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Multidrug-Resistant Gram-negative Bacilli Colonization Risk Factors among Trauma Patients

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Abstract

Prior studies have demonstrated high rates of colonization and infection with multidrug-resistant Gram-negative bacilli (MDR-GNB) in injured military personnel. Our analysis shows that injuries inflicted during peak combat periods, massive blood transfusion requirement, and post-trauma cefazolin prophylaxis (additive effect with fluoroquinolones) were risk factors for MDR-GNB colonization.

Keywords

multidrug-resistant organisms; multidrug-resistant gram-negative bacilli; military health; colonization

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Infections with multidrug-resistant Gram-negative bacilli (MDR-GNB), such as extended-spectrum β-lactamase (ESBL)-producing *Escherichia coli*, pose a dangerous threat to individuals worldwide, and in particular to those with battlefield injuries (Murray, 2008). One prospective longitudinal study showed that individuals deployed in Afghanistan and Iraq were almost three times more likely to be colonized with MDR-GNB than those who were not deployed, and personnel with recent injuries possessed a greater risk (Weintrob et al., 2013). Prior research demonstrates colonization occurs primarily through hospital-associated transmission, and rates increase as patients transit through the medevac chain, which involves multiple facilities. Given these findings, our objective was to further ascertain risk factors for MDR-GNB colonization by examining data from wounded military personnel.

Injured service members evacuated through Landstuhl Regional Medical Center (LRMC; Germany) and admitted to a participating US hospital (National Capital Region or San Antonio Military Medical Center) between June 2009 and May 2012 were included. Patient characteristics were captured through the DoD Trauma Registry, while antimicrobial administration, infections, and active surveillance cultures (ASC) were obtained through the supplemental Trauma Infectious Disease Outcomes Study infectious disease module (Tribble et al., 2011). An ASC specimen was obtained from groin or axilla swabs performed within two days of US hospital admission. Gram-negative bacilli were determined to be multidrug-resistant if they showed resistance to at least three of four antibiotic classes or were producers of ESBL or carbapenemases (Division of Healthcare Quality Promotion, 2008.). Antimicrobial susceptibility tests were performed using automated systems (BD Phoenix [BD Biosciences, Sparks, MD] or Vitek 2 [bioMerieux Inc., Hazelwood, MO]) along with disc diffusion and E-test methods (Clinical and Laboratory Standards Institute, 2009). Characteristics were compared using Chi-square or Fisher's exact test (SAS version 9.4, SAS, Cary, NC). Logistic regression models were performed to evaluate risk factors associated with MDR-GNB colonization.

Among the 2079 trauma patients, 289 (14%) were colonized with MDR-GNB (Table 1). *E. coli* was the most common MDR (or ESBL-producing) organism isolated (74% out of 301 bacterial isolates), followed by *Acinetobacter baumannii* complex (15%), *Klebsiella pneumoniae* (10%), *Enterobacter cloacae* (1%), and *Citrobacter* spp. (<1%). Two hundred and twenty-two (77%) colonized patients were classified as having ESBL-producing *E. coli*.

Overall, 284 (98%) colonized patients received antibiotics (for prophylaxis or empiric therapy) prior to ASC with most being prescribed more than one. Cefazolin, with/without fluoroquinolones, was most commonly prescribed and was received by 823 (40%) and 839 (40%) trauma patients, respectively. Among MDR-GNB colonized patients, 57% and 35% received cefazolin with/without fluoroquinolones, respectively. Administration of antibiotics was so common that 93% of colonized patients who were given fluoroquinolones also received cefazolin. Few patients received only cefazolin or fluoroquinolones alone (111 and 5 patients, respectively). A total of 101 patients received only doxycycline (for malaria prophylaxis and not active infection) and 136 did not receive any antibiotics, of which 2% and 4% were colonized, respectively.

Since the population of patients receiving only one antibiotic was small, and because colonization rates among those who received either doxycycline alone or no antibiotics were similar, both were considered as a reference group. Statistically significant variables in the univariate analysis were assessed using stepwise selection for inclusion in the multivariate logistic regression analysis (Table 1). Peak combat period, blood transfusion requirement, and cefazolin administration with/without fluoroquinolones were independently associated with MDR-GNB colonization. It is noteworthy that cefazolin plus fluoroquinolones had a higher risk than cefazolin without fluoroquinolones. When the model was restricted to ESBL-producing *E. coli*, similar statistically significant results were observed (data not shown). A restricted model directly compared cefazolin with/without fluoroquinolones, excluding all other patients. In this model, cefazolin with fluoroquinolones was an independent predictor of MDR-GNB colonization (odds ratio: 1.57; 95th confidence interval: 1.12–2.19).

Our study demonstrates that wounded military personnel are often colonized with MDR-GNBs, predominantly ESBL-producing *E. coli*, which corresponds with previous data showing a 5.5-fold increase of ESBL-producing *E. coli* colonization among personnel in Afghanistan compared with those in the US (Vento et al., 2013). Being injured during peak combat periods was associated with increased risk for colonization and may be reflective of the large number of patients transitioning through combat support hospitals during this time or possibly represents a seasonal pattern among these healthcare-associated infections. Blood transfusions were also associated with MDR-GNB colonization, which is not unexpected as large-volume transfusions are a marker of injury severity and likely related to patients' increased exposure to antibiotics and location in a critical care setting.

One important finding was an additive risk for MDR-GNB colonization when cefazolin was combined with fluoroquinolone administration. A possible explanation for the synergistic effect seen for increased MDR-GNB colonization with multiple antibiotics is the high prevalence of antibiotic-resistance gene transfer and acquisition between bacteria. Our results show that MDR-GNB colonization risk remains high with antibiotic exposure regardless of intensive care unit admission, suggesting a major role for resistance emergence in addition to endogenous bacteria flora within high-risk critical care environment. While we recognize the limitation of using doxycycline as a reference group, and would have preferred instead to analyze colonization rates of individual antibiotics and their combinations against cefazolin exposure alone, there is value in understanding this complicated, but likely important, relationship. It also reflects the reality that the majority of patients receive several antibiotics concurrently.

Antibiotics, such as fluoroquinolones and cephalosporins, are routinely prescribed for surgical prophylaxis and treatment of infectious complications. Cefazolin forms the backbone of most post-trauma prophylactic guidelines (Hospenthal et al., 2011) and research shows that fluoroquinolones offer no advantage over cefazolin and may even deter wound healing. Current civilian and military guidelines do not recommend coverage for Gram-negative organisms except in wounds with extensive damage (Hoff et al., 2011). Thus, with the results of our study demonstrating an increased risk for MDR-GNB colonization with administration of multiple antibiotics, adherence to current practice guidelines and evidence-

based infection control measures is a critical component of antibiotic stewardship required to halt the precipitous rise of colonization in MDR-GNB infections.

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Highlights

- 14% of wounded soldiers colonized with multidrug-resistant Gram-negative bacilli
- 74% of bacteria was extended spectrum β-lactamase-producing *Escherichia coli*
- Combat period, blood transfusion, and cefazolin use associated with colonization
- Additive risk for colonization when cefazolin combined with fluoroquinolones

Table 1

Analysis of Potential Risk Factors Associated with Multidrug-Resistant Gram-Negative Bacilli (MDR-GNB) Colonization

Potential Risk Factor	**Patients without MDR-GNB Colonization (N=1790)**	**Patients with MDR-GNB Colonization (N=289)[a]**	**Univariate Odds Ratio (95% Confidence Interval)[b]**	**Multivariate Odds Ratio (95% Confidence Interval)[b]**
Age at injury, median years (IQR)	24.5 (21.9, 28.7)	23.4 (21.6, 27.5)	0.98 (0.95, 0.99)	0.98 (0.96, 1.01)
Combat-related injury	1580 (88%)	276 (96%)	2.82 (1.59, 5.01)	–
Peak combat period (April – September)[c]	1040 (58%)	200 (69%)	1.62 (1.24, 2.1)	1.76 (1.31, 2.35)
Military Operational Theater				
Afghanistan	1571 (88%)	267 (92%)	Reference	Reference
Iraq	178 (10%)	18 (6%)	0.60 (0.36, 0.98)	0.89 (0.49, 1.59)
Non-theater	41 (2%)	4 (1%)	0.57 (0.20, 1.62)	1.55 (0.43, 5.65)
Blast mechanism of injury	1200 (67%)	224 (78%)	1.69 (1.26, 2.27)	–
Traumatic amputation (excluding digits)	337 (19%)	91 (32%)	1.99 (1.51, 2.62)	–
Injury Severity Score[d]				
0–9 (mild)	731 (41%)	73 (25%)	Reference	Reference
10–15 (moderate)	394 (22%)	74 (26%)	1.88 (1.33, 2.66)	1.29 (0.86, 1.92)
16 – 24 (severe)	419 (23%)	92 (32%)	2.20 (1.58, 3.06)	1.28 (0.83, 1.97)
≥25 (life-threatening)	245 (14%)	50 (17%)	2.04 (1.39, 3.01)	0.94 (0.56, 1.60)
First 24 hour blood transfusion				
No units[e]	1056 (59%)	100 (35%)	Reference	Reference
1–9 units	403 (23%)	88 (30%)	2.31 (1.69, 3.14)	1.77 (1.22, 2.57)
10 – 20 units	184 (10%)	55 (19%)	3.16 (2.19, 4.54)	2.53 (1.55, 4.11)
> 20 units	147 (8%)	46 (16%)	3.31 (2.24, 4.88)	2.80 (1.63, 4.81)
Infection prior to US hospital admission	155 (9%)	45 (16%)	1.95 (1.36, 2.78)	1.35 (0.88, 2.09)
ICU Admission[f]				
LRMC only	263 (15%)	46 (16%)	1.46 (1.01, 2.12)	0.93 (0.60, 1.45)
US hospitals ± LRMC	643 (36%)	138 (48%)	1.79 (1.36, 2.36)	0.84 (0.57, 1.26)
Non-ICU	877 (49%)	105 (36%)	Reference	Reference
Mechanical ventilation at LRMC	571 (32%)	130 (45%)	1.75 (1.36, 2.25)	–
Central line at LRMC	270 (15%)	65 (22%)	1.63 (1.20, 2.22)	–
Indwelling orthopedic hardware at LRMC	545 (30%)	102 (35%)	1.25 (0.96, 1.62)	0.87 (0.65, 1.16)

Potential Risk Factor	Patients without MDR-GNB Colonization (N=1790)	Patients with MDR-GNB Colonization (N=289)[a]	Univariate Odds Ratio (95% Confidence Interval)[b]	Multivariate Odds Ratio (95% Confidence Interval)[b]
Antibiotic administration[g]				
No antibiotics or only doxycycline	230 (13%)	7 (2%)	Reference	Reference
Cefazolin (with/without other antibiotics; excluding fluoroquinolones)	722 (40%)	101 (35%)	4.60 (2.11, 10.03)	3.50 (1.54, 7.93)
Cefazolin plus fluoroquinolone (with/without other antibiotics)	675 (38%)	164 (57%)	7.98 (3.69, 17.26)	5.42 (2.39, 12.31)

ASC - active surveillance for asymptomatic colonization; ICU - intensive care unit; IQR - interquartile range; LRMC - Landstuhl Regional Medical Center (Germany; first evacuation hospital leaving combat zone)

[a] All listed variables, except for indwelling orthopedic hardware, are statistically significantly different ($p<0.05$) compared to the non-colonized group.

[b] Variables with a significance cut-off value of $p=0.3$ in the logistic regression univariate model were assessed for inclusion in the multivariate model using stepwise selection.

[c] A higher proportion of combat-related injuries occurred during the listed months compared to October-March, which was used as the reference.

[d] The injury severity score is an overall value based on anatomical regional injury values.

[e] Ninety-five patients received no units of blood and 1061 were missing blood transfusion data. The median injury severity score for the patients with missing data was 8 (mild injury), 97% did not sustain an amputation, and 73% were not admitted to the intensive care unit in Landstuhl, which is suggestive that they likely did not require large-volume blood transfusions. The group of patients with missing blood data or no units (N=1156) had significantly less ICU admissions, amputations, lower injury severity scores, and lower shock indexes compared to the patients with 1–9 units of blood ($p<0.0001$). Therefore, the group of patients with no units of blood or missing blood data was used as the reference category.

[f] Admission to the ICUs is recorded within the first week of care at each facility. Data are missing from seven patients without MDR-GNB colonization.

[g] Administration of doxycycline alone was likely for antimalarial chemoprophylaxis. It was included with ‘no antibiotics’ as the reference group due to the low numbers of patients who only received doxycycline and had MDR-GNB colonization (N=2). Other antibiotics given to patients who received cefazolin include (but are not limited to) amoxicillin/clavulanate, clindamycin, doxycycline, and metronidazole. There were 163 non-colonized and 17 colonized patients who received varying combinations of other antibiotics without cefazolin, including (but not limited to) clindamycin, meropenem, piperacillin/tazobactam, and vancomycin. Due to their lack of receipt of cefazolin, these 180 patients were not included in the logistic regression univariate/multivariate analyses.