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MAJOR ARTICLE

The Effect of Systemic Antibiotics for Suppurative Skin and Soft Tissue Infections on the Skin Microbiome

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Background. Skin and soft tissue infections (SSTIs) are very common bacterial infections. There are few data on the microbiome of persons with and without SSTIs and the effects of systemic antibiotic therapy.

Methods. We sampled the skin microbiome from 10 outpatients with acute suppurative SSTI before and after systemic antibiotic therapy and enrolled 10 matched controls. Samples were collected at 6 skin body sites (occipital scalp, axilla, interdigital hand web spaces, gluteal crease, inguinal creases, and popliteal fossa), 2 mucosal sites (throat, anterior nares), and the site of skin infection (for case subjects) at baseline and a week later after abscess incision, drainage, and oral antibiotics.

Result. Among 10 SSTI cases, mean age was 41.5 years and 3 had diabetes mellitus. The gluteal crease at baseline had higher α-diversity in controls vs cases ($P = .039$); β-diversity analysis showed significant differences in overall bacterial community composition ($P = .046$). However, at other body sites there were no significant differences by either α- or β-diversity. Systemic antibiotic use did not affect body site diversity indices except at the SSTI site (α-diversity increased, $P = .001$).

Conclusions. We surprisingly found no significant differences in microbiome comparing noninfected skin sites before and after systemic SSTI antibiotic therapy nor significant differences at noninfected skin sites between SSTI cases and uninfected controls. We also found minimal significant differences between microbiome diversity and bacterial signatures at noninfected skin sites between patients with acute skin infection and uninfected controls. Our findings challenge the dogma that systemic antibiotics impact the skin microbiome.

Keywords. antibiotics; longitudinal cohort; microbiome; skin infection; treatment.

Staphylococcus aureus is a ubiquitous pathogen that causes infections, ranging from skin and soft tissue infections (SSTIs) to severe sepsis [1]. *Staphylococcus aureus* is the most common known cause of community-associated SSTIs [2, 3]. SSTIs are an extremely common cause of community infections with an incidence of 4 per 100 persons per year, which is more than twice that of urinary tract infections and 10 times that of pneumonia [4]. Recurrent *S aureus* SSTIs are common and problematic [5–7], occurring in 10%–72% of patients with *S aureus* SSTIs [8]. Reasons for *S aureus* infection recurrence remain elusive, and pathogen-specific factors and human behaviors have been found to have little role [9]. There are few data on the microbiome of persons with SSTIs and the effect of systemic antibiotic therapy on the skin microbiome. Human microbiota disturbances have a role in susceptibility to other infectious diseases such as *Clostridioides difficile*–associated diarrhea [10, 11]. Studies of the human nares have found that Esp-secreting *Staphylococcus epidermidis* and *Corynebacterium* colonization are associated with prevention of *S aureus* colonization [12, 13]. Therefore, we hypothesized that the microbiome of patients with SSTIs may differ from persons without SSTIs.

We also hypothesized that systemic antibiotic therapy would change the skin microbiome. Presumed antibiotic-induced changes in skin microbiome have provided the basis for skin microbiome study protocols [14, 15], in which subjects must avoid systemic antibiotic therapy for months prior to skin sampling. Therefore, we also tested whether systemic antibiotic therapy affected skin microbiome composition in patients with acute SSTI by comparing the microbiome before and after antibiotic treatment.

MATERIALS AND METHODS

Study Population

We enrolled 10 adult participants with acute SSTIs and 10 healthy controls. A control was matched to each case by age (±10 years),

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sex, and race/ethnicity. All participants were recruited from outpatient sites at Harbor-UCLA (University of California, Los Angeles) Medical Center (Torrance, California). Potential participants were excluded if there were plans for hospitalization, or if they had or used any of the following in the prior 30 days: systemic or topical antibiotics, skin creams or disinfectants, immunosuppressive therapy, or hospitalization. We also excluded persons with known chronic dermatologic conditions, such as psoriasis and atopic dermatitis, and recent (<30 days) antibiotic use from the control group. Written informed consent was obtained from participants in their preferred language (English or Spanish). The protocol was approved by the Institutional Review Board of the Lundquist Institute at Harbor-UCLA Medical Center.

Data Collection

Participants completed a standardized questionnaire on demographics, comorbidities, and risk factors of *S aureus* infection (eg, hemodialysis, home intravenous catheters, and healthcare worker status), daily hygiene routines, and use of skin products. Participants were asked to keep their same routine for 7–14 days in terms of bathing/showering, use of any skin products, and use of mouthwash (Table 1).

Case participants were seen after completion of their physician-prescribed antibiotic course (range, 7–14 days) to repeat all of the above procedures. Controls were seen for a follow-up visit the same number of days after the case to which they were matched. During the follow-up visit, all participants were asked about changes in comorbidities and hygienic habits. Controls were asked if they had used antibiotics since baseline visit.

Sample Collection

A sterile flocked swab (Copan, Italy) was used to sample the skin microbiome during the 2 visits: before and after the antibiotic course. Research associates performed hand hygiene; then donned face mask, hair net, gown, and shoe covers; repeated hand hygiene; and then donned gloves. To minimize microbiome specimen contamination, one associate swabbed participants' body sites and a second associate handed the materials to the first research associate. Gloves were changed prior to each new body site swabbed.

Samples were collected at the following 9 body sites: nares, oropharynx, occiput, bilateral axilla, bilateral interdigital web spaces of the hands, bilateral inguinal crease, gluteal crease, and bilateral popliteal fossa, and infection site(s) in cases only located in the arm (n = 3), buttock (n = 2), groin (n = 1), leg (n = 2), and multiple sites (n = 2: arm and leg [n = 1] and axilla and labia [n = 1]) based on current standards established by the Human Microbiome Project [15, 16]. Sites chosen for sampling were based on prior work that demonstrated that the skin microbiome is highly dependent on the "microenvironment" of the sampled body site (ie, moist, dry, or sebaceous skin area) [14]. We also chose body sites for their high likelihood of *S aureus* colonization, specifically nares and notable extranasal sites [17]. For nonmucosal sites, a flocked swab was moistened with SCF-1 buffer (50 mM Tris buffer, 1 mM ethylenediaminetetraacetic acid, 0.5% Tween-20) prior to specimen collection. Swabbing was performed for 30 seconds in a circular motion at each site. In addition, bilateral interdigital web spaces of hand and bilateral popliteal fossa were also scraped with feather disposable scalpels (#10 blade). Specifically, these sites were swabbed for 30 seconds then scraped for 30 seconds and swabbed again for 30 seconds. Residuals from the scalpel were wiped onto the swab after the second swabbing.

The following negative controls or environmental controls were included: air swabs waved in the room for 30 seconds during sample collection, empty Eppendorf tube during DNA extraction, and no template controls during sequencing. Collected samples were placed in SCF-1 buffer in sterile Eppendorf tubes on ice and then immediately transferred to a −80°C freezer after all samples were collected. They were stored at −80°C until DNA extraction was performed.

DNA Extraction

Bacterial genomic DNA was isolated using the Power Soil DNA Isolation Kit (MoBio, Qiagen, Hilden, Germany). DNA extraction was performed following the manufacturer's instructions with modifications to pretreatment of gram-positive bacteria. In brief, 0.2 mg/mL of lysozyme (Sigma-Aldrich, Darmstadt, Germany) was added to the samples collected in the MoBio bead tubes for at least 20 minutes at 25°C. After lysozyme lysis, 0.5 mg/mL of Qiagen's Proteinase K was added to the reaction mixture with incubation at 56°C for 60 minutes. Extracted DNA was quantified using a NanoDrop 2000 spectrophotometer (Thermo Fisher Scientific).

DNA Sequencing and Processing

16S ribosomal RNA gene V1–V3 amplicon sequencing was performed by the UCLA Microbiome Core on the MiSeq instrument (Illumina, San Diego, California) using NEXTflex kits for preparing libraries (Bioo Scientific). NEXTflex polymerase chain reaction I primer mix was used for 16S V1–V3 amplification (forward, 5′-CTCTTTCCCTACACGACGCTCTTCCGATCTAGAGTTTGATCCTGGCTCAG-3′, reverse 5′-CTGGAGTTCAGACGTGTGCTCTTCCGATCTGTATTACCGCGGCTGCTGG-3′). Libraries were sequenced using the MiSeq V3 reagent kit (2 × 300) according to the manufacturer's instructions. A total of 17 million paired-ends were generated. The mothur (v1.44.3) tool and pipeline was used to process the sequencing reads for an approximate total of 7.5 million high-quality reads across 382 samples. Distribution of the high-quality reads are summarized in Supplementary Figure 1. To explain the pipeline briefly, after assembling contigs, reads with any ambiguous reads or reads shorter than 450 reads were excluded. The reads

Table 1. Demographic, Behavioral, and Clinical Data

Characteristic	No. (%)		
	Cases (n = 10)	Controls (n = 10)	Total (N = 20)
Age, y			
18–25	1 (10)	1 (10)	2 (10)
26–50	6 (60)	6 (60)	12 (60)
≥50	3 (30)	3 (30)	6 (30)
Sex			
Male	4 (40)	4 (40)	8 (40)
Female	6 (60)	6 (60)	12 (60)
Ethnicity/race			
Hispanic	6 (60)	6 (60)	12 (60)
African American/Black	2 (20)	1 (10)	3 (15)
White	2 (20)	2 (20)	4 (20)
Asian/Pacific Islander	0	1 (10)	1 (5)
Shower in the last week, frequency			
Once a week or less	0	0	0
2–3 times a week	0	0	0
4–6 times a week	2 (20)	1 (10)	3 (15)
Once a day	8 (80)	8 (80)	16 (80)
≥2 times a day	0	1 (10)	1 (5)
Length of shower, on average			
<1 minute	0	0	0
1–5 minutes	1 (10)	2 (20)	3 (15)
6–10 minutes	3 (30)	3 (30)	6 (30)
11–15 minutes	3 (30)	2 (20)	5 (25)
>15 minutes	3 (30)	3 (30)	6 (30)
Last shower			
<2 hours ago	0	0	0
2–6 hours ago	4 (40)	0	4 (20)
7–12 hours ago	0	2 (20)	2 (10)
13–24 hours ago	5 (50)	6 (60)	11 (55)
24–48 hours ago	1 (10)	2 (20)	3 (15)
>48 hours ago	0	0	0
Soap use			
Yes	10 (100)	10 (100)	20 (100)
No	0	0	0
Frequency			
Never	0	0	0
Rarely	1 (10)	0	1 (5)
About half the time	0	0	0
Most of the time	1 (10)	2 (20)	3 (15)
All of the time	8 (80)	8 (80)	16 (80)
Brand of soap			
Dove	7 (70)	4 (40)	11 (55)
Dial	1 (10)	1 (10)	2 (10)
Axe	1 (10)	0	1 (5)
Suave	0	1 (10)	1 (5)
Generic bar soap	1 (10)	1 (10)	2 (10)
Generic body wash	0	0	0
Shea butter	0	1 (10)	1 (5)
Unknown	0	2 (20)	2 (10)
Mouthwash use in the last week			
Yes	3 (30)	4 (40)	7 (35)
No	7 (70)	6 (60)	13 (65)
Brand			
Listerine	2 (20)	4 (40)	6 (30)
Colgate	1 (10)	0	1 (5)

Table 1. Continued

Characteristic	No. (%)		
	Cases (n = 10)	Controls (n = 10)	Total (N = 20)
Frequency			
Once a week or less	0	0	0
2–3 times a week	1 (10)	1 (10)	2 (10)
4–6 times a week	0	1 (10)	1 (5)
Once a day	2 (20)	0	2 (10)
≥2 times a day	0	2 (20)	2 (10)
Time since last application			
<2 hours ago	0	0	0
2–6 hours ago	1 (10)	2 (20)	3 (15)
7–12 hours ago	0	1 (10)	1 (5)
13–24 hours ago	0	0	0
24–48 hours ago	1 (10)	1 (10)	2 (10)
>48 hours ago	0	0	0
Antibiotic name/dosage			
TMP-SMX, 800 mg-160 mg (twice daily)	7[a](10)	…	…
Cephalexin, 500 mg (4 times daily)	2[a] (20)	…	…
Doxycycline, 100 mg (twice daily)	1 (10)	…	…
Clindamycin, 150 mg (every 8 hours)	1 (10)	…	…

Abbreviation: TMP-SMX, trimethoprim-sulfamethoxazole.

[a]One patient was prescribed both TMP-SMX and cephalexin.

were then aligned to the SILVA SEED v132 reference database, allowing for 6 maximum homopolymers. Chimeras were removed using vsearch (v2.13.3) [18]. The RDP v16 training set was used to first exclude unknown and nonbacterial reads. Finally, taxonomy was assigned using Greengenes v13_8_99 by binning sequences in to phylotypes according to their taxonomic classification [19]. Low-frequency singletons were removed. Potential contaminating bacteria were detected by comparing species prevalence in samples vs the environmental controls using R package "decontam" with a probability threshold of 0.01 [20]. After subtracting the background noise in the environmental controls, the remaining samples were rarefied to a sequencing depth of 2000 reads. Forty-three samples with insufficient high-quality reads were omitted. Supplementary Table 2 shows the final sample set used in the analysis and Supplementary Table 3 shows the summary of the 39 contaminating bacteria excluded from the analysis.

Statistical Analysis

Alpha-diversity (effective number of species, binning by phylotype) was calculated using Shannon index prior to data rarefaction. After rarefying data to a sequencing depth of 2000 high-quality reads, β-diversity metrics was calculated using Bray-Curtis dissimilarity. Permutational multivariate analysis of variance (adonis test) was performed to determine whether the samples clustered by their β-diversity partition distance. Plots for β-diversity analysis were constructed by nonmetric multidimensional scaling, in which each point on the plot represents a sample, and the shorter distance between points indicate increasingly similar microbiome signature [21]. Mann-Whitney-Wilcoxon test was used for 2-group comparisons. Comparison between visits 1 and 2 at the skin infection site used a mixed-effects model, which includes the random effect of repeated measures from the same individual. Analysis of variance *F*-test determined whether the fixed effect of "visit" was significantly associated with bacteria relative abundance.

To validate that the initial finding was not dependent on rarefaction, we analyzed the data at a higher sampling depth of 5000 reads. We also performed the differential abundance analysis using DESeq2, which uses raw count data without any rarefaction, which therefore does not omit any available valid data [22]. Last, we used the "dada2" (v1.16) pipeline to determine whether findings are consistent when processing the sequencing reads into amplicon sequence variants instead of clustering into phylotypes through the "mothur" pipeline [23].

RESULTS

Demographic and Clinical Characteristics of the Participants

We enrolled 21 participants: 11 cases and 10 controls. One case did not return for the second visit and was removed from further analyses. Among the final 10 cases, mean age was 41.5 years, 60% were female, and 60% were Hispanic (Table 1). Comorbidities included diabetes mellitus (30%) and hypertension (30%); other comorbidities and hygienic habits are summarized in Table 1. Seven (70%) were treated with trimethoprim-sulfamethoxazole, 2 (20%) with cephalexin, 1 (10%) with doxycycline, 1 (10%) with clindamycin, and 1 (10%)

with both trimethoprim-sulfamethoxazole and cephalexin (Table 1). All patients' skin infections were cured with their prescribed treatment.

Among controls, mean age was 38.8 years, 60% were female, and 60% were Hispanic (Table 1). Comorbidities included diabetes mellitus (20%); other comorbidities and hygienic habits were similar to those of cases (Table 1, Supplementary Table 1).

Of note, there were no significant changes in risk factors and hygienic measures between study visits in cases and controls (data not shown). No controls took antibiotics between study visits.

Microbial Diversity Across Body Sites (Cases Versus Controls)

Supplementary Table 2 shows the final sample set used in the analysis. The number of high-quality reads were significantly lower in the environmental controls compared to the true samples (Supplementary Figure 1). The samples did not show clustering or batch effect by either the sequencing plates or the date of DNA extraction (Supplementary Figure 2).

The microbiome samples from body sites were dominated by Firmicutes, Actinobacteria, and Bacteroidetes except for the throat, from which Bacteroidetes were most abundant. This phyla distribution was neither significantly different between cases and controls, nor different between visits in both groups (Figure 1). At nearly all noninfected body sites and at both visits, the microbiome of SSTI cases and controls did not show significant differences by α-diversity (Shannon index, Figure 2A) nor β-diversity (Bray-Curtis dissimilarity, Figure 2B). The exception was that the gluteal crease at visit 1 (before antibiotics) showed higher bacterial α-diversity in controls compared to

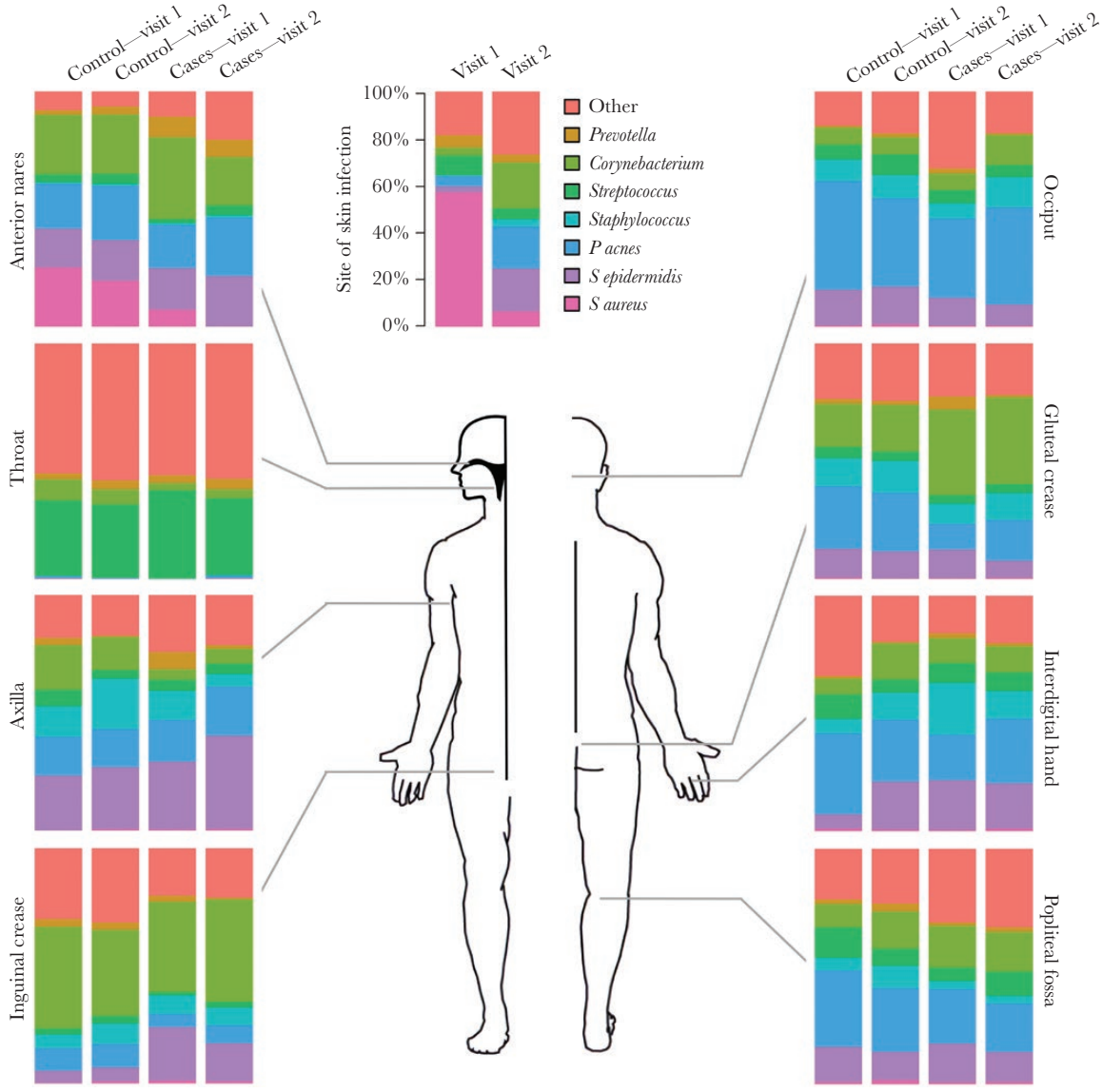

Figure 1. Top bacteria distribution in cases vs controls and in visit 1 vs visit 2 at each body site. Bar plots show the average relative abundance for the top bacteria at each body site and timepoint. Each bar sums to approximately 100%.

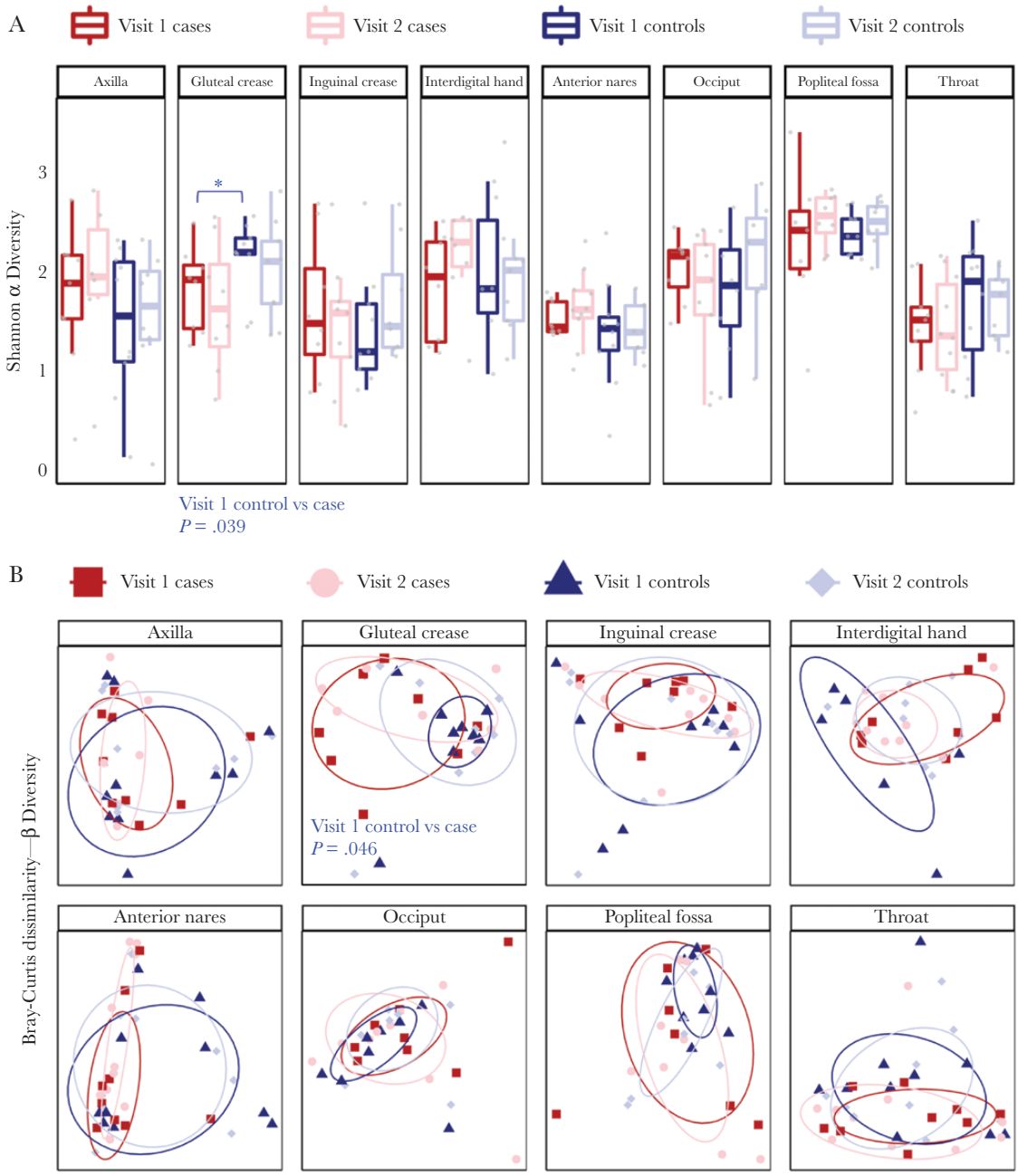

Figure 2. Microbiome diversity in cases vs controls and in visit 1 vs visit 2 at noninfected body sites. *A*, Shannon α-diversity in cases (red) vs controls (blue) and at visit 1 (darker shade) vs visit 2 (lighter shade). The nonparametric Mann-Whitney-Wilcoxon test was used for 2-group comparisons. *B*, Nonmetric multidimensional scaling using Bray-Curtis dissimilarity index (β-diversity). Ellipses represent 80% confidence intervals. Permutational multivariate analysis of variance (adonis test) was performed to determine whether the samples clustered by their β-diversity partition distance.

cases ($P = .039$, Figure 2A) and differences in β-diversity by Bray-Curtis dissimilarity index ($P = .047$, Figure 2B). More specifically, the gluteal crease showed higher relative abundance of *Corynebacterium* species and lower *Propionibacterium acnes* in cases compared to controls at visit 1 (Figure 3).

Effect of Systemic Antibiotic Therapy on Microbial Diversity

Systemic antibiotic therapy did not significantly affect noninfected sites as evidenced by both α-diversity (Figure 2A) and β-diversity (Figure 2B). However, at the SSTI site, systemic antibiotic therapy was associated with shifts in bacterial community composition ($P = .005$) and the α-diversity at the SSTI site was increased at visit 2 ($P = .001$). After antibiotic therapy, the relative abundance of *Corynebacterium* species was increased while *S aureus* was decreased at the SSTI site (Figure 4). At the SSTI site at visit 1, 4 of 13 samples had no or very low relative abundance of *S aureus* and by visit 2 after antibiotics, 11 of the 13 samples had very low *S aureus* (Supplementary

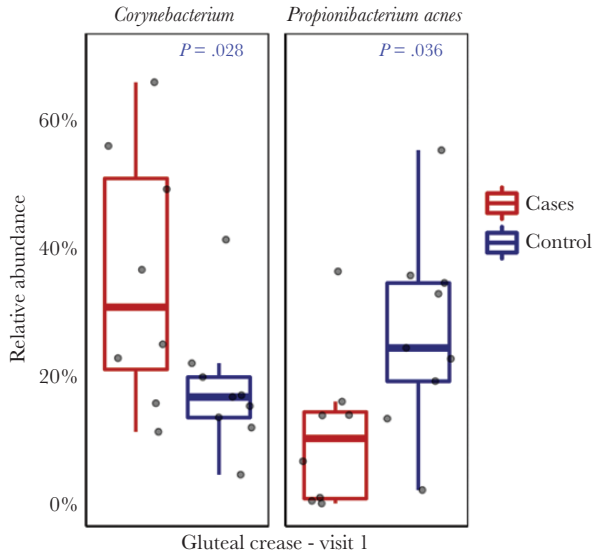

Figure 3. Bacteria that were significantly different between cases (red) and controls (blue) at the gluteal crease visit 1. Differences in relative abundance between the 2 groups were tested using Mann-Whitney-Wilcoxon test.

Figure 3). Additional validation studies showed our findings were consistent (Supplementary Table 4), with significant differences in microbiome (by α- and β-diversity) between cases vs controls at visit 1 gluteal crease and between visit 1 and visit 2 at the SSTI site; there were no significant differences at noninfected skin sites before and after antibiotic therapy. DESeq2 analysis similarly showed that case samples from the visit 1 gluteal crease had higher *Corynebacterium* unclassified, but lower *P acnes* compared to controls. DESeq2 analysis at the SSTI site showed that *S aureus* was higher at visit 1 and that *Corynebacterium* unclassified was higher at visit 2 after antibiotics.

DISCUSSION

Our investigation of skin microbiome at 9 uninfected body sites in persons with and without acute suppurative SSTI, with the exception of the gluteal crease, revealed no significant differences in microbiome diversity between infected and uninfected persons. Cranendonk et al similarly found no significant differences in microbiota between patients with nonsuppurative SSTIs (cellulitis) and controls [24]. They found a correlation between the microbiota of the affected lesion and the microbiota of the unaffected, contralateral limb. In contrast, we studied patients with suppurative SSTIs, which likely represents a distinctly different disease pathogenesis. Interestingly, our findings differ somewhat from that of Horton et al, who, in a cross-sectional study of skin microbiome of skin areas adjunct to a suppurative SSTI, found similar microbiota in areas of infection and the contralateral side, including high levels of *S aureus*, not seen in uninfected controls [25]. However, our study differed in that rather than examine contralateral sides from infected sites, we focused on body sites that represented heterogenous skin types (dry, moist, sebaceous) and mucous membranes known to be commonly *S aureus* colonized (nares, throat).

The only site in which microbiome differences were found, the gluteal crease, was characterized by higher species diversity and higher relative abundance of *Corynebacterium* species compared to controls. Among cases, the posttreatment SSTI site also showed higher species diversity and higher relative abundance of *Corynebacterium* species. Consistent with other reports, we found no significant difference in community structure at the nares and inguinal body sites between infected and uninfected persons [26, 27]. Also consistent, we found an inverse relationship between *Corynebacterium* species and *S aureus* before and after a typical antibiotic course at the SSTI site [26, 27].

Surprisingly and importantly, we found no significant difference in skin microbiome after systemic antibiotic treatment. This finding contrasts with generally held dogma that antibiotics affect the skin microbiome [28], although such dogma has been based on theoretical concerns and supportive data are limited. Our findings may be explained by antibiotic penetration into skin tissues. Skin bacteria are found largely external to epidermis and in the upper parts of the hair follicle. However, systemic antibiotics do not significantly penetrate skin layers superficial to dermis [29], where skin microbiota are sampled. Last, we expected a larger effect size due to antibiotics but it was not statistically detectable, which may be due to a small sample size.

Also surprisingly, we also found little microbiome diversity differences in nares and throat between cases and controls. Some systemic antibiotics can decolonize the anterior nares in a sizeable proportion of persons [30–32], especially clindamycin [30, 31]. Most of our cases received non-clindamycin therapy, possibly explaining the lack of observed mucosal microbiome changes. The lack of effect of systemic antibiotic therapy on skin, nose, and throat microbiome composition by 16S analysis is particularly interesting, given that antibiotic use within the past year has dogmatically been an exclusion criteria for subjects entering microbiome studies [28]. Furthermore, this starkly contrasts with the gut microbiome, which has major and sustained microbiome changes after antibiotic use [33].

Two previous studies have investigated SSTI microbiome, one used the 16S ribosome variable region V1–V3 [26] and the other V3–V4 [27] for amplicon sequencing. While V1–V3 (ours) is prone to more sequencing error due to its longer insert size, the longer insert size also gives better taxonomic classification, for example at the staphylococcal species level [34], which is particularly important for skin microbiome studies. Furthermore, whereas Johnson et al used the Roche GS FLX Titanium 454 sequencer, our study used the now more common Illumina MiSeq, which is more accurate and has higher throughput [26, 27, 35].

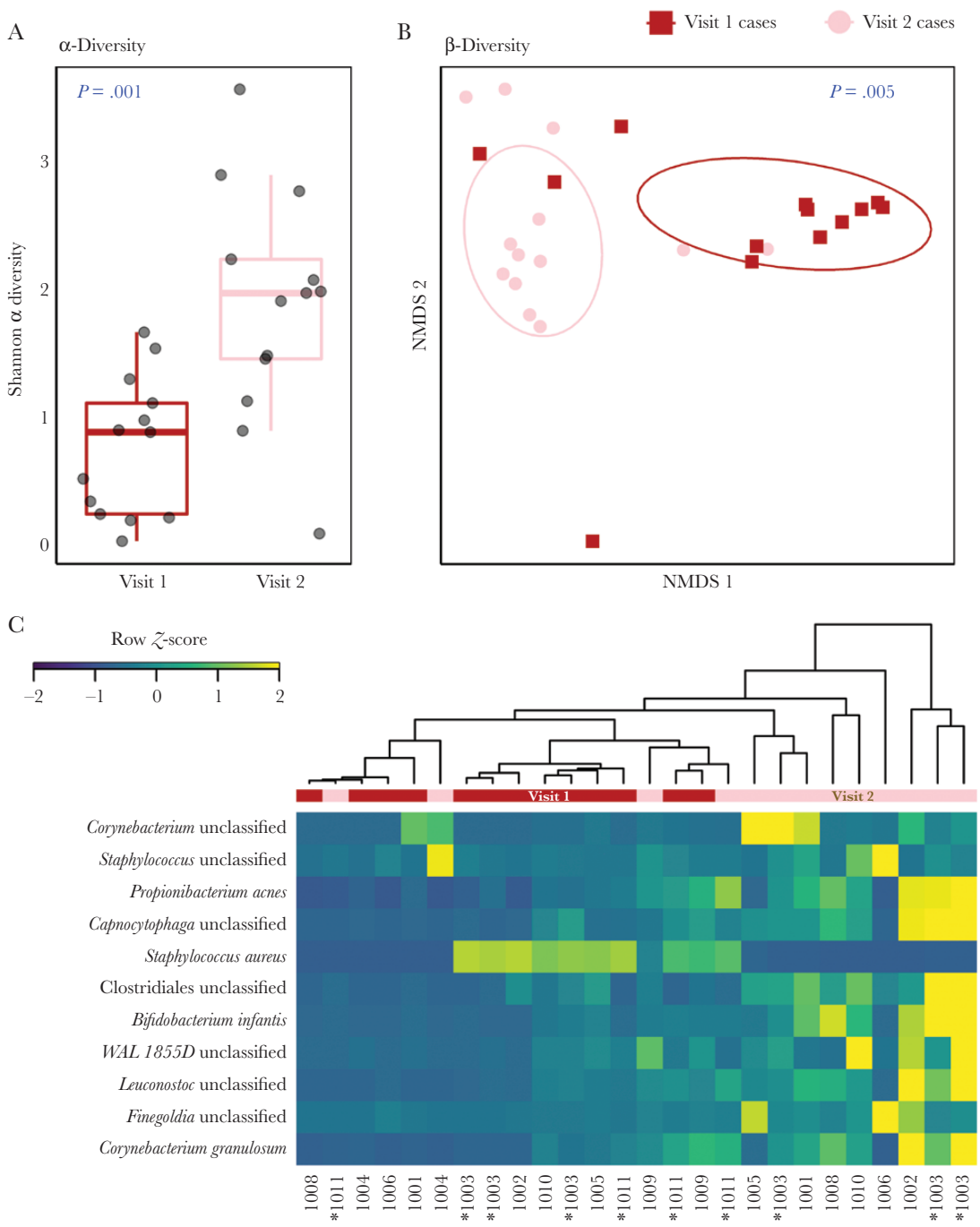

Figure 4. Microbiome comparison at the site of skin infection between visit 1 and visit 2 (after antibiotics). Comparison of α-diversity (*A*) and β-diversity (*B*) at the site of skin infection between visit 1 and visit 2. Ellipses represent 80% confidence intervals. *C*, Heatmap of bacteria significantly different between the 2 visits ($P < .05$). Columns represent a sample and * points to samples with skin and soft tissue infection involving >1 body site. Heatmap *Z*-scores are calculated for each row, which helps visualize expression patterns by centering and normalizing the values. Differences between visit 1 and visit 2 were tested using a mixed-effects model, which includes the random effect of repeated measures from the same individual. Analysis of variance *F*-test determined whether the fixed effect of "visit" was significantly associated with bacteria relative abundance. Abbreviation: NMDS, nonmetric multidimensional scaling.

Our study has strengths. First, we used environmental control samples and nontemplate controls, which are important especially when dealing low biomass samples such as from the skin. Prior studies did not have access to reagents and equipment specific for microbiome analysis and may have been subject to "kit-ome" contaminants [36]. Second, our study is the first to investigate the skin microbiome in areas far distant from the infected site compared to matched control subjects. Our results are somewhat surprising, given that *S aureus* colonization is much higher in patients with acute suppurative SSTI

compared to uninfected controls [37, 38], but we found the microbiomes between these groups was relatively similar even in sites such as the nares and throat, which are more commonly colonized in those with suppurative SSTIs. Last, we conducted a standardized survey collecting information regarding subjects' hygienic habits. Time from last hand washing is associated with skin bacterial composition [39], but effects of bathing and topical product use are relatively unstudied. However, given our relatively small sample size, further studies are warranted to correlate hygienic habits and microbiome. Regardless, we found that instructions to not change bathing habits and skin product use were reportedly nearly always followed, providing the feasibility for future studies.

Our study has limitations. First, we studied suppurative SSTIs, which were presumed to be caused by *S aureus* given that this pathogen causes the majority of suppurative SSTIs [5]. While 6 of 10 cases had high relative abundance of *S aureus* at the SSTI site measured by 16S sequencing, not all had such abundance (Supplementary Figure 3). Therefore, although our goal was to recruit subjects with *S aureus* SSTIs, not all may have had *S aureus* SSTIs, though all infections cleared with empiric antistaphylococcal antibiotic therapy. Second, we had a limited sample size (n = 10), and there may be peculiarities in our study population's genetics and habits. Third, we examined a limited number of heterogenous body sites including mucosal and nonmucosal, oily and dry skin sites, which we expected to have no association with infection to serve as "control" body sites. However, there are more body sites that might differ from those we chose, before and after systemic antibiotics. We speculate that other dry body sites (eg, foot, abdomen) are unlikely to provide additional significant or differing results. Finally, another limitation is the nature of the 16S sequencing technology, which only provides relative abundances instead of absolute bacterial counts, and does not inform us of nutrient use or metabolic pathways.

In conclusion, we found no significant differences between microbiome diversity and bacterial signatures at noninfected skin sites before and after antibiotics in persons with acute SSTIs. Aside from the gluteal crease, we found no significant differences between microbiome diversity and bacterial signatures at noninfected skin sites between patients with acute SSTI and uninfected controls. Our findings also strongly suggest that systemic antibiotics have minimal to negligible effect on skin microbiome diversity and signatures at noninfected skin sites. This finding has major implications for investigations of skin microbiome; specifically, persons with recent systemic antibiotic use should likely not be excluded from surveillance of skin microbiome since such treatment is unlikely to affect microbiota composition. To determine if there are other associations with skin microbiome and SSTI, further work should consider examining the effect of topical antibiotics or antiseptics, such as chlorhexidine gluconate and other populations (eg, children and those suffering from recurrent infections, among others).

Supplementary Data

Supplementary materials are available at *Open Forum Infectious Diseases* online. Consisting of data provided by the authors to benefit the reader, the posted materials are not copyedited and are the sole responsibility of the authors, so questions or comments should be addressed to the corresponding author.

Notes

Acknowledgments. We appreciate the assistance of the University of California, Los Angeles (UCLA) Microbiome Core for performing sequencing.

Data availability. The sequencing data are available on the NCBI Sequence Read Archive under the accession number: PRJNA742681.

Patient consent. This study was approved by the Institutional Review Board of the Lundquist Institute at Harbor-UCLA Medical Center, Torrance, California (internal project number 31237-01). All patients provided informed consent before inclusion.

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Potential conflicts of interest. All authors: No reported conflicts of interest.

All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

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