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MRSA Nasal Burden and Risk of MRSA Infection

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Master of Science

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MRSA Nasal Burden and Risk of MRSA Infection

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An abstract of
A thesis submitted to the Faculty of the
James T. Laney School of Graduate Studies of Emory University
in partial fulfillment of the requirements for the degree of
Master of Science
in Clinical Research
2012

Abstract

MRSA Nasal Burden and Risk of MRSA Infection
By Edward Alan Stenehjem

Introduction: The burden of nasal *Staphylococcus aureus* colonization has been linked to an increased risk of staphylococcal infection. The cycle threshold (C_t) of molecular diagnostic tests used to identify methicillin-resistant *S. aureus* (MRSA) nasal colonization is a quantitative value of burden and may have clinical implications.

Methods: A retrospective cohort study was used to assess the effect MRSA nasal colonization burden has on subsequent MRSA infection. Veterans were classified as non-carriers, low burden MRSA ($C_t > 24$) carriers or high burden MRSA carriers ($C_t \leq 24$) based on admission nasal surveillance swabs. MRSA infections were identified prospectively. Chart reviews were performed to obtain important clinical information such as demographics, comorbidities, and presence of wounds or devices. A multivariate logistic regression model was used to assess the association of MRSA nasal colonization burden (negative, low, and high) and risk of subsequent MRSA infection.

Results: From October 1st, 2007, to February 1st, 2008, 205 patients were admitted to the Atlanta VA Medical Center with positive MRSA nasal colonization, 68.8% had a $C_t > 24$ and 31.2% had a $C_t \leq 24$. A comparison group from the same time period, consisting of 141 patients with negative MRSA nasal colonization, was randomly selected. 6 MRSA infections occurred in patients with negative colonization (4.3%), 26 with low burden (18.5%), and 11 with high burden (17.2%). In multivariate analysis, MRSA nasal colonization was a risk factor for subsequent MRSA infection ($p = 0.0081$). Low burden (RR 3.62, 95% CI 1.47 – 8.93) and high burden (RR 2.71, 95% CI 0.95 – 7.72) were both associated with subsequent MRSA infection when compared to patients without nasal MRSA colonization. High burden of nasal MRSA was not a significant risk factor (RR 0.75, 95% CI 0.36 – 1.55) when compared to low nasal burden of MRSA.

Conclusions: Among Atlanta veterans, MRSA nasal colonization was a risk factor for subsequent MRSA infection. However, patients with a higher nasal burden of MRSA, as defined by the C_t from the Xpert MRSA assay, were not at an increased risk of MRSA infection when compared to patients with low nasal burden of MRSA.

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Table of Contents

Introduction

Staphylococcus aureus is a well-known, gram-positive bacteria that can cause significant morbidity and mortality in those infected. *S aureus* is one of the most common causes of skin and soft tissue infection in community settings but can also cause severe nosocomial infections. During the past twenty years an epidemic of drug resistant *S aureus* has emerged. This drug resistant *S aureus,* known as methicillin-resistant *S aureus* or MRSA, is more virulent and leads to greater morbidity and mortality than drug susceptible *S aureus(1).* The MRSA epidemic has lead researchers and healthcare professionals to investigate methods of preventing the spread of MRSA within hospitals and also identify risk factors for infection.

In addition to causing clinically important infections, *S aureus* is also a human commensal. Although multiple anatomic body sites can be colonized with *S aureus,* the anterior nares is the most frequent carriage site. The association of nasal colonization and subsequent staphylococcal disease was first reported in 1931 by Danbolt(2) and is now a well known risk factor for subsequent infection. However, not all nasal colonization carries the same risk of subsequent infection. *S aureus* nasal carriage can be classified into three main groups: persistent carriage, intermittent carriage, and non-carriage. This distinction is important because persistent carriers have a higher *S aureus* nasal burden and higher risk of acquiring *S aureus* infection when compared to those with intermittent or non-carriage(3, 4).

The MRSA epidemic led many healthcare institutions to perform active surveillance of nasal MRSA carriage in an attempt to reduce transmission of MRSA within healthcare settings. In most instances, while admitted to acute care facilities, MRSA carriers are

placed in contact isolation in an attempt to reduce transmission to healthcare workers and subsequently to other patients. The Veterans Health Administration (VHA) was an early adopter of active nasal surveillance and now mandates nasal MRSA surveillance of all patients admitted to acute care settings in the United States.

The VHA uses a molecular test to identify carriers of MRSA, the Xpert MRSA assay (Cepheid, Sunnyvale, CA). This test, although reported as positive or negative, has a quantitative component that has yet to be evaluated in clinical settings. The Cycle Threshold (C_t), the time it takes for the test to become positive, is inversely proportional to the burden of MRSA in the specimen tested. The shorter the C_t, the more MRSA is present.

We hypothesize nasal MRSA burden, as measured by the C_t of the Xpert MRSA assay, is a risk factor for subsequent MRSA infection. Our study identified a cohort of veterans admitted to the Atlanta Veterans' Affairs Medical Center (AVAMC) and followed them for four years for the development of MRSA infections. This cohort was used to analyze the affect admission nasal MRSA burden had on the development of subsequent MRSA infection. The results of this study will begin to identify potential uses of quantitative nasal colonization results.

Background

The Evolution of MRSA

Prior to the introduction of penicillin in the early 1940's, invasive *S aureus* infection caused significant morbidity and mortality. The introduction of penicillin greatly reduced this burden of disease but its effect was short lived (5). Only one year after the introduction of penicillin, the first penicillin-resistant *S aureus* was reported (6). As use of penicillin increased in hospitals after World War II, penicillin-resistant *S aureus* began to increase dramatically in response to increased antibiotic pressure (7). Paralleling the current MRSA epidemic, resistance to penicillin was first identified in hospitalized patients and subsequently spread to the community. The production of a β-lactamase was responsible for the resistance of *S aureus* to penicillin. This increase in resistance was the stimulus for the development of semi-synthetic penicillin, which would remain active in the presence of the inactivating enzymes. Methicillin was subsequently introduced in 1961 and was active against β-lactamase-producing *S aureus.*

Within one year of methicillin's introduction, MRSA emerged as an important clinical pathogen in the United Kingdom (8). MRSA is characterized by the expression of the *mecA* gene, which encodes a protein (PBP2a) with low affinity for β-lactam antibiotics. This *mecA* gene, carried by the staphylococcal cassette chromosome *mec* (SCC*mec*), was likely acquired from coagulase-negative staphylococcus via horizontal transfer (9-11).

MRSA outbreaks in U.S. hospitals were commonly described in the medical literature during the late 1960's and early 1970's (12, 13). Over the next 20 years, MRSA infections became increasingly more common in hospitalized populations. The

National Nosocomial Infections Surveillance (NNIS) system began collecting data on nosocomial infections in 1970. In the early 1980's, data from the NNIS system demonstrated a slow increase in MRSA nosocomial infections to a rate of 5 – 10% of all *S aureus* isolates collected. Rates among small, community hospitals remained low (<5%) at that time. The epidemiology of nosocomial MRSA infections dramatically changed in the late 1980's and early 1990's. By the early 1990's, the proportion of *S aureus* isolates due to MRSA had increased to 20% among small community hospitals and was 40% in large urban medical centers (14, 15). In 2003, MRSA accounted for 64% of all nosocomial *S aureus* infections (16).

As nosocomial MRSA infections became more common in the late 1990's, sporadic cases of MRSA infections began to appear in the general community without identifiable MRSA risk factors (17). Mimicking the trends seen with penicillin-resistant *S aureus,* rates of community-acquired MRSA rose dramatically and now account for the majority of *S aureus* skin and soft tissue infections. Although community-acquired strains and hospital-acquired strains share a common resistance to methicillin, the genetic lineage of the strains and resistance profiles were originally distinct. Now, the once clear distinction between hospital-acquired and community-acquired strains has become blurred as strains commonly associated with CA-MRSA are causing nosocomial infections(18, 19).

Nasal Colonization and Risk of Infection

S aureus is both a human commensal and a frequent cause of infections that cause significant morbidity and mortality. *S aureus* colonizes the skin and mucosa of

humans at multiple sites, with the anterior nares being the most frequent carriage location. The association between *S aureus* nasal colonization and staphylococcal infection was first described in the 1930's. Multiple studies have subsequently confirmed nasal colonization as a risk factor for infection. At the Brooke Army Medical Center in Texas, 19% of patients with MRSA nasal colonization developed a subsequent MRSA infection within one year compared to 2% without nasal colonization(20). Huang et al. found similar results in a Boston cohort of patients with 29% of colonized or infected patients developing subsequent MRSA infection within 18 months of discharge from the hospital(21) . Nasal colonization is also a significant risk factor for development of surgical site infections. In a study of United States (US) veterans, preoperative nasal colonization was a significant risk factor for the development of post operative surgical site infections after controlling for multiple other confounding variables (Relative Risk 8.1)(22). In addition to MRSA causing more severe infections as compared to methicillin-susceptible *S aureus* (MSSA)*(1),* MRSA colonization is a greater risk factor for subsequent infection than MSSA colonization(23).

A casual relationship between *S aureus* nasal carriage and subsequent infection is supported by multiple studies demonstrating colonizing strains are identical to those that cause subsequent infection. Von Eiff et al. identified 14 patients originally colonized with *S aureus* who subsequently developed *S aureus* bacteremia. In 12 of 14 patients (86%), the original colonizing strain was clonally identical to the isolates obtained from the blood 1 day to 14 months later(24). Wertheim et al. showed similar results with 32 of 40 (80%) invasive *S aureus* strains being identical to the nasal strain detected on

admission(25). Although speculative, identifying and typing *S aureus* clones at other body sites (perineum, axilla, etc.) may have increased this percentage to close to 100%.

One major limitation of studies evaluating nasal colonization and risk of subsequent infection is the determination of colonization status by a single nasal culture. Longitudinal studies clearly distinguish at least three patters of *S aureus* carriage: Persistent carriage, intermittent carriage, and non-carriage (some studies include a fourth category of occasional carriers)(26-29). Most studies define persistent nasal carriage as individuals with ≥ 80% of weekly nasal swabs positive for *S aureus*. In addition to having persistently positive nasal cultures, persistent carriage is associated with a higher burden of *S aureus* in the nose (i.e. more colony forming units per swab). In 1960, Arthur White described his work exploring persistent carriage and quantitative nasal cultures. He found only 5.3% of the positive cultures from patients with intermittent nasal colonization contained more than 10^5 colonies per swab. In contrast, 47.2% of patients with persistent colonization had cultures containing more than 10^5 colonies per swab(30). These findings have been validated in a number of recent publications as well(31, 32).

Higher nasal bacterial burden and the persistently colonized state are also associated with an increased risk of subsequent infection. This trend was initially seen in 1963 with increased postoperative staphylococcal infections among patients with heavy nasal carriage (more than 100,000 colonies per swab). White et al. found an 8% infection rate in those without nasal colonization and a progressively increasing rate of infection with increasing nasal *S aureus* burden, up to 29% in those with > 10^6 colonies per swab(3). This finding has also been observed in two more recent studies. In a

population of patients undergoing orthopedic surgery in the Netherlands, patients with a high nasal staphylococcal burden preoperatively were 16 times more likely to develop a postoperative infection with *S aureus* compared to those without high level carriage (either low burden or negative colonization)(33). In patients undergoing continuous peritoneal dialysis, increased *S aureus* infections related to dialysis were observed in persistently colonized patients compared to those with intermittent or negative colonization*(4)*.

The above findings demonstrate the increased risk of staphylococcal infection among high-risk patients with a high nasal *S aureus* burden. To date, there has not been a study strictly evaluating MRSA nasal burden among average risk patients in the US during the current MRSA epidemic. Also, all previous studies have utilized standard quantitative culture techniques for determining nasal burden, a burdensome and labor intensive process not likely useful in clinical practice. The clinical utility of evaluating quantitative nasal *S aureus* burden has yet to be defined and studies evaluating its role in average risk patients are needed.

Active Surveillance and Molecular Methods of Detection and Quantification

In the 1990's, during the height of the hospital based MRSA epidemic in the US, the Society of Healthcare Epidemiology of America encouraged the use of active surveillance cultures to identify patients colonized with MRSA(34). The goal of active surveillance cultures was to identify patients colonized with MRSA and take measures to prevent transmission of MRSA within healthcare settings. Typically, once patients were identified with MRSA colonization, healthcare workers would wear gowns and gloves during routine patient care to prevent transmission to healthcare workers and the

subsequent transmission to non-colonized patients. The VHA was an early adopter of the active surveillance strategy and mandated MRSA nasal screening on admission to all VA acute care facilities in the US.

One of the limitations of surveillance cultures is the time required for cultures to grow. In some studies, delays up to 5 days have been reported from the time a surveillance culture was obtained until the result was reported(35). This delay can allow colonized patients to serve as persistent reservoirs of MRSA until culture results are obtained and the patient is placed on contact isolation. In an attempt to reduce the time from testing to results, molecular methods began to be used for detection of nasal MRSA colonization. Molecular methods utilize polymerase chain reaction (PCR) technology to detect and amplify regions of MRSA specific DNA. These tests can detect the presence of MRSA DNA within 2 hours and dramatically reduce the time to result in surveillance testing. In addition to the rapid response times, PCR based tests can also be used for quantification. The number of cycles the test completes before target DNA is detected (cycle threshold - C_t) is inversely proportional to the amount of DNA in the sample. The fewer the cycles necessary, the more target DNA is present.

In a previous study conducted by our group at the Atlanta VA Medical Center, we compared standard quantitative nasal cultures with the C_t of the Xpert MRSA assay on simultaneously collected nasal swabs. Using this method, we identified a strong linear correlation between the log of the colony forming units per/ml and the C_t, Pearson's correlation coefficient = -0.89(36). The relationship between quantitative cultures and the C_t from the Xpert MRSA assay is logarithmic and dichotomizes patients into high and low nasal MRSA burden at a C_t of 24.

The C_t from MRSA nasal surveillance swabs is a convenient and accurate measure of MRSA burden and may represent an accessible marker of quantification that can be used as a risk factor of subsequent MRSA infection. Using the C_t as a marker of MRSA nasal burden, we performed a retrospective cohort study to assess the affect of nasal MRSA burden on the risk of subsequent MRSA infection among veterans in Atlanta.

Methods

Study Design

A retrospective cohort study design was used to assess the effect MRSA nasal colonization burden has on subsequent MRSA infection while controlling for other risk factors associated with MRSA infection. The primary outcome variable was subsequent MRSA infection during four years of follow-up. MRSA nasal colonization burden was the main exposure variable and stratified on three levels: Negative colonization, low burden of colonization (CT > 24), and high burden of colonization (CT ≤ 24).

Study Setting

The study population consisted of US veterans who obtain medical care at the AVAMC. The AVAMC is a large, integrated healthcare system with approximately 200 inpatient beds, eight community-based outpatient clinics, and one nursing home care unit. Approximately 82,000 veterans receive care through the AVAMC and account for over 30,000 annual bed days of care at the acute care facility. All AVAMC medical facilities utilize the VA's computerized patient records system (CPRS) to access medical information. The AVAMC uses one central microbiology laboratory that receives specimens from the surrounding VA outpatient clinics, nursing home, and acute care facility. Most veterans at the AVAMC do not have private insurance coverage and rely solely on the VA for their medical needs.

Inclusion / Exclusion Criteria

All patients admitted to the AVAMC acute care medical facility from October 1st, 2007, through February 1st 2008 (4 months), were eligible to be included in the study population. All patients with positive admission nasal MRSA surveillance results were

included in the study. A random sample of patients from the same time period with negative admission nasal MRSA surveillance results was also included. Patients without admission nasal colonization results were excluded. Patients sent to the AVAMC for a surgical procedure with no prior or subsequent follow-up within the AVAMC system were also excluded. The inpatient psychiatric department does not perform MRSA nasal surveillance routinely and thus were excluded from the study population.

Colonization Data

In an attempt to reduce nosocomial MRSA transmission, the VHA issued a Directive in 2007 mandating the use of a MRSA bundle in all acute care settings. The MRSA bundle consists of active surveillance for MRSA nasal colonization in all patients admitted to the hospital, transferred between units, and upon discharge from the hospital; contact precautions for patients with (or a history of) MRSA colonization or infection; increased emphasis on hand hygiene; and a change in the institutional culture so infection control and prevention is the responsibility of all care providers. At the AVAMC, the MRSA bundle was fully implemented by October 1st, 2007, with greater than 90% compliance with admission surveillance swabs.

MRSA admission screening at the AVAMC is performed with one double swab (liquid Stuart Copan swabs; Cepheid) inserted 1 cm into each nasal vestibule and rotated 4 revolutions while maintaining even contact with the nasal mucosa. Nursing staff collects all nasal surveillance specimens within 24 hours of admission. All nursing staff has undergone training on appropriate collection techniques but no system is in place to ensure adequate collection techniques are utilized. Admission swabs are sent directly to the microbiology laboratory for testing using the Xpert MRSA assay. The Xpert MRSA

assay is performed according to the manufacturer's instructions with a C_t of 15 – 36 considered positive for MRSA. All C_t data is archived and easily extracted from the Xpert computer system. Extranasal sites are not routinely screened for MRSA and decolonization strategies are not routinely recommended for colonized patients.

Colonization results (positive or negative) from October 1st, 2007, through February 1st, 2008, were obtained from the Veterans Health Information Systems and Technology Architecture (VISTA) with the use of TheraDoc (Hospira, Lake Forest, IL), a web-based hospital surveillance system. The C_t for all positive admission screens was recorded. Patients with a $C_t > 24$ were considered to have low nasal MRSA burden and those ≤ 24 were considered to have high MRSA nasal burden.

MRSA Infection Data

MRSA infections were identified prospectively on a monthly basis by utilizing the microbiology option for specific organisms in VISTA. Surveillance of MRSA positive clinical cultures from all body sites began October 1st, 2005. All clinical MRSA cultures and corresponding clinical data (electronic medical record, anatomic site of culture, radiographic studies, laboratory results, and physician notes) were reviewed by the same experienced infectious disease physician on a monthly basis to identify true infections and exclude cultures representing colonization.

Infections were classified according to the Centers for Disease Control and Prevention criteria(37). Cultures not associated with a true infection were excluded. The infections were categorized according to primary site of infection into the following categories: skin and soft tissue, bone and joint, bloodstream, genitourinary, lower respiratory tract, surgical site, and other.

MRSA infections were classified into two mutually exclusive categories. Infections were considered hospital-onset if the clinical culture, from which MRSA was isolated, was obtained > 48 hours after admission to the AVAMC and not present on admission, all other infections were considered community–onset(38).

Identification of Covariates

The electronic medical record for each study participant was reviewed. Admission history and physical, discharge summaries, operative notes, last primary care note, progress notes within 30 days of admission, and pertinent laboratory values were reviewed. Demographic variables and covariates were recorded on a standard case report form. Problem lists were not used as a source of covariate information. External devices were considered anything foreign that entered the body and had an externalized segment (i.e. urinary catheter, central vascular access, suprapubic urinary catheter, tracheostomy, feeding tube). End stage renal disease (ESRD) included only those patients on renal replacement therapy. Patients not known to be human immunodeficiency virus (HIV) positive were assumed to be negative. Wounds were considered anything that caused the integrity of the skin to be compromised and included severe psoriasis, decubitus ulcers, chronic diabetic wounds, surgical wounds not yet healed, and burns. Chronic liver disease was defined as cirrhosis or chronic liver failure and did not include Hepatitis A, Hepatitis B, or Hepatitis C without liver failure of acute liver failure. Malignancies were considered active if the patient was actively being treated with chemotherapy, was under hospice care secondary to malignancy, had metastatic disease, or had active disease in which treatment was recommended. Localized prostate cancer was not considered as an active malignancy. All other co-morbidities were obtained from the medical record.

Sample Size Calculations

Estimated rates of subsequent MRSA infection were based on the available literature. For patients without nasal colonization it was estimated that 2.5% of the population would develop a MRSA infection during follow-up while 10% of those with low nasal colonization ($C_t > 24$) and 30% of those with high ($C_t \leq 24$) nasal colonization would subsequently develop an infection. Using a power of 80% and an alpha of 0.05, approximately 28 patients would be needed to show an effect when comparing high colonization to no colonization, 62 patients needed to compare high colonization to low colonization, and 150 patients comparing low colonization to no colonization. Approximately 50 patients per month are admitted to the AVAMC with MRSA nasal colonization. Of these patients, 30% have high nasal MRSA burden (≈ 15/month) and 70% have low nasal MRSA burden (≈ 35/month). Based on the above estimates and admission patterns at the AVAMC, it was estimated that four months worth of admission data would be needed to provide an adequate sample size.

Statistical Analysis

Descriptive statistics were used to compare the study population stratified by colonization status (negative, low burden, high burden). The purpose of this comparison was to identify potential factors that are associated with the exposure variable (colonization status). Differences in proportions of categorical variables (including demographic and clinical characteristics and co-morbidities) were tested using χ^2. If expected cell counts were less than 5, Fisher's exact test was utilized. Continuous variables were analyzed with a one-way analysis of variance to compare means or two-sample T test. A p-value of ≤ 0.05 was considered significant.

Unadjusted risk ratios were obtained for all covariates and the outcome in univariate analysis. Due to the limited published clinical data on nasal colonization burden, an exploratory analysis for potential interaction terms was performed. In this analysis, the study population was stratified on individual covariate levels and risk ratios for each level of the exposure variable (colonization status) were compared with the Breslow-Day test. A p-value ≤ 0.10 was considered significant in addition to biologically plausible interaction terms.

A multivariate logistic regression model was used to analyze the relationship between MRSA nasal colonization and subsequent infection. All covariates significant in univariate analysis ($P < 0.10$) and those considered clinically or epidemiologically relevant were evaluated in the initial model. Variables classified as collinear were not used in combination in the model. Model selection was based on a purposeful selection of covariates and not on statistical algorithms (backward, forward, or stepwise). Confounding was evaluated by assessing the effect covariates had on the parameter estimate for nasal colonization. Interaction terms with nasal colonization status were evaluated using the likelihood ratio test to compare models with and without interaction terms.

Data were analyzed using SAS software, version 9.3 (SAS Institute Inc., Cary, NC). Proc Genmod was used for model building and analysis assuming a binary distribution with the log link function. A Poisson distribution was used if models failed to converge.

IRB Approval and Funding Source

The Emory University Institutional Review Board and the VA Research and Development Committee approved this study. No external funding was used to conduct this study.

Results

Description of Cohort

From October 1st, 2007, to February 1st, 2008, 205 patients were admitted to the AVAMC with positive MRSA nasal colonization. Of those colonized, 141/205 (68.8%) had a $C_t > 24$ and 64/205 (31.2%) had a $C_t \leq 24$ (Figure 1). One hundred and fifty patients with negative MRSA nasal colonization were randomly selected during the same time period and nine patients were excluded due to lack of follow-up data (n = 141). The study cohort was predominately Caucasian (53.5%) or African American (43.9%) and the majority were male (95.1%). The mean age was 63.4 years old (standard deviation = 12.8). Comparing patients with MRSA nasal colonization to those without nasal colonization demonstrates many important differences (Table 1). Male sex, admission to a non-ICU unit, antibiotics received within 30 days, device present on admission, wound present on admission, and previous or concurrent MRSA infection were all more common in those colonized with MRSA.

During the four-years of follow-up, 43 subsequent MRSA infections were identified. Six MRSA infections occurred in those without MRSA nasal colonization (6/141, 4.3%), 26 MRSA infections occurred in those with low nasal MRSA burden (26/141, 18.5%), and 11 occurred in those with high nasal MRSA burden (11/64, 17.2%). The distribution of infection types and epidemiologic categories (hospital onset vs. community onset) was not significantly different in each colonization strata (negative, low burden, high burden) as is seen in Table 2. Time to subsequent MRSA infection did not differ between colonization categories ($P = 0.7979$). Death during the four years of follow-up was more common in patients with any MRSA nasal colonization (> 50%)

when compared to those without MRSA nasal colonization (34%). Patients with a high burden of nasal MRSA ($C_t \leq 24$) were the most likely to have persistent nasal carriage on readmission to the AVAMC (Table 2).

Characterization of Colonization Strata

In the cohort, 141 patients were negative for MRSA on admission, 141 had low MRSA nasal burden, and 64 had high MRSA nasal burden. A descriptive comparison of the three groups revealed significant differences between categories (Table 3). Patients not colonized were more likely to be admitted to the intensive care unit ($P = 0.0305$). Patients with either low or high nasal colonization were more likely to have a wound or device present on admission ($P < 0.0001$), a previous or concurrent MRSA infection ($P < 0.0001$), received an antibiotic within 30 days of admission ($P = 0.0463$), or were diagnosed with ESRD ($P = 0.0034$) or HIV ($P = 0.0494$) when compared to those without nasal colonization.

When comparing demographics and covariates between patients with low and high nasal colonization burden only, ESRD and having a device present on admission were more common in those with high burden ($P =0.0042$ and $P = 0.0036$ respectively) while patients with low burden were more likely to have three or more co-morbidities on admission ($P = 0.0640$). Interestingly, the proportion of patients that had a history of MRSA infection or a concurrent MRSA infection on admission did not differ between low and high nasal burden patients (18.4% vs. 21.9% respectively, $P = 0.5652$).

Multivariate Analysis

A univariate analysis was first performed to calculate unadjusted risk ratios for all potential risk factors of subsequent MRSA infection among patients admitted to the

AVAMC. The results are summarized in Table 4. Our main predictor variable, colonization status, was found to be associated with subsequent infection when comparing low burden to negative (RR 4.33, 95% CI 1.84 – 10.20) and high burden to negative (RR 4.04, 95% CI 1.56 – 10.44). Comparing patients with high burden to low burden was not significant in univariate analysis (RR = 0.93, 95% CI 0.49 – 1.76). The following factors were found to be significantly associated with subsequent MRSA infection and were included in the original model: Wounds present on admission (RR 2.56, 95% CI 1.46 – 4.46), device present on admission (RR 2.63, 95% CI 1.44 – 4.79), previous or concurrent MRSA infection (RR 2.25, 95% CI 1.20 – 4.22), hospital admission in the year prior to admission (RR 1.87, 95% CI 1.02 – 3.40), antibiotics within 30 days of admission (RR 2.14, 95% CI 1.23 – 3.73), diabetes (RR 1.69, 95% CI 0.96 – 2.96), and HIV infection (RR 2.43, 95% CI 1.00 – 5.85). No factors were identified on univariate analyses that were protective from subsequent MRSA infection.

A purposeful selection strategy including assessment of confounding and interaction was used to select the covariates for multivariate analysis. No evidence of interaction with colonization status was observed with the following interaction terms (significant Breslow-Day test or clinically relevant): Wounds, antibiotic use within 30 days, HIV, diabetes, ESRD, device present on admission, race, or previous/concurrent MRSA infection (Table 1, Appendix). The final model included colonization status, wounds, and device present on admission (Goodness-of-fit test, $p = 0.2960$). The rate ratios, 95% confidence intervals, and chi-square p-values for all variables included in the final multivariable analysis are listed in Table 5. MRSA colonization status was a significant risk factor for subsequent MRSA infection ($p = 0.0081$). After adjustment for

confounding variables, low colonization burden (compared to no colonization) was associated with increased risk of subsequent infection (RR = 3.62, 95% CI 1.47 – 8.93) while high colonization burden (compared to no colonization) did not reach the predefined level for significance (RR = 2.71, 95% CI 0.95 – 7.72, $P = 0.0615$). No variables were added to the final model solely as confounders and no interaction was detected.

Discussion

In this retrospective cohort study, we evaluate MRSA nasal colonization burden as a risk factor for subsequent MRSA infection using the C_t from the Xpert MRSA assay as a surrogate for colonization burden. MRSA nasal colonization was again found to be a risk factor for subsequent infection; however, patients with high MRSA nasal burden did not have an increased risk for subsequent infection as compared to those with low nasal burden. In addition to MRSA nasal colonization, the presence of wounds and invasive devices were also independent risk factors for the development of MRSA infections.

Colonization with *S aureus* is a well-known risk factor for subsequent staphylococcal infection and is the basis for many infection control interventions(2, 23). The literature on nasal colonization burden and its clinical effects is much less robust and based on studies from the 1960's(3, 30, 39). It is believed that a high nasal burden of *S. aureus* leads to a persistently colonized state and a high risk of subsequent staphylococcal disease. Studies assessing this relationship of staphylococcal burden and disease have mainly been conducted in countries with low levels of MRSA, included patients at very high risk of subsequent staphylococcal infection, and collected nasal culture specimens under strict study protocols(4, 31, 33). Our study is the first to assess an easily accessible quantification measure (C_t) obtained through routine infection control surveillance as a risk factor for subsequent MRSA disease among a population of veterans with significant MRSA infection rates(40, 41).

Our study did not find high colonization burden, as defined by the C_t of ≤ 24 from the Xpert MRSA assay, to be a significant risk factor for subsequent MRSA infection

when compared to low colonization burden. This unanticipated result can likely be attributed to one of number of factors. First, the two previous studies that have shown a higher infection rates among those with a high nasal burden of *S. aureus* only included high risk patients, peritoneal dialysis patients and post-surgical patients(4, 33). Our study uses both medical and surgical patients with the majority not having the high-risk features identified previously (wounds, external devices). Colonization burden may play a more significant role in patients with altered skin integrity than in a general patient mix that was used in our analysis.

Second, nasal swabs are collected on admission to the AVAMC by nursing staff throughout the hospital and not under strict study protocols. This collection, although intended to be standardized by the VHA Directive, is likely plagued by variable nasal swabbing technique due to different nurses collecting specimens throughout the hospital. For example, one patient had 9 admission or transfer nasal MRSA surveillance tests performed over the course of 6 months. Of these 9 tests, 4 had a $C_t \leq 24$ and 5 had a $C_t > 24$ without systemic antibiotics or nasal decolonization being prescribed. This variability is likely due to differing collection techniques used within the hospital and not actual changes in the nasal burden of MRSA. Due to this variability, our study likely suffers from measurement error and resultant misclassification bias. In a previous study, we validated the C_t as a measure of bacterial burden collected on nasal swabs in a sample of veterans; however, the C_t has not been validated as a measure of actual patient MRSA burden when collected during routine infection control surveillance. In order to effectively use the C_t as a measure of nasal MRSA burden, one must ensure strict collection techniques are followed to ensure reliability of results.

Third, our analysis did not use the C_t as a time dependent variable. We defined the C_t and the patients' colonization status based on one admission and one nasal swab. It is clear, as demonstrated above, the C_t is prone to variability and also colonization status is known to change, especially among intermittent carriers. Using the results of admission swabs on re-admission to the hospital (over 54% of the study population was readmitted during the 4 year follow up period – table 2) would likely add bias into the analysis as only patients needing re-hospitalization would be prone to retesting and thus reclassification. In order to adequately use the C_t as a time dependent variable, one must obtain repeat swabs on the entire study cohort and not just those needing re-hospitalization. Such a study would be ideal to define MRSA carriage status overtime and risk of subsequent infection but would require considerable financial and personnel support given the follow-up time and large sample size needed.

Our study has several unique strengths. Most importantly, all infections during the four years of follow-up were analyzed prospectively using a consistent clinical and microbiologic definition and reviewed by the same experienced infectious disease physician. In addition, most veterans do not have outside medical insurance coverage and do not seek care outside of the local VA system. Because of this, we are confident our study captured the majority of subsequent MRSA infections within out cohort and data obtained on co-morbidities and pre-admission characteristics were accurate and not subject to bias.

Our study does have several limitations. First, the AVAMC population of adult, primarily male, veterans living in the southeast US may not generalize to children, women, or other regions of the US(42-44). Second, our sample size calculations were

based on estimates of infection risks that were supported by minimal prior data. Due to our estimates, which included much higher rates of infection among those with high burden of colonization, our final model failed to demonstrate a statistically significant risk ratio when comparing high colonization burden to negative colonization ($P = 0.0615$). Even though it did not reach our predefined level of significance, the risk ratio and confidence intervals are very similar to that of low colonization burden and would likely reach significance if the sample size were increased. Also, colonization burden was dichotomized based on our previous work demonstrating a logarithmic relationship between C_t and quantitative cultures. Exploring the C_t as a continuous variable among colonized individuals would be a reasonable alternative as well.

In addition to better defining colonization as a risk factor, our study demonstrates the effect external devices (urinary catheters, central lines, tracheostomy, etc.) and wounds have on MRSA infections. Wounds and external devices both disrupt the natural barrier of the skin and allow a portal of entry for bacteria. Our findings add to the literature demonstrating similar findings in hospitalized patients(45-47).

The mortality rate seen in our study also highlights the complex patient population seen at the AVAMC. Interestingly, the death rate among MRSA colonized patients was considerably greater than those without nasal colonization (53% vs. 34%). This finding has not been well studied but is likely attributed to the increased level of co-morbidities found in those with MRSA nasal colonization. In our study, HIV and ESRD were both more common in patients colonized with MRSA while other studies have shown increased rates of CHF and COPD among colonized patients(45). Whether patients are more prone to MRSA colonization due to immune dysfunction related to

multiple co-morbidities and a reduction of antimicrobial peptides or due to more contact with the healthcare system due to their co-morbidities is yet to be fully elucidated.

Future Directions

Although the C_t as a quantitative measure of burden was not predictive of subsequent MRSA infection, its utility has yet to be fully defined and studied. A more thorough evaluation of nasal MRSA colonization burden is warranted with a focus on consistent and reliable nasal swabbing techniques. Future studies would include a case-control study evaluating the C_t as a risk factor for development of MRSA hospital acquired infections. A case-control study design would reduce the misclassification bias by studying infections that were temporally associated with a nasal colonization measurement. If a significant association is found between nasal colonization burden and hospital acquired infections, this may represent a population that would benefit from MRSA decolonization upon admission.

High nasal colonization burden has also been associated with colonization at other body sites(48) and increased rates of transmission to the surrounding environment(49, 50). Whether quantitative cultures or the C_t can be used in infection control measures to identify patients at high risk of transmission needs further study. Quantitative measures could potentially identify a subgroup of colonized patients that require contact isolation while in acute care settings instead of universal isolation of all MRSA carriers. Identifying patients at the highest risk of MRSA transmission could potentially lead to a more cost effect strategy of isolation and better patient care(51).

The relationship between MRSA strain type, colonization burden, and risk of subsequent infection has yet to be studied. The MRSA USA300 strain has been

associated with patients requiring hemodialysis and those with HIV infection(52). In our study, ESRD was associated with higher nasal burden of MRSA. A more comprehensive study evaluating strain types, virulence factors, patient demographics, and nasal burden is warranted.

References

1. Cosgrove SE, Sakoulas G, Perencevich EN, et al. Comparison of mortality associated with methicillin-resistant and methicillin-susceptible Staphylococcus aureus bacteremia: a meta-analysis. *Clin Infect Dis* 2003;36(1):53-9.
2. Wertheim HF, Melles DC, Vos MC, et al. The role of nasal carriage in Staphylococcus aureus infections. *Lancet Infect Dis* 2005;5(12):751-62.
3. White A. Increased Infection Rates in Heavy Nasal Carriers of Coagulase-Positive Staphylococci. *Antimicrob Agents Chemother (Bethesda)* 1963;161:667-70.
4. Nouwen JL, Fieren MW, Snijders S, et al. Persistent (not intermittent) nasal carriage of Staphylococcus aureus is the determinant of CPD-related infections. *Kidney Int* 2005;67(3):1084-92.
5. Shinefield HR, Ruff NL. Staphylococcal infections: a historical perspective. *Infect Dis Clin North Am* 2009;23(1):1-15.
6. Kirby WM. Extraction of a Highly Potent Penicillin Inactivator from Penicillin Resistant Staphylococci. *Science* 1944;99(2579):452-3.
7. Barber M, Rozwadowska-Dowzenko M. Infection by penicillin-resistant staphylococci. *Lancet* 1948;2(6530):641-4.
8. Jevons MP, Coe AW, Parker MT. Methicillin resistance in staphylococci. *Lancet* 1963;1(7287):904-7.
9. Berglund C, Soderquist B. The origin of a methicillin-resistant Staphylococcus aureus isolate at a neonatal ward in Sweden-possible horizontal transfer of a staphylococcal cassette chromosome mec between methicillin-resistant Staphylococcus haemolyticus and Staphylococcus aureus. *Clin Microbiol Infect* 2008;14(11):1048-56.
10. Archer GL, Niemeyer DM. Origin and evolution of DNA associated with resistance to methicillin in staphylococci. *Trends Microbiol* 1994;2(10):343-7.
11. Wielders CL, Vriens MR, Brisse S, et al. In-vivo transfer of mecA DNA to Staphylococcus aureus [corrected]. *Lancet* 2001;357(9269):1674-5.
12. Barrett FF, McGehee RF, Jr., Finland M. Methicillin-resistant Staphylococcus aureus at Boston City Hospital. Bacteriologic and epidemiologic observations. *N Engl J Med* 1968;279(9):441-8.
13. O'Toole RD, Drew WL, Dahlgren BJ, et al. An outbreak of methicillin-resistant Staphylococcus aureus infection. Observations in hospital and nursing home. *JAMA* 1970;213(2):257-63.
14. Chambers HF. The changing epidemiology of Staphylococcus aureus? *Emerg Infect Dis* 2001;7(2):178-82.
15. Panlilio AL, Culver DH, Gaynes RP, et al. Methicillin-resistant Staphylococcus aureus in U.S. hospitals, 1975-1991. *Infect Control Hosp Epidemiol* 1992;13(10):582-6.
16. Klevens RM, Edwards JR, Tenover FC, et al. Changes in the epidemiology of methicillin-resistant Staphylococcus aureus in intensive care units in US hospitals, 1992-2003. *Clin Infect Dis* 2006;42(3):389-91.

17. Four pediatric deaths from community-acquired methicillin-resistant Staphylococcus aureus - Minnesota and North Dakota, 1997-1999. *MMWR Morb Mortal Wkly Rep* 1999;48(32):707-10.
18. Klein E, Smith DL, Laxminarayan R. Community-associated methicillin-resistant Staphylococcus aureus in outpatients, United States, 1999-2006. *Emerg Infect Dis* 2009;15(12):1925-30.
19. Stranden AM, Frei R, Adler H, et al. Emergence of SCCmec type IV as the most common type of methicillin-resistant Staphylococcus aureus in a university hospital. *Infection* 2009;37(1):44-8.
20. Davis KA, Stewart JJ, Crouch HK, et al. Methicillin-resistant Staphylococcus aureus (MRSA) nares colonization at hospital admission and its effect on subsequent MRSA infection. *Clin Infect Dis* 2004;39(6):776-82.
21. Huang SS, Platt R. Risk of methicillin-resistant Staphylococcus aureus infection after previous infection or colonization. *Clin Infect Dis* 2003;36(3):281-5.
22. Gupta K, Strymish J, Abi-Haidar Y, et al. Preoperative nasal methicillin-resistant Staphylococcus aureus status, surgical prophylaxis, and risk-adjusted postoperative outcomes in veterans. *Infect Control Hosp Epidemiol* 2011;32(8):791-6.
23. Safdar N, Bradley EA. The risk of infection after nasal colonization with Staphylococcus aureus. *Am J Med* 2008;121(4):310-5.
24. von Eiff C, Becker K, Machka K, et al. Nasal carriage as a source of Staphylococcus aureus bacteremia. Study Group. *N Engl J Med* 2001;344(1):11-6.
25. Wertheim HF, Vos MC, Ott A, et al. Risk and outcome of nosocomial Staphylococcus aureus bacteraemia in nasal carriers versus non-carriers. *Lancet* 2004;364(9435):703-5.
26. Eriksen NH, Espersen F, Rosdahl VT, et al. Carriage of Staphylococcus aureus among 104 healthy persons during a 19-month period. *Epidemiol Infect* 1995;115(1):51-60.
27. VandenBergh MF, Yzerman EP, van Belkum A, et al. Follow-up of Staphylococcus aureus nasal carriage after 8 years: redefining the persistent carrier state. *J Clin Microbiol* 1999;37(10):3133-40.
28. Williams RE. Healthy carriage of Staphylococcus aureus: its prevalence and importance. *Bacteriol Rev* 1963;27:56-71.
29. Maxwell JG, Ford CR, Peterson DE, et al. Long-term study of nasal staphylococci among hospital personnel. *Am J Surg* 1969;118(6):849-54.
30. White A. Quantitative studies of nasal carriers of staphylococci among hospitalized patients. *J Clin Invest* 1961;40:23-30.
31. Nouwen JL, Ott A, Kluytmans-Vandenbergh MF, et al. Predicting the Staphylococcus aureus nasal carrier state: derivation and validation of a "culture rule". *Clin Infect Dis* 2004;39(6):806-11.
32. Stone ND, Lewis DR, Lowery HK, et al. Importance of bacterial burden among methicillin-resistant Staphylococcus aureus carriers in a long-term care facility. *Infect Control Hosp Epidemiol* 2008;29(2):143-8.

33. Kalmeijer MD, van Nieuwland-Bollen E, Bogaers-Hofman D, et al. Nasal carriage of Staphylococcus aureus is a major risk factor for surgical-site infections in orthopedic surgery. *Infect Control Hosp Epidemiol* 2000;21(5):319-23.
34. Muto CA, Jernigan JA, Ostrowsky BE, et al. SHEA guideline for preventing nosocomial transmission of multidrug-resistant strains of Staphylococcus aureus and enterococcus. *Infect Control Hosp Epidemiol* 2003;24(5):362-86.
35. Huskins WC, Huckabee CM, O'Grady NP, et al. Intervention to reduce transmission of resistant bacteria in intensive care. *N Engl J Med* 2011;364(15):1407-18.
36. Stenehjem E, Rimland D, Crispell EK, et al. Cephid Xpert MRSA Cycle Threshold in Discordant Colonization Results and as a Quantitative Measure of Nasal Colonization Burden. *J Clin Microbiol* 2012.
37. Horan TC, Andrus M, Dudeck MA. CDC/NHSN surveillance definition of health care-associated infection and criteria for specific types of infections in the acute care setting. *Am J Infect Control* 2008;36(5):309-32.
38. Klevens RM, Morrison MA, Nadle J, et al. Invasive methicillin-resistant Staphylococcus aureus infections in the United States. *JAMA* 2007;298(15):1763-71.
39. Solberg CO. A study of carriers of Staphylococcus aureus with special regard to quantitative bacterial estimations. *Acta Med Scand Suppl* 1965;436:1-96.
40. Caffrey AR, Laplante KL. Changing epidemiology of methicillin-resistant Staphylococcus aureus in the Veterans Affairs Healthcare System, 2002-2009. *Infection* 2011.
41. Tracy LA, Furuno JP, Harris AD, et al. Staphylococcus aureus infections in US veterans, Maryland, USA, 1999-2008. *Emerg Infect Dis* 2011;17(3):441-8.
42. Hudson LO, Murphy CR, Spratt BG, et al. Differences in methicillin-resistant Staphylococcus aureus (MRSA) Strains Isolated from Pediatric and Adult Patients from Hospitals in a Large California County. *J Clin Microbiol* 2011.
43. Liu C, Graber CJ, Karr M, et al. A population-based study of the incidence and molecular epidemiology of methicillin-resistant Staphylococcus aureus disease in San Francisco, 2004-2005. *Clin Infect Dis* 2008;46(11):1637-46.
44. Laupland KB, Church DL, Mucenski M, et al. Population-based study of the epidemiology of and the risk factors for invasive Staphylococcus aureus infections. *J Infect Dis* 2003;187(9):1452-9.
45. Honda H, Krauss MJ, Coopersmith CM, et al. Staphylococcus aureus nasal colonization and subsequent infection in intensive care unit patients: does methicillin resistance matter? *Infect Control Hosp Epidemiol* 2010;31(6):584-91.
46. Coello R, Glynn JR, Gaspar C, et al. Risk factors for developing clinical infection with methicillin-resistant Staphylococcus aureus (MRSA) amongst hospital patients initially only colonized with MRSA. *J Hosp Infect* 1997;37(1):39-46.
47. Harinstein L, Schafer J, D'Amico F. Risk factors associated with the conversion of meticillin-resistant Staphylococcus aureus colonisation to healthcare-associated infection. *J Hosp Infect* 2011;79(3):194-7.

48. Mermel LA, Cartony JM, Covington P, et al. Methicillin-resistant Staphylococcus aureus colonization at different body sites: a prospective, quantitative analysis. *J Clin Microbiol* 2011;49(3):1119-21.
49. Ehrenkranz NJ. Person-to-Person Transmission of Staphylococcus Aureus. Quantitative Characterization of Nasal Carriers Spreading Infection. *N Engl J Med* 1964;271:225-30.
50. White A. Relation between quantitative nasal cultures and dissemination of staphylococci. *J Lab Clin Med* 1961;58:273-7.
51. Stelfox HT, Bates DW, Redelmeier DA. Safety of patients isolated for infection control. *JAMA* 2003;290(14):1899-905.
52. Mermel LA, Eells SJ, Acharya MK, et al. Quantitative analysis and molecular fingerprinting of methicillin-resistant Staphylococcus aureus nasal colonization in different patient populations: a prospective, multicenter study. *Infect Control Hosp Epidemiol* 2010;31(6):592-7.

Tables and Figures

Figure 1. Distribution of the cycle threshold (CT) of positive admission nasal MRSA screens (Xpert MRSA assay) among Atlanta veterans (n = 205)

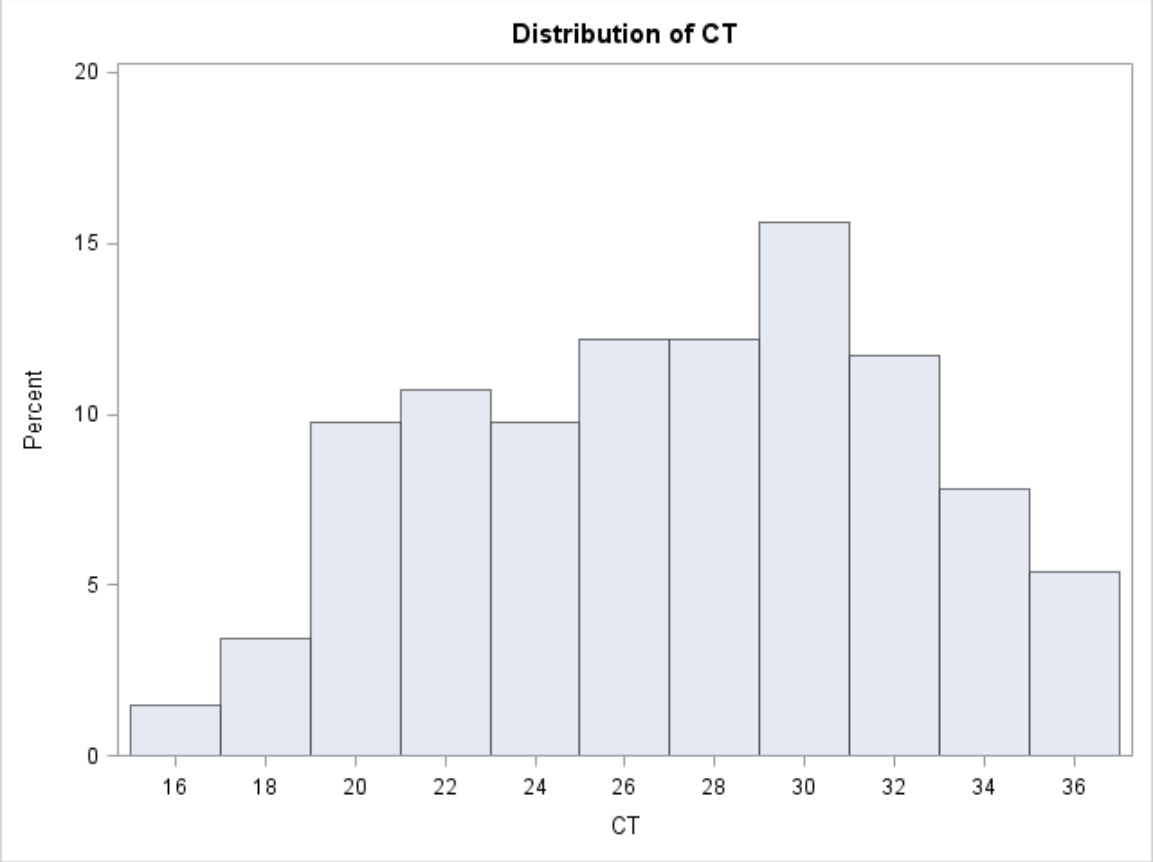

Table 1: Baseline patient characteristics among MRSA colonized and non-colonized (N = 346)

	Nasal Colonization Status		
Patient Demographics	**Negative**	**Positive**	***P* - value[a]**
	N (%)	N (%)	
Total	141	205	
CT	n/a	27.1	
Age (years)			
Mean (SD)	63.0 (13.3)	63.6 (12.4)	0.6876[b]
> 71.7 (4th quartile)	39 (27.7)	51 (24.9)	0.5622
Gender			
Male	130 (92.2)	199 (97.1)	0.0393
Female	11 (7.8)	6 (2.9)	
Race			
Black	55 (39.0)	97 (47.3)	0.1571
White	81 (57.4)	104 (50.7)	
Other	5 (3.6)	4 (2.0)	
Clinical Characteristics			
Admit from other than home	11 (7.8)	29 (14.2)	0.0697
Admission to ICU	44 (31.2)	40 (19.5)	0.0127
Surgery in 12 months prior	35 (24.8)	63 (30.7)	0.2307
Admission in 12 months prior	67 (47.5)	115 (56.1)	0.1163
Antibiotics within 30 days	26 (18.4)	61 (29.8)	0.0171
Co-morbidities			
Wound present	10 (7.1)	55 (26.8)	<0.0001
Device Present	8 (5.7)	32 (15.6)	0.0045
Previous / Concurrent MRSA	1 (0.7)	40 (19.5)	<0.0001
CAD	55 (39.0)	68 (33.2)	0.2651
CHF	34 (24.1)	62 (30.2)	0.2108
PVD	16 (11.4)	38 (18.5)	0.0702
COPD	26 (18.4)	55 (26.8)	0.0702
DM	60 (42.6)	88 (42.9)	0.9450
Smoker	45 (31.9)	52 (25.4)	0.1826
Advanced Liver dz	5 (3.6)	15 (7.3)	0.1650
Active Malignancy	31 (22.0)	28 (13.6)	0.0430
ESRD	6 (4.3)	13 (6.3)	0.4026
CVA	18 (12.8)	38 (18.5)	0.1521
HIV	2 (1.4)	12 (5.9)	0.0507
Other	11 (7.8)	25 (12.2)	0.1884
≥ 3 co-morbidities	58 (41.1)	90 (43.9)	0.6091

*CAD = coronary artery disease, CHF = congestive heart failure, PVD = peripheral vascular disease, COPD = chronic obstructive pulmonary disease, DM = diabetes mellitus, ESRD = end stage renal disease, CVA = cerebrovascular accident, HIV = human immunodeficiency virus.

[a] *P*-value for Chi Square or Fisher's exact test

[b] *P*-value for two-sample T test

Table 2. A comparison of subsequent MRSA infection types, death during follow-up, and readmission within 4 years stratified by colonization status among a cohort of Atlanta veterans

Characteristic	Negative Colonization (N = 141)	Low Colonization Burden (N = 141)	High Colonization Burden (N = 64)	*P*-value [a]
Total Subsequent Infections	6 (4.3%)	26 (18.5%)	11 (17.2%)	0.0007
Subsequent Infection				
Skin/Soft Tissue	2	10	4	0.2040
Bone and Joint		4		
Primary Bloodstream		7	1	
Genitourinary	2	2	4	
Lower Respiratory	2	2	2	
Surgical Site		1		
Mean Time to Infection in Days (SD)	310.8 (283.9)	385.8 (398.4)	445.6 (444.9)	0.7979[b]
Death				
Death during admission	7 (5.0%)	9 (6.4%)	4 (6.3%)	0.8683
Death during follow up	48 (34.0%)	73 (51.8%)	35 (54.7%)	0.0026
Readmission during 4 years of follow up				
≥ 1 readmission	77 (54.6%)	82 (58.2%)	41 (64.1%)	0.4438
Admission nasal swab positive on readmission	7 (9.5%)	48 (59.3%)	28 (70.0%)	<0.0001

[a] *P*-value for Chi Square Test or Fisher Exact Test
[b] *P*-value for one-way analysis of variance, F test

Table 3. Baseline patient characteristics among patients without MRSA colonization, low MRSA colonization burden, and high MRSA colonization burden (N = 346)

	Colonization Status			
Patient Demographics	**Negative**	**Low Burden**	**High Burden**	***P* – value[a]**
	N (%)	**N (%)**	**N (%)**	
Total	141	141	64	
CT	n/a	29.8	21.0	
Age (years)				
Mean (SD)	63.0 (13.3)	62.7 (12.5)	65. 5 (12.3)	0.3411[b]
> 71.7 (4th quartile)	39 (27.7)	33 (23.4)	18 (28.1)	0.6552
Gender				
Male	130 (92.2)	136 (96.5)	63 (98.4)	0.0993
Female	11 (7.8)	5 (3.5)	1 (1.8)	
Race				
Black	55 (39.0)	67 (47.5)	30 (46.9)	0.3544
White	81 (57.4)	70 (49.6)	34 (53.1)	
Other	5 (3.6)	4 (2.9)	0 (0)	
Clinical Characteristics				
Admit from other than home	11 (7.8)	17 (12.1)	12 (18.8)	0.0736
Admission to ICU	44 (31.2)	30 (21.3)	10 (15.6)	0.0305
Surgery in 12 months prior	35 (24.8)	41 (29.1)	22 (34.4)	0.3597
Admission in 12 months prior	67 (47.5)	75 (53.2)	40 (62.5)	0.1356
Antibiotics within 30 days	26 (18.4)	40 (28.4)	21 (32.8)	0.0463
Co-morbidities				
Wound Present	10 (7.1)	34 (24.1)	21 (32.8)	<0.0001
Device Present	8 (5.7)	15 (10.6)	17 (26.7)	<0.0001
Previous / Concurrent MRSA	1 (0.7)	26 (18.4)	14 (21.9)	<0.0001
CAD	55 (39.0)	44 (31.2)	24 (37.5)	0.3673
CHF	34 (24.1)	43 (30.1)	19 (29.7)	0.4537
PVD	16 (11.4)	23 (16.3)	15 (23.4)	0.0831
COPD	26 (18.4)	39 (27.7)	16 (25.0)	0.1779
DM	60 (42.6)	60 (42.6)	28 (43.8)	0.9849
Smoker	45 (31.9)	40 (28.4)	12 (18.8)	0.1500
Advanced Liver Disease	5 (3.6)	12 (8.5)	3 (4.7)	0.1861
Active Malignancy	31 (22.0)	21 (14.9)	7 (10.9)	0.1011
ESRD	6 (4.3)	4 (2.9)	9 (14.1)	0.0034
CVA	18 (12.8)	26 (18.4)	12 (18.8)	0.3581
HIV	2 (1.4)	10 (7.1)	2 (3.1)	0.0494
Other	11 (7.8)	18 (12.8)	7 (10.9)	0.3891
≥ 3 co-morbidities	58 (41.1)	68 (48.2)	22 (34.4)	0.1563

*CAD = coronary artery disease, CHF = congestive heart failure, PVD = peripheral vascular disease, COPD = chronic obstructive pulmonary disease, DM = diabetes mellitus, ESRD = end stage renal disease, CVA = cerebrovascular accident, HIV = human immunodeficiency virus.

[a] *P*-value for Chi Square or Fisher's exact test

[b] *P*-value for one-way analysis of variance, F test

Table 4. Univariate analysis of potential risk factors for subsequent MRSA infection in veterans (n=346)

Potential Risk Factors	Risk Ratio[a]	95% CI	*P*-value[b] (If ≤0.10)
Male vs. Female	2.17	0.31 – 14.83	-
Black vs. White	1.40	0.79 – 2.44	-
Age: 4th quartile vs. other	0.75	0.37 – 1.50	-
Clinical Characteristics			
Admit from other than home	1.24	0.55 – 2.75	-
Admission to ICU vs. Floor	0.61	0.28 – 1.31	-
Surgery in 12 months prior	1.65	0.94 – 2.91	-
Admission in 12 months prior	1.87	1.02 – 3.40	0.0376
Antibiotics within 30 days	2.14	1.23 – 3.73	0.0070
Co-morbidities			-
Low vs. Negative Nasal Burden	4.33	1.84 – 10.20	0.0008
High vs. Negative Nasal Burden	4.04	1.56 – 10.44	0.0040
High vs. Low Nasal Burden	0.93	0.49 – 1.76	-
Wound Present	2.56	1.46 – 4.46	0.0010
Device Present	2.63	1.44 – 4.79	0.0022
Previous / Concurrent MRSA	2.25	1.20 – 4.22	0.0135
CAD	0.70	0.37 – 1.31	-
CHF	0.90	0.47 – 1.70	-
PVD	1.23	0.60 – 2.51	-
COPD	1.12	0.59 – 2.12	-
DM	1.69	0.96 – 2.96	0.0652
Smoker	1.11	0.60 – 2.04	-
Advanced Liver Disease	1.22	0.41 – 3.61	-
Active Malignancy	0.64	0.26 – 1.55	-
ESRD	1.77	0.70 – 4.42	-
CVA	1.57	0.82 – 2.99	-
HIV	2.43	1.00 – 5.85	0.0620
Other	1.67	0.80 – 3.48	-
≥ 3 co-morbidities	1.40	0.80 – 2.45	-

[a] Unadjusted risk ratios

[b] *P*-value for Chi Square Test

Table 5. Multivariate Analysis of predictors of subsequent MRSA infection among veterans

Parameters	Rate Ratio[a]	95% CI	*P*-value[b]
Low vs. Negative Nasal Colonization Burden	3.62	1.47 – 8.93	0.0052
High vs. Negative Nasal Colonization Burden	2.71	0.95 – 7.72	0.0615
High vs. Low Nasal Colonization Burden	0.75	0.36 – 1.55	0.4378
Wounds	1.86	0.98 – 3.52	0.0559
Device Present on Admission	2.12	1.04 – 4.32	0.0393

[a] Adjusted rate ratios

[b] *P*-value for Chi Square Test

Appendix

Table 1. Testing for statistical significance of interaction terms between colonization status and selected covariates

Interaction	Variables	*P* – value[a]
Colonization status	Wounds	0.0734
	Antibiotic use 30 days prior	0.1044
	HIV	0.1650
	Diabetes	0.0901
	ESRD	0.2202
	Device Present on Admit	0.2651
	Race	0.0756
	Previous/Concurrent MRSA	0.1761

[a]*P*-value for chi-square test (likelihood ratio test for interaction terms)