reports that have informed these interventions. Specifically, little is known about the extent of patient skin colonization or of environmental contamination with this pathogen.⁹⁻¹¹

We hypothesized that extrarectal colonization and environmental contamination with KPC-producing Enterobacteriaceae are common. To test this hypothesis and to develop a biologic basis for KPC-producing Enterobacteriaceae control interventions, we conducted a multicenter culture survey of LTACH patient body sites and inanimate surfaces in patients' environments.

METHODS

We conducted a cross-sectional microbiologic and medical record survey at 6 LTACHs in metropolitan Chicago over a 4-month period in 2011. Patients who were colonized with KPC-producing Enterobacteriaceae were identified through whole-facility point prevalence surveys (1 point prevalence survey per LTACH) that were performed 1–7 weeks earlier as part of an investigation of the emergence of KPC-producing Enterobacteriaceae in the region. Colonization prevalence at individual LTACHs ranged from 10% to 53% of patients.¹² Clinical microbiology laboratory information system databases at each LTACH were also reviewed in order to identify patients with a clinical culture positive for KPC-producing Enterobacteriaceae.

Study subjects were selected so that patients with different total body and potential environmental burdens of KPC-producing Enterobacteriaceae would be evaluated. Subjects were assigned to 1 of 3 groups: patients with a rectal surveillance culture that was negative for KPC-producing Enterobacteriaceae and who did not have a history of KPC-producing Enterobacteriaceae in a clinical culture (group 1), patients with a rectal surveillance culture that was positive for KPC-producing Enterobacteriaceae but who did not have a history

of KPC-producing Enterobacteriaceae in a clinical culture (group 2), and patients with a positive rectal surveillance culture and a history of KPC-producing Enterobacteriaceae in a clinical culture (group 3). We hypothesized that the number of anatomic and environmental sites positive for KPC-producing Enterobacteriaceae would be greatest, on average, for group 3 patients, followed by group 2 patients and then group 1 patients.

Each LTACH was visited only once, at which time all patient and environmental specimens were collected. On the day of the survey, all patients with a previous rectal surveillance culture result were divided into the 3 groups described above. Up to 3 patients in each group were studied; the intention was to test a total of 10 patients in each group.

Swab specimens were collected from 10 × 10-cm areas of intact skin (inguinal, axillary, antecubital fossae, and upper back); from the tracheostomy site, if present, or from the oropharynx if the patient did not have a tracheostomy; from urine, if the patient was not anuric; and from the rectum. If stool was present on perianal skin, this was cultured in lieu of rectal swabbing. Sterile double Dacron swabs (Becton Dickinson) moistened in modified Stuart's medium were used to sample each skin site. Single swabs were used for rectal culture. To sample urine, a double swab was dipped in urine collected in a sterile cup. The time from bathing to swabbing was not recorded, although all facilities had a policy that required daily patient baths.

Sterile, moistened double Dacron swabs were also used to culture environmental sites in occupied patient rooms and in common areas. Sites varied according to facility but included dresser, call button, bed rails, overbed table, infusion pump control panel, ventilator control panel, toilet flusher, computer keyboard, and medicine cabinet door. Common-area sites included equipment in the physical therapy gym and the hemodialysis room and items in staff break rooms. A 10 × 10-cm area of the surface of each site was swabbed when feasible. For smaller surfaces, the entire site was swabbed. For large surfaces, high-touch regions were swabbed. Rooms and common areas were cleaned daily with a quaternary ammonium compound. The time from cleaning to environmental sampling was not recorded.

Swabs were processed within 4 hours of collection. For anatomic site specimens, the first swab was tested for KPC-producing Enterobacteriaceae using a direct ertapenem disk method.¹³ The second swab was placed in 5 mL tryptic soy broth (Remel) containing a 10- μ g meropenem disk (Becton Dickinson; final concentration of meropenem, 2 μ g/mL). If there was no growth in the ertapenem disk test but the broth indicated growth after overnight incubation, a 25- μ L broth aliquot was plated onto a fresh MacConkey agar (Remel), streaked for isolation, and incubated overnight. Because single swabs were used for rectal cultures, broth enrichment was not performed.

For environmental site cultures, both swabs were inoculated into 5 mL tryptic soy broth containing a 10- μ g meropenem (Becton Dickinson) disk. After overnight incubation, turbid broth cultures were plated onto MacConkey agar, and results were interpreted as described above.

Isolated colonies from positive cultures underwent real-time polymerase chain reaction (PCR) testing for *bla*_{KPC}.¹⁴ All *bla*_{KPC}-positive isolates and all gram-negative rods identified

in environmental cultures, regardless of *bla*_{KPC} PCR results, were identified to the species level by the MicroScan Walk-away System (Siemens). Susceptibility of *bla*_{KPC}-positive isolates to chlorhexidine gluconate (Sigma Chemicals) was determined by an agar dilution method.¹⁵

Patient medical records were reviewed for the following information: age, sex, length of stay, medical comorbidities, indwelling medical devices, mobility, incontinence, and antibiotic therapy. To calculate the sensitivity of various anatomic sites for detection of KPC-producing Enterobacteriaceae carriage, we defined the criterion standard for patient carriage as culture positivity for KPC-producing Enterobacteriaceae in any 1 or more anatomic sites. Descriptive statistics and associations between colonization with KPC-producing Enterobacteriaceae and clinical or demographic variables were analyzed by the Fisher exact test and Kruskal Wallis test, where appropriate, using SAS, version 9.1.3 (SAS Institute). The project was reviewed by the Rush University Medical Center Institutional Review Board, which granted expedited approval.

RESULTS

Thirty-three subjects were enrolled: 12 in group 1, 11 in group 2, and 10 in group 3 (Table 1). The number of patients tested at each LTACH ranged from 3 to 8. Overall, 18 (54%) subjects were men, mean age was 66 years, and median length of stay in the LTACH prior to enrollment was 50 days. The majority of subjects surveyed had multiple comorbidities: 30 (91%) were mechanically ventilated, 32 (97%) were bedbound, 28 (85%) had a gastrostomy tube in place, 27 (82%) had fecal incontinence, and 30 (91%) had received at least 1 antibiotic since admission to the LTACH.

Twenty-four patients (3 patients from group 1 and all patients from groups 2 and 3) had at least 1 anatomic site that grew a *bla*_{KPC}-positive isolate, and 23 (96%) patients had more than 1 positive anatomic site (Table 2). The single most sensitive anatomic site was rectal (88%, 95% confidence interval [CI], 68%–97%). Culture of the inguinal and rectal sites together detected all positive patients. Direct plating without broth enrichment was sufficient to detect KPC-producing Enterobacteriaceae at 190 of 191 colonized sites (99.5%).

Colonization of intact skin by KPC-producing Enterobacteriaceae was common. Of the 24 patients who were colonized at any anatomic site, 23 patients (96%) had KPC-producing Enterobacteriaceae detected at 1 or more skin sites. A higher burden of skin site colonization was found in patients from groups 2 and 3 compared to those from group 1. Among patients with negative rectal surveillance cultures (group 1), 3 (25%) grew KPC-producing Enterobacteriaceae from 1 or more inguinal, axillary, back, or antecubital skin sites (Figure 1). In comparison, 10/11 (90%) patients in the rectal surveillance-positive/clinical culture-negative group (group 2) and 10/10 (100%) patients in the rectal surveillance-positive/clinical culture-positive group (group 3) had a KPC-producing Enterobacteriaceae isolate detected at a skin site ($P < .001$; Figure 1). The mean number of skin sites that grew KPC-producing Enterobacteriaceae was 0.5 sites per patient in group 1, 1.8 sites per patient in group 2, and 2.3 sites per patient in group 3 ($P < .001$).

Nineteen patients were colonized with *bla*_{KPC}-positive *K. pneumoniae*, 2 were colonized with *K. pneumoniae* and *Enterobacter aerogenes*, 1 was colonized with only *E. aerogenes*, 1 was colonized with *K. pneumoniae* and *Escherichia coli*, and 1 was colonized with *K. pneumoniae*, *E. aerogenes*, and *E. coli*. For the 32 *bla*_{KPC}-positive isolates identified from patient skin sites, minimum inhibitory concentrations (MICs) of chlorhexidine ranged from 1 to 128 µg/mL. The geometric mean MICs were 41.4 µg/mL for 27 *K. pneumoniae* isolates and 48.5 µg/mL for 5 other isolates (4 *E. aerogenes* and 1 *E. coli*).

A total of 371 environmental sites were cultured. Of these, 2 sites (0.5%; bed rails and call button) were positive for KPC-producing Enterobacteriaceae. Both of these sites were in the room of a group 3 patient who had a history of KPC-producing Enterobacteriaceae from a sputum culture; 6/7 (86%) of this patient's anatomic sites grew KPC-producing Enterobacteriaceae in our surveillance cultures. Fifty-seven (15%) environmental sites grew other carbapenem-resistant gram-negative bacteria: 53 *Acinetobacter baumannii*, 2 *Pseudomonas aeruginosa*, 1 *Achromobacter xylosoxidans*, and 1 *Chryseobacterium indologenes*.

DISCUSSION

The best strategies for surveillance and control of KPC-producing Enterobacteriaceae in acute and long-term care facilities are still being defined. Our study provides information that should prove useful to infection preventionists and others who are working to prevent cross transmission of KPC-producing Enterobacteriaceae in LTACHs. First, we demonstrated that although rectal/stool swab culture was the single most sensitive specimen type for detecting KPC-producing Enterobacteriaceae carriage, adding culture of inguinal skin improved sensitivity. Inguinal and rectal screening together detected all colonized patients. These findings are consistent with those of other investigators who determined that the inguinal skin was a frequent site of ESBL-producing Enterobacteriaceae colonization among short-term acute care hospital patients.^{16,17} Culture of multiple anatomic sites may be necessary to optimize detection of colonized patients.

Colonization of other skin sites with KPC-producing Enterobacteriaceae was also common, especially among patients with rectal carriage or with a prior positive clinical culture. Since KPC-producing Enterobacteriaceae are assumed to be transmitted from patient to patient via contaminated healthcare worker hands, these findings suggest that reducing the burden of KPC-producing Enterobacteriaceae on patients' skin—a likely source of healthcare worker hand contamination—should be explored as a means of reducing cross transmission in LTACHs. Cleansing the skin of critically ill patients daily with chlorhexidine gluconate has been shown to decrease horizontal spread of VRE, MRSA, and *Acinetobacter* spp. in short-term acute care hospital intensive care units.^{18–20}

When planning a chlorhexidine gluconate skin-cleansing intervention in an LTACH, one must consider at least 4 issues. First, a recent report of reduced susceptibility to chlorhexidine in the hyperepidemic KPC-producing *K. pneumoniae* strain ST258 raises the concern that chlorhexidine skin cleansing may prove less effective for these pathogens than for the MDROs studied earlier. Chlorhexidine MICs of the isolates tested in our study

remain within a range that would be expected to be inhibited by commercially available chlorhexidine skin cleansers, since, for example, 2% chlorhexidine solutions contain 20,000 μg of chlorhexidine per milliliter. Second, cleaning protocols need to anticipate the risk of skin recontamination by these patients, who often have fecal incontinence (Table 1). Third, the high rates of axillary skin positivity (Figure 1) raise concerns about the modes of such contamination and the need for special attention to cleaning this easily missed site. Fourth, the potential role of KPC-producing Enterobacteriaceae as resident skin flora, rather than as transient skin colonizers, should be explored.

While skin colonization with KPC-producing Enterobacteriaceae was common, contamination of the environment was rare. Only 2 (0.5%) sites (bedrail and call button) were found to be contaminated. Both of these sites were in the room of a patient with a relatively high body burden of KPC-producing Enterobacteriaceae, with 6 of 7 anatomic sites tested culture positive. Additionally, unlike many of the patients studied, this patient was able to intentionally touch his immediate environment, which may have led to its contamination. Overall, current environmental cleaning protocols at the LTACHs appeared to have been adequate to limit environmental contamination with KPC-producing Enterobacteriaceae. Alternatively, these pathogens may not survive as well on dry inanimate surfaces as other gram-negative bacilli.

Our study has limitations. Although we included patients from 6 of 7 LTACHs in metropolitan Chicago, the sample size was small and may not have been representative of all LTACH patients. Broth enrichment was not used for culturing the rectal swab specimens, which may have resulted in lower sensitivity of detecting KPC-producing Enterobacteriaceae at that anatomic site compared to other sites. However, for non-rectal sites, broth enrichment had minimal effect on sensitivity; only 1 (0.5%) additional site was detected by broth enrichment, suggesting that that broth enrichment would not have improved sensitivity of rectal swab screening greatly. The use of double swabs to collect environmental specimens may be an insensitive method of environmental sampling because of swabs' relatively small surface area. Sensitivity of our environmental culture method was enhanced by using broth enrichment, however, and we were able to identify other carbapenem-resistant gram-negative bacteria in 15% of environmental cultures, suggesting that our method was adequate. Additionally, because our study was performed at LTACHs with high prevalence of KPC-producing Enterobacteriaceae, these results may not be generalizable to facilities with lower prevalence.

In the United States, LTACHs are increasingly being identified as reservoirs for KPC-producing Enterobacteriaceae.^{3,4,21} Focused interventions to reduce cross transmission of KPC-producing Enterobacteriaceae at these facilities are therefore warranted. The frequent skin contamination identified in our study suggests that careful attention to patient skin antisepsis and to healthcare worker hand hygiene may be essential interventions to decrease spread. These hypotheses should be investigated.

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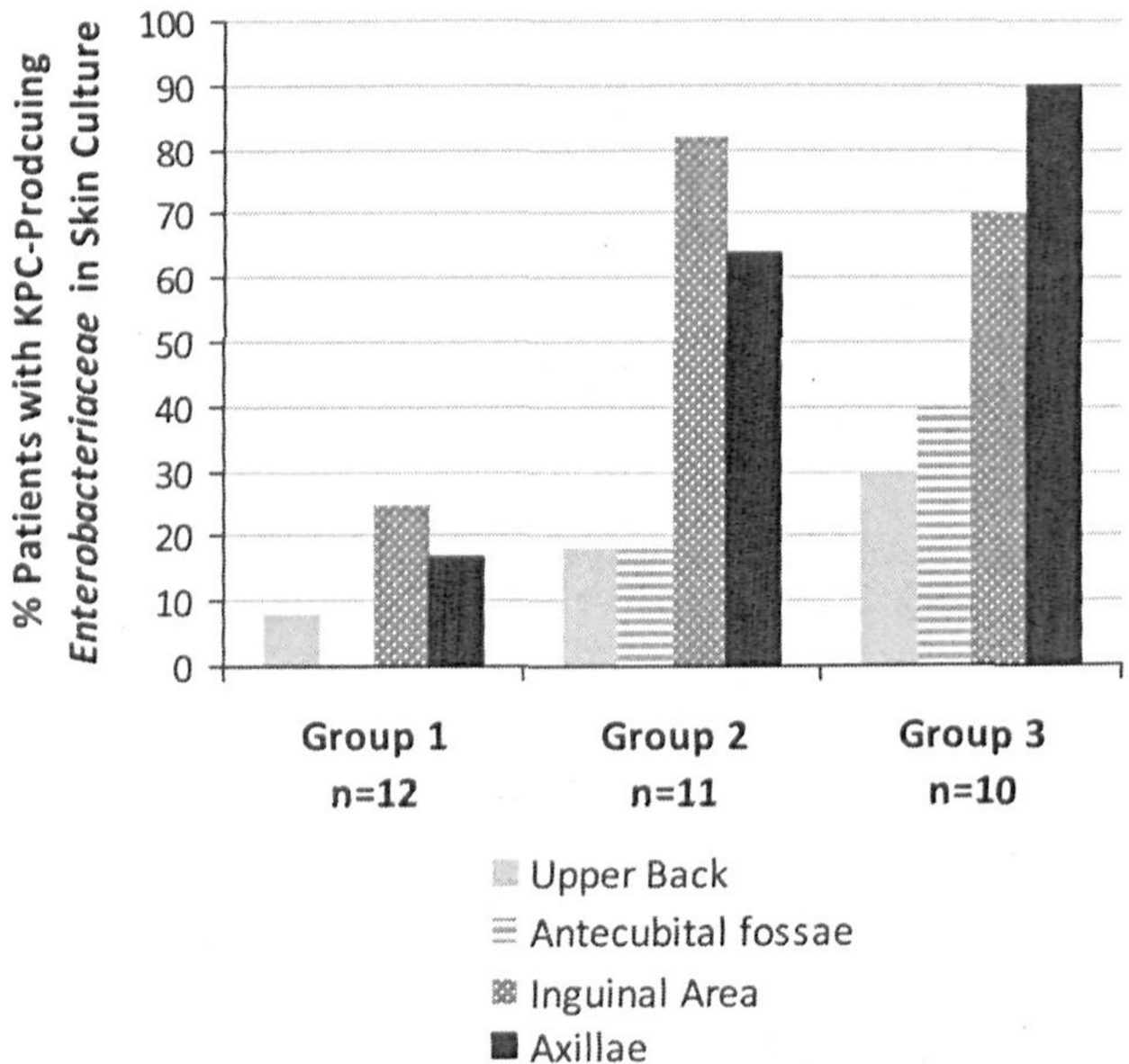


FIGURE 1.

Skin colonization with *Klebsiella pneumoniae* carbapenemase (KPC)-producing Enterobacteriaceae among the 3 groups of patients surveyed, by skin site. Group 1, patients with rectal surveillance and clinical cultures negative for KPC-producing Enterobacteriaceae; group 2, patients with a positive rectal surveillance culture for KPC-producing Enterobacteriaceae but no history of KPC-producing Enterobacteriaceae in a clinical culture; group 3, patients with a positive rectal surveillance culture and a history of KPC-producing Enterobacteriaceae in a clinical culture.

TABLE 1.

Demographic and Clinical Characteristics of Study Subjects

Characteristic	Group 1 (n = 12)	Group 2 (n = 11)	Group 3 (n = 10)
Male sex	5 (42)	8 (73)	5 (50)
Mean age, years (SD)	66 (14)	68 (11)	64 (18)
Median length of stay prior to culture survey, days (IQR)	41 (28–93)	54 (42–65)	39 (24–50)
Bedbound	11 (92)	11 (100)	10 (100)
Gastrostomy tube	9 (75)	10 (91)	9 (90)
Fecal incontinence	9 (75)	11 (100)	7 (70)
Mechanically ventilated	10 (83)	10 (91)	10 (100)
Diabetes mellitus	5 (42)	8 (73)	6 (60)
Malignancy	2 (17)	2 (18)	2 (20)
Cardiovascular disease	6 (50)	8 (73)	3 (30)
Dementia	1 (8)	3 (27)	1 (10)
Hemodialysis	3 (25)	2 (18)	4 (40)
Cerebrovascular accident	1 (8)	5 (46)	2 (20)
Treatment with any antibiotic at time of culture survey	8 (67)	9 (82)	8 (80)
Treatment with a carbapenem at time of culture survey	2 (17)	2 (18)	0 (0)
Treatment with a fluoroquinolone at time of culture survey	1 (8)	0 (0)	1 (10)
Treatment with a cephalosporin at time of culture survey	1 (8)	0 (0)	1 (10)

NOTE. Data are no. (%) unless otherwise indicated. Group 1, patients with rectal surveillance and clinical cultures negative for *Klebsiella pneumoniae* carbapenemase (KPC)-producing Enterobacteriaceae; group 2, patients with a positive rectal surveillance culture for KPC-producing Enterobacteriaceae but no history of KPC-producing Enterobacteriaceae in a clinical culture; group 3, patients with a positive rectal surveillance culture and a history of KPC-producing Enterobacteriaceae in a clinical culture. IQR, interquartile range; SD, standard deviation.

TABLE 2.
Sensitivity of Culture of Different Anatomic Sites for *Klebsiella pneumoniae* Carbapenemase-Producing Enterobacteriaceae

	No. of positive cultures (N = 24)	Sensitivity, % (95% CI)
Skin sites		
Inguinal	19	79 (58–93)
Axillary	18	75 (53–90)
Upper back	6	25 (10–47)
Antecubital fossae	6	25 (10–47)
Nonskin sites		
Rectal ^a	21	88 (68–97)
Urine (N = 19) ^b	10	53 (29–76)
Oropharyngeal/tracheal secretions	10	42 (22–63)
Combined sites		
Rectal and inguinal	24	100 (86–100)
Rectal and axillary	23	96 (79–100)
Axillary and inguinal	22	92 (73–99)

NOTE. CI, confidence interval.

^aThree patients had negative rectal swab cultures but positive cultures of inguinal skin.

^bFive patients were anuric, so urine was not collected for culture.