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Interplay of Personal, Pet, and Environmental Colonization in Households Affected by Community-Associated Methicillin-Resistant Staphylococcus aureus

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Abstract

Objective—We sought to determine the prevalence, molecular epidemiology, and factors associated with Staphylococcus aureus environmental surface and pet colonization in households of children with community-associated methicillin-resistant S. aureus (CA-MRSA) infection.

Methods—Between 2012 and 2015, 150 children with CA-MRSA infections and their household contacts and pets were enrolled in this cross-sectional study in metropolitan Saint Louis, MO. Cultures to detect S. aureus were collected from 3 anatomic sites of household members, $2 \log \alpha t$ sites, and 21 environmental surfaces in each household. Molecular epidemiology of S. aureus isolates was determined via repetitive-sequence PCR. Generalized linear models were developed to identify factors associated with S. aureus/MRSA household contamination.

CONFLICTS OF INTEREST

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Results—MRSA was recovered from environmental surfaces in 69 (46%) households (median 2 surfaces $[range 1–18]$). The enrollment infecting strain type was the most common strain recovered from the environment in most (64%) households. In generalized linear models, factors associated with a higher proportion of MRSA-contaminated environmental surfaces were household member MRSA colonization burden, MRSA as the dominant S. aureus strain colonizing household members, more strain types per household member, index case African-American race, and renting (vs. owning) the home. Of 132 pets, 14% were colonized with MRSA. Pets whose primary caretaker was MRSA-colonized were more likely to be MRSA-colonized than pets whose primary caretaker was not MRSA-colonized (50% vs. 4%, p<0.001).

Conclusions—Household environments and pet dogs and cats serve as reservoirs of MRSA. Household member MRSA colonization burden predicts environmental MRSA contamination. Longitudinal studies will inform the directionality of household transmission.

Keywords

community-associated methicillin-resistant Staphylococcus aureus; household reservoirs; environmental contamination; pets

> Community-associated methicillin-resistant Staphylococcus aureus (CA-MRSA) is an important cause of skin and soft tissue infection (SSTI) in children [1]. Incomplete understanding of the epidemiology of carriage and transmission of these strains has contributed to their widespread dissemination in communities and, recently, into hospitals, where healthcare-associated (HA)-MRSA strains once predominated [2, 3]. While preventive measures in hospitalized patients have curtailed the incidence of HA-MRSA infections [4, 5], similar strides have not been achieved in the community setting. Indeed, after performing decolonization, up to 50% of SSTI patients develop recurrent infections over the ensuing year [6].

CA-MRSA is a disease of households, facilitated by complex transmission dynamics yet to be fully understood [2, 7]. Dogma holds that S. aureus colonization poses risk for recurrent SSTI and spread among household contacts [8]. Instances of discordance between infecting S. aureus strains and those colonizing index cases and their household contacts [9, 10], and varied effectiveness of decolonization trials [6, 11], indicate that exclusive focus on human colonization may be inadequate. The household environment is increasingly recognized as a community reservoir of S. aureus. Indeed, household MRSA environmental contamination is significantly more prevalent in homes of MRSA-infected patients than in control households [12, 13]. Additionally, household companion animals can carry *S. aureus* and may contribute to *S. aureus* household transmission $[7, 14]$. Studies to date $[9, 12, 13, 15, 16]$ have been limited by examining only a subset of household members, discounting socioeconomic status, sampling a small number of surfaces, or disregarding pet colonization.

Understanding these complex community MRSA reservoirs is imperative to mitigate transmission. The objectives of this study were to determine the prevalence, molecular epidemiology, and factors associated with S. aureus, and specifically MRSA, environmental surface contamination and pet carriage in households of children with CA-MRSA infection.

METHODS

Participants

Between January 2012 and October 2015, 377 children with SSTI were screened for the HOME: Household Observation of MRSA in the Environment study. Screening took place at St. Louis Children's Hospital (SLCH), Cardinal Glennon Children's Hospital, and community pediatric practices affiliated with the Washington University Pediatric and Adolescent Ambulatory Research Consortium. Children with HA-MRSA infections [17] (e.g., recent hospitalization, invasive medical device, residing in long-term care facility) were excluded from screening. One hundred thirty subjects did not meet eligibility criteria (e.g., SSTI not verified as MRSA, time interval from infection to screening, distance from medical center). Of 247 eligible patients with CA-MRSA infection, 150 index cases (149 with SSTI, 1 invasive infection) were enrolled, along with their household contacts (individuals sleeping in the home ≥4 nights/week) and indoor pet dogs and cats. The Washington University Institutional Review Board and Institutional Animal Care and Use Committee approved study procedures. Written, informed consent/assent was obtained for all household members (by participant and/or guardian) and pets (by primary caretaker).

Data collection

An enrollment visit was conducted in the index case's primary home. Participants were queried about demographics, socioeconomic surrogates (e.g., insurance status, home ownership, household crowding), prior S. aureus infections, personal hygiene practices, activities, pet characteristics, and household layout and cleaning practices. To control for potential bias introduced by participants misrepresenting their cleaning habits, the research team assigned each household an objective 'home cleanliness score' from 1 (above average) to 4 (very dirty), which considered odor, clutter, and grime, modified from the Environmental Cleanliness and Clutter Scale [18].

Index case infecting isolates were obtained from the clinical microbiology laboratory when available. At enrollment, colonization cultures were collected by trained study personnel from the anterior nares, axillae, and inguinal folds of each household member (Eswab, Becton Dickinson [BD], Franklin Lakes, NJ) and from the nares (minitip Eswab, BD) and dorsal fur (Eswab) of indoor pet dogs and cats. Twenty-one environmental surfaces [19] were sampled (Table 1); standardized environmental sampling employed the Baird Parker Agar contact plate (Hardy, Santa Maria, CA) and the Eswab [19, 20].

Laboratory procedures

From Eswabs, S. aureus was recovered using broth-enrichment. From the contact plate, colonies were selected based on morphology. S. aureus identification and antibiotic susceptibility testing were performed in accordance with established techniques [21] (Supplementary Methods). Molecular typing was performed on all recovered S. aureus isolates by repetitive-sequence PCR (repPCR) [22, 23]. Isolates with unusual repPCR patterns and all isolates recovered from pets were confirmed as S. aureus by matrix-assisted laser desorption ionization time-of-flight mass spectrometry (VITEK MS v2.0) [24].

Statistical analysis

(*numbero f S* . *aureus*(*MRSA*)*anatomicalsitescolonized*) (*numbero f sampledhouseholdmembers*) * (*numbero f sampledanatomicsites*)

To compare the proportion of household environmental surfaces contaminated with S. aureus or MRSA, as well as the number of unique strains (by repPCR) recovered from households, across various attributes, nonparametric Mann-Whitney U and Kruskal-Wallace tests were performed. The relationship between environmental surface contamination prevalence and household colonization pressure was determined by linear regression. Pearson's χ^2 tests compared pet carriage with primary caretaker colonization and various pet factors. Data analyses were performed with SPSS 23 for Windows (IBM SPSS, Chicago, IL).

Generalized linear Poisson models were developed (in R language [25]; Supplementary Methods) in a Bayesian framework to define the impact of household member colonization pressure and individual and household-level cleanliness variables on the proportion of household environmental surfaces contaminated with S. aureus or MRSA. The 'Personal Colonization and Pet Carriage' model considered how colonization status and strain distribution of household members and pets and socioeconomic factors influence household contamination. The 'Household Practices and Personal Behaviors' model examined how hygiene and epidemiological factors influence household contamination. See Supplementary Tables 1 and 2 for primary and secondary covariates included.

RESULTS

Study population and S. aureus colonization prevalence

The 150 index cases (53% female) had 546 household contacts, of which 521 (52% female) enrolled (96% participation). The median age of index cases and household contacts was 3 years (range 0.1–18) and 27 years (range 0.1–82), respectively. Index cases were primarily Caucasian (68%) or African-American (25%); 6% were of Hispanic/Latino ethnicity. The majority of participants lived in houses (81%, vs. apartments/townhomes) that they owned (66%, vs. rented) in urbanized areas (87%, vs. urban clusters or rural areas) [26, 27]. Median household size was 4 (range 2–13). Enrollment visits were conducted a median of 20 days (range 3–95) following MRSA infection. The study area encompassed $4,035$ miles² (household distance from SLCH: median 17 miles, range 1–76). Fifty-seven (38%) index cases and 218 (43%) household contacts were colonized with S. aureus at enrollment (30% and 23% with MRSA, respectively). Sixty-two (41%) index cases and 87 (17%) household contacts had a history of confirmed S. aureus infection (excluding the infection prompting study enrollment); 59% and 27%, respectively, reported an SSTI (of any etiology) in the year prior to enrollment.

Prevalence of S. aureus environmental contamination and pet colonization

S. aureus was recovered from $\frac{1}{2}$ environmental surface in 95 (63%) of 150 households (median 3 surfaces [range 1–18]); MRSA was recovered from surfaces in 69 (46%) households (median 2 surfaces [range 1–18]) (Table 1). Of households with *S. aureus*

contamination, 39 (41%) were contaminated exclusively with MRSA, 26 (27%) exclusively with methicillin-susceptible S. aureus (MSSA), and 30 (32%) with both.

The most common sites of environmental MRSA contamination were the index case's bed linens (15%), television remote control (15%), kitchen table (12%), computer keyboard/ mouse (12%), bathroom countertop (11%), and refrigerator door handle (11%) (Table 1). Surfaces that were less frequently MRSA-contaminated overall (index case bath towel, kitchen sink faucet handle or hand towel, and bathtub soap bar/dish) were more likely to be contaminated in households with a higher proportion of MRSA-contaminated environmental surfaces (Figure 1).

Pet dogs or cats were present in 81 (54%) households (median 2 [range 1–9] pets per household). Overall, 100 dogs and 32 cats from 71 households were swabbed; 16 (16%) dogs and 3 (9%) cats carried MRSA (Table 1). While 50% of pets whose primary caretaker (i.e., the human primarily responsible for their feeding, exercising, and waste cleanup) was MRSA-colonized also carried MRSA, just 4% of pets whose primary caretaker was not MRSA-colonized carried MRSA ($p<0.001$); for 3 of the 4 pets who carried MRSA without an MRSA-colonized caretaker, an alternate household member was colonized. Pets with MRSA carriage lived in households with higher household member MRSA colonization pressure (median 0.50, interquartile range [IQR] 0.25–0.75) than pets without carriage (median 0.08, IQR 0.00–0.27; p<0.001). No relationship was observed between pet MRSA carriage and pet daycare/boarding attendance, sleeping with colonized household members, overall health, or infection history.

Factors associated with environmental contamination

In univariate analyses, a higher proportion of MRSA-contaminated environmental surfaces was associated with the index case being African-American or multiracial (vs. Caucasian, p=0.01), renting the home (vs. owning, p=0.001), index case MRSA colonization (p<0.001), and any household member MRSA colonization $(p<0.001)$ (Supplementary Table 3). In addition to the covariates significantly associated with a higher proportion of MRSAcontaminated environmental surfaces, a higher proportion of S. aureus-contaminated environmental surfaces was associated with Medicaid or no health insurance (vs. private or Tricare, $p=0.03$) and presence of more unique *S. aureus* strains in the household ($p<0.001$). Neither prior S. aureus infections nor past-year SSTIs in household members were associated with S. aureus or MRSA surface contamination.

Predicting environmental contamination: multivariable models

Personal Colonization and Pet Carriage Model—Covariates significantly predictive of a higher proportion of MRSA-contaminated environmental surfaces included increased household member MRSA colonization pressure, MRSA as the dominant S. aureus strain colonizing household members, increased number of unique S . aureus strains (by repPCR) per household member, and renting (vs. owning) the home (Table 2).

Household Practices and Personal Behaviors Model—Given the strong influence of personal colonization on environmental contamination in the 'Personal Colonization and

Pet Carriage' model, we developed models that excluded human colonization and pet carriage to discern the influence of personal behaviors on household environmental contamination. Renting (vs. owning) the home and index case African-American (or multiracial) race (vs. Caucasian) were predictive of a higher proportion of MRSAcontaminated environmental surfaces (Table 2, Supplementary Figure 1).

Molecular epidemiology of S. aureus strains in households

Overall (including index cases, household contacts, environmental surfaces, and pets), up to 7 (median 2) unique strains (by repPCR) were recovered from a single household. Up to 4 strains (median 1) were recovered from the environmental surfaces of a single household. Crowded households (>2 people per bedroom) harbored more strains overall (median 2, interquartile range $[IQR] 2–4$) than non-crowded households (median 2, IQR 1–2; p=0.004). Households assigned a home cleanliness score of below average or very dirty harbored more strains (median 2, IQR 1–3) than average or above average households (median 1, IQR 1–2; $p=0.01$). Households with more strains also had a higher proportion of S. aureuscontaminated environmental surfaces $(p=0.001,$ Supplementary Table 3, Supplementary Figure 1).

The MRSA isolate from the enrollment infection was available for 91 (61%) index cases, and a concordant strain was recovered from an environmental surface in 44 (48%) of these households. The enrollment infecting strain was the most common strain recovered in 37 (64%) of 58 MRSA-contaminated household environments. An environmental surface was contaminated with a S. aureus strain concordant with an index case colonizing or infecting strain in 60 (52%) of 116 households with an available index case strain (Supplementary Figure 2A); an environmental surface was contaminated with a household contactconcordant strain in 68 (45%) of 150 households. Strains discordant with index case strains were recovered from the environment in 18 (23%) of 78 households with both an available index case strain and environmental contamination (Supplementary Figure 2B); only 4 (5%) of these 78 household environments yielded a strain not recovered from a human household member. Among household contacts, siblings of index cases were the most likely to be colonized with a strain recovered from an environmental surface (31%), while adults other than the index case's parents were least likely to be colonized with environmental strains $(11\%, p=0.03)$.

Overall, the index case's bed linens, TV remote control, and refrigerator door handle were most often colonized with a S. aureus strain concordant with an index case strain (Supplementary Figure 2A), reflective of environmental sites with high colonization prevalence (Table 1). Focusing on contaminated surfaces, in households with an available index case strain, the bathroom soap bar/dish, bathroom hand towel, and bathroom door handle were most often colonized with a strain concordant with an index case strain (Supplementary Figure 2B). Certain high-touch surfaces shared by all household members, such as the toilet handle, kitchen hand towel, and videogame controller were more often colonized with a household contact strain than an index case strain (Supplementary Figure 2B).

Of 130 swabbed pets with a swabbed primary caretaker, 104 (80%) did not carry S. aureus. Sixteen (12%) pets carried a S. aureus strain concordant with a primary caretaker strain, while 4 (3%) pets carried a strain discordant from primary caretaker strains; the primary caretaker was not colonized for 6 (5%) pets carrying S. aureus (Figure 2). Overall, 6 (5%) of 132 swabbed pets carried a distinct strain not recovered from a human household member. Likelihood of strain concordance did not differ between dogs and cats or between pets who did and did not sleep with a household member. In 14 households with both environmental contamination and pet carriage, 13 (93%) displayed strain concordance between these sites.

DISCUSSION

In the present study characterizing households of children with MRSA SSTI, nearly half of homes were found to harbor environmental MRSA contamination. In addition to the index case's bed linens, commonly touched and shared surfaces (TV remote control, computer keyboard/mouse, and kitchen table) were the fomites most likely to be MRSA-contaminated. In most households, the index case infecting strain was the predominant environmental strain. Moreover, while >20% of households with pets had dogs and/or cats with MRSA carriage, such carriage rarely occurred in the absence of a MRSA-colonized household member. While the cross-sectional nature of our study precludes defining directionality of transmission, our data support the premise that infected/colonized household members contaminate their home environment and subsequently, environmental reservoirs perpetuate the cycle of reacquisition and transmission.

The prevalence of household environmental MRSA contamination (46%) was higher in the present study compared to two prior US studies in homes of patients with MRSA SSTI. Studies conducted in Chicago/Los Angeles and New York recovered MRSA from environmental surfaces in 27% and 30% of homes, respectively [12, 28]. Our higher measured prevalence of environmental contamination may reflect inclusion of more surfaces (21 vs. ~8–11) and solely pediatric index cases (vs. households without children), who may be in closer contact with their environment; indeed, strains recovered from children had the highest concordance with environmental strains. While several surfaces with high MRSA prevalence (e.g., TV remote control, refrigerator door handle, computer, index case bed linens) were sampled across all studies, the present study identified other frequently contaminated sites (e.g., kitchen table, bathroom countertop) that represent potential targets for intervention.

It has been demonstrated that occupants leave an identifiable bacterial signature on household surfaces. Lax and colleagues monitored household microbial communities before and after a physical move; bacterial communities of participants' new homes promptly converged on the communities of their original homes, implicating household members as the vector [29]. Collectively, our predictive models indicate that household member colonization pressure is the most significant predictor of the burden of MRSA environmental contamination. In the majority of households in our study, the index case infecting strain was the predominant environmental strain; in prior studies, environmental contamination with clinical S. aureus strains has been associated with household transmission, reacquisition, and recurrent infection [12, 28, 30]. In total, the available data indicate that household

The present study of household MRSA contamination is strengthened by the sampling of companion animals. While *S. aureus* has been recovered from pets, such carriage can resolve without antimicrobial treatment, suggesting that pets are not a natural *S. aureus* reservoir [7]. A study in the United Kingdom sequenced genomes of 46 isolates from MRSA-infected pets, all of which clustered with human colonizing strains from unrelated patients, representing the circulating epidemic clade [32]. These data suggest S. aureus transmission from human to pet, though the study did not include colonizing isolates of human household contacts. In the present study of households of children with MRSA SSTI, more than 20% of sampled companion animals carried S. aureus (14% carried MRSA). Importantly, pets were more likely to carry MRSA if their primary caretaker was MRSA-colonized and if household member MRSA colonization pressure was higher. Additionally, pets almost exclusively carried strains concordant with both the index case infecting strain and strains found on environmental surfaces. Collectively, these studies indicate that while pets may participate in household S. aureus transmission, humans represent the primary source of S. aureus in pets.

Compared to other recent studies [9, 12], the present work systematically sampled a much higher proportion of household members and more environmental surfaces, and included companion animals, to provide the most comprehensive available picture of household S. aureus dynamics. Our conclusions are based on substantial epidemiologic data and advanced statistical modeling techniques. We quantified the proportion of contaminated surfaces rather than recording only presence or absence of environmental S. aureus, though we did not measure the density of S. aureus at each surface. As the primary objective of this study was to identify household reservoirs of MRSA to inform interventions to interrupt MRSA transmission and prevent recurrent infections, we only sampled households of children with MRSA SSTI. Strain concordance between the MRSA enrollment infection isolate and other recovered isolates could only be evaluated in the 91 households where it was available. A further limitation of the study is that due to its cross-sectional nature, directionality of transmission among humans, pets, and the environment were not definitively established. Finally, while whole-genome sequencing would provide the highest strain resolution, repPCR has been shown to be highly discriminatory, especially in the household context [23].

In this comprehensive investigation of households affected by CA-MRSA, we have demonstrated that household member colonization pressure is the most significant predictor of household environmental contamination. These findings suggest that environmental reservoirs, particularly those shown here to harbor a high burden of MRSA, may perpetuate the cycle of reacquisition and transmission. Thus, in addition to personal decolonization,

integrating environmental hygiene measures (which have been effective in healthcare settings [33]) may prove beneficial in household infection prevention efforts. Longitudinal studies are needed to further elucidate *S. aureus* household transmission dynamics, prevalence and factors associated with persistent colonization, and how these phenomena relate to recurrent infection, ultimately informing the design of interventions to disrupt transmission and prevent infection.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Highlights

- **•** MRSA contaminated 46% of environments in households of children with MRSA SSTI
- **•** The most prevalent sites of MRSA were index case bed linens and TV remote control
- **•** Risk factors included household member MRSA colonization burden and home rental
- **•** 14% of pet dogs and cats were colonized with MRSA
- **•** The infecting strain type was the most common strain recovered from the environment

Figure 1.

Prevalence of MRSA contamination at specific surfaces (blue dots/trendline) is inversely related to the proportion of environmental surfaces contaminated with MRSA in households when given surface is contaminated with MRSA (red dots/trendline). Environmental surfaces are listed from left to right in order of increasing MRSA prevalence. When infrequently colonized surfaces (e.g., the index case bathroom towel) are contaminated with MRSA, the proportion of environmental surfaces contaminated with MRSA is high, suggesting these surfaces require a high burden of MRSA in the household prior to becoming colonized. Conversely, the most frequently contaminated surfaces (e.g., the index case bed linens) are contaminated in the context of a low proportion of environmental surfaces contaminated with MRSA, suggesting these surfaces are the first fomites to be colonized (inverse relationship between colonization prevalence and the proportion of environmental surfaces contaminated with MRSA demonstrated via linear regression analysis; $β = -0.696$, $p < 0.001$).

Figure 2.

Molecular epidemiology of S. aureus strains recovered from pet dogs $(N=99)$ and cats (N=31) and their primary caretakers (i.e., the human who is primarily responsible for their feeding, exercising, and waste cleanup). Pets carrying strains concordant or discordant with their primary caretaker are shaded in blue and red, respectively; pets shaded in white did not carry S. aureus. Darker and lighter shades represent MRSA and MSSA pet carriage, respectively. Asterisk (*) denotes that the pet carried S. aureus while the primary caretaker was not colonized with S. aureus (and thus, colonization status was discordant). Figure excludes one cat and one dog who each carried S. aureus but whose primary caretakers were not sampled. Artwork created by Llisole from Noun Project (https://thenounproject.com)

Table 1.

Prevalence of S. aureus on Household Environmental Surfaces and Pets

Abbreviations. MRSA, methicillin-resistant S. aureus; MSSA, methicillin-susceptible S. aureus Note. Culturing technique used for each surface: Eswab (Living room - TV remote control, computer keyboard and mouse, videogame controller, telephone; Bathroom - sink faucet handle, light switch, hand towel, index case bath towel, toilet handle, door handle; Kitchen - hand towel, sponge or cloth, sink faucet handle; Bedroom - index case bed linens) or Baird Parker Agar contact plate (Bathroom - countertop, toilet seat, bathtub, sink, soap bar and dish; Kitchen - refrigerator door handle, table top).

 A Il objects and surfaces were not present in all homes; if present, the object/surface was sampled. N represents the number of households in which this surface was present.

b A dog or cat was present in 81 (54%) households; 132 dogs and cats were cultured in 71 (47%) households.

 $c_{\rm Both MRSA}$ and MSSA were recovered from 1 computer keyboard/mouse and 1 index case bed linens.

d If a landline was not present, the mobile phone of the index case or his/her mother was cultured.

Table 2.

Factors Associated with Proportion of Environmental Surfaces Contaminated with S. aureus and MRSA, Multivariable Model

Note. Poisson generalized linear mixed models constructed in the R library 'MCMCglmm'. See Supplementary Methods for runtime parameters and Supplementary Tables 1 and 2 for primary and secondary covariates used in model selection. "-" denotes variable not included in applicable model.

Abbreviations: MRSA, methicillin-resistant S. aureus; CrI, credible interval; SSTI, skin and soft tissue infection.

 a^2 Colonization pressure calculated as (number of S. aureus [MRSA] colonized anatomic sites) / [(number of sampled household members)*(number of sampled anatomic sites)].

 b_T This does not include the infection that prompted enrollment into the study.

 c_c Excludes personal colonization and pet carriage information.

 d
Multiracial participants include African-American/Caucasian (N=9), Caucasian/American Indian (N=1), and African-American/Caucasian/ American Indian (N=1).