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Commensal *Staphylococcus epidermidis* Defends against *Staphylococcus aureus* through SaeRS Two-Component System

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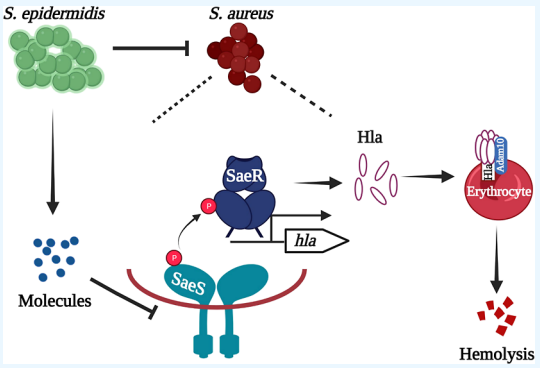
ABSTRACT: *Staphylococcus aureus* is a high-virulent Gram-positive pathogen that is responsible for a serious of diseases. The emergence of antibiotic-resistant *S. aureus* poses a significant challenge in terms of treatment. The recent research on the human microbiome suggested that the application of commensal bacteria is a new strategy for combating pathogenic infections. *Staphylococcus epidermidis*, one of the most abundant species in the nasal microbiome, is able to inhibit the colonization of *S. aureus*. However, during bacterial competition, *S. aureus* undergoes evolutionary changes to adapt to the diverse environment. Our study has demonstrated that the nasal colonized *S. epidermidis* possesses the ability to inhibit the hemolytic activity of *S. aureus*. Moreover, we deciphered another layer of mechanism to inhibit *S. aureus* colonization by *S. epidermidis*. The active component present in the cell-free culture of *S. epidermidis* was found to significantly reduce the hemolytic activity of *S. aureus* in SaeRS- and Agr-dependent manner. Specifically, the hemolytic inhibition on the *S. aureus* Agr-I type by *S. epidermidis* is primarily dependent on the SaeRS two-component system. The active component is characterized as a small molecule that is heat sensitive and protease resistant. Critically, *S. epidermidis* significantly inhibit the virulence of *S. aureus* in a mouse skin abscess model, suggesting that the active compound could potentially be used as a therapeutic agent for managing *S. aureus* infections.



INTRODUCTION

The extensive investigation of the human microbiome has led to the recognition of commensal bacteria as a crucial anti-infective strategy.[1] The human commensal affect the susceptibility to host infectious diseases by several mechanisms including secreting antimicrobial peptides, promoting human immune system, etc.[2] Coagulase-negative *Staphylococci* (CoNS) play a crucial role in defending against the nasal colonization of pathogens in the respiratory tract.[3] *Staphylococcus epidermidis* is one of the most prevalent species found in the human nasal microbiome.[4] *S. epidermidis* not only contributes to wound repair by promoting host immunity but also eradicates pathogens by inducing the production of antimicrobial peptides in nasal epithelial cells.[1,5] Moreover, *S. epidermidis* affects the pathogens through direct interaction. Phenol-soluble modulins (PSMs) produced by *S. epidermidis* displayed antimicrobial activity against pathogens including *Staphylococcus aureus*.[6] *S. epidermidis* disrupts biofilm formation of *S. aureus* by producing serine protease Esp or some unknown small molecules.[7,8]

S. aureus is a common Gram-positive pathogen causing a series of infectious diseases, ranging from skin and soft tissue infection to bacteremia.[9] The pathogenesis of *S. aureus* is attributed to the production of virulence factors, which are controlled by several regulatory systems including the SaeRS two-component system (TCS), accessory gene regulator (Agr) quorum sensing system, SarA transcription factor, etc.[10] Meanwhile, *S. aureus* can asymptomatically colonize in human nostrils, skin, and other mucosal surfaces, which is considered a risk factor for subsequent infections.[11] During the interaction with commensal bacteria, *S. aureus* adapts to different environments through various mechanisms. The naturally occurring polymorphism of virulence regulators is advantageous for the colonization of *S. aureus*.[12] Autoinducing peptide (AIP) originated from CoNS, including *S. epidermidis* or *Staphylococcus caprae*, have been reported to cross-inhibit the Agr system of *S. aureus*.[13,14] It has been observed that Agr-defective *S. aureus* strains are frequently found in human nares,[15] suggesting that *S. aureus* adopts a commensal lifestyle by suppressing the expression of key virulence regulators.

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Based on our previous findings, it appears that the activity of the SaeRS TCS contributes to nasal colonization. Specifically, we observed enhanced survival of strains with reduced SaeRS TCS activity in the nares of human subjects.[16] SaeRS TCS promotes the pathogenesis of *S. aureus* by controlling the expression of over 20 virulence factors including α-hemolysin (*hla*).[17,18] As one of the representative virulence factors, Hla is responsible for the hemolytic activity of *S. aureus* by disrupting red blood cells.[19] We hypothesized that *S. aureus* SaeRS TCS might involve in the competition with commensal bacteria. In this study, we investigated the potential mechanisms underlying the inhibition of *S. aureus* by *S. epidermidis* in healthy individuals through in vitro screening the hemolytic activity of *S. aureus* and in vivo animal models.

RESULTS

Commensal *S. epidermidis* Inhibits the Hemolytic Activity of *S. aureus* in Agr-Dependent and Agr-Independent Manner. Our previous findings have indicated that the production of antimicrobials by *S. epidermidis* is not the primary mechanism involved in its interaction with *S. aureus*.[5] In order to investigate whether *S. epidermidis* promotes the transition of *S. aureus* to a commensal state, we collected the spent culture of 210 commensal *S. epidermidis* isolates and co-cultured them with *S. aureus* ST398-2012-3 for a period of 24 h. Subsequently, we evaluated the growth, hemolytic ability, and Agr activity of *S. aureus* (Figure 1A). Our in vitro co-culture experiments revealed that only one *S. epidermidis* isolate was able to significantly inhibit the growth of *S. aureus* (Figure 1B). Out of the 210 *S. epidermidis* isolates tested, approximately 46.7% (98/210) were found to inhibit the hemolytic activity of *S. aureus* by 50%. However, only 8 isolates were found to inhibit the Agr activity of *S. aureus* by 50% (Figure 1B), indicating that commensal *S. epidermidis* can inhibit the hemolytic activity of *S. aureus* through both Agr-dependent and Agr-independent mechanisms.

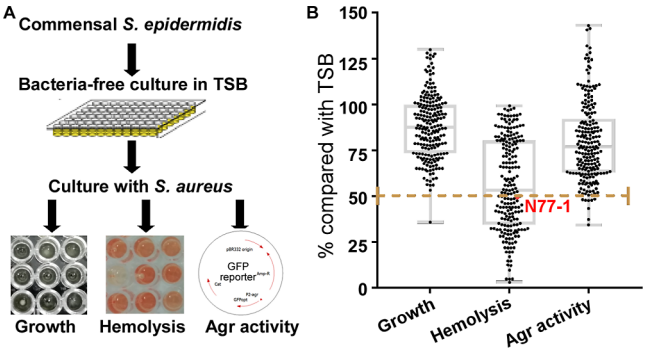

Figure 1. Commensal *S. epidermidis* inhibits the hemolytic activity of *S. aureus* independent of Agr. (A) Experiment procedures to screen for the commensal *S. epidermidis* by the co-culture assay. (B) Growth, hemolytic activity, and Agr activity of *S. aureus* co-cultured with 10% spent culture of *S. epidermidis* were tested compared with the TSB medium alone. The growth of *S. aureus* was determined by testing the OD_{600}. The hemolytic activity was determined by incubating with human red blood cells. The Agr activity was determined by testing the *gfp* activity of the P2 promoter of the Agr system. The red point shows the *S. epidermidis* N77-1 isolate selected for the following experiment.

Inhibition of Commensal *S. epidermidis* on *S. aureus* Hemolytic Ability Is a General Phenomenon. Our study demonstrated that commensal *S. epidermidis* can inhibit the hemolytic activity of *S. aureus* ST398-2012-3. To investigate whether *S. epidermidis* can also affect *S. aureus* strains isolated from the human nares, we randomly selected one commensal *S. epidermidis* isolate, N77-1, which exhibited moderate hemolytic activity. The sequence type for N77-1 was ST520, and it was found to inhibit the hemolytic activity of ST398-2012-3 by 50% (Figure 1B). We also randomly selected 36 commensal *S. aureus* isolates and evaluated their hemolytic activity. Our results showed a significant decrease in the hemolytic ability of *S. aureus* when co-cultured with the spent culture of *S. epidermidis* N77-1 (Figure 2A), suggesting that the inhibitory effect of *S. epidermidis* on the hemolytic activity of *S. aureus* is a general phenomenon.

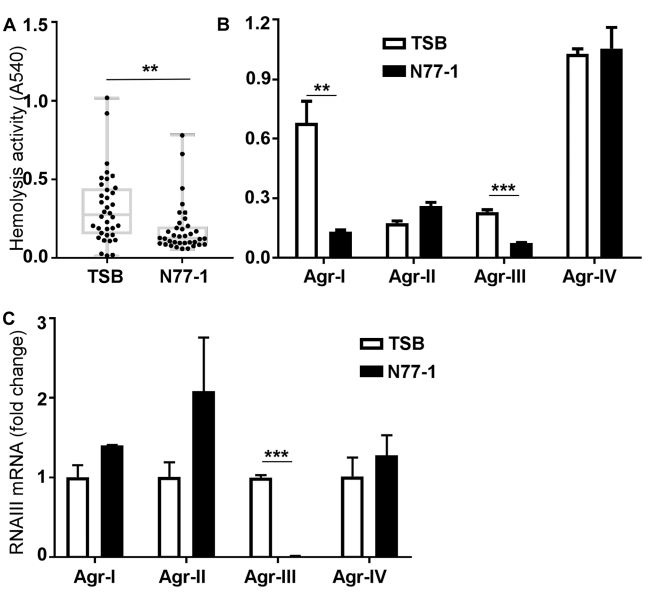

Figure 2. Inhibition of commensal *S. epidermidis* on *S. aureus* hemolytic ability is a general phenomenon. (A) The hemolytic activity of different *S. aureus* isolates was evaluated in co-culture with TSB or TSB with 10% spent culture of *S. epidermidis* N77-1. (B) The hemolytic activity of *S. aureus* isolates with different Agr types was compared with culture in TSB or TSB with 10% spent culture of *S. epidermidis* N77-1. (C) The transcription level of RNAIII was tested by RT-PCR. The statistical analysis was tested by unpaired, two-tailed Student' *t*-test. $^{**}P < 0.001$; $^{***}P < 0.0001$.

The Agr system in *S. aureus* has been classified into four types (I–IV) based on the polymorphism in the amino acid sequences of Agr.[20] The hemolytic activity of all four Agr types of *S. aureus* strains was evaluated when co-cultured with the spent culture of *S. epidermidis* N77-1. Surprisingly, the hemolytic ability of *S. aureus* strains with Agr-I and -III was significantly reduced in the presence of *S. epidermidis* N77-1 (Figure 2B). Despite the significant decrease in the hemolytic activity of *S. aureus* Agr-I type in the presence of *Staphylococcus epidermis* N77-1, the transcription of *RNAIII* remained unaffected (Figure 2C), suggesting that the observed reduction in the hemolytic activity of *S. aureus* Agr-I type is not dependent on Agr activity.

Commensal *S. epidermidis* Inhibits *S. aureus* Hemolytic Activity by Down-Regulating the SaeRS TCS. In order to investigate the mechanism underlying the observed decrease in the hemolytic activity of *S. aureus* in response to commensal *S. epidermidis*, we examined the transcription levels

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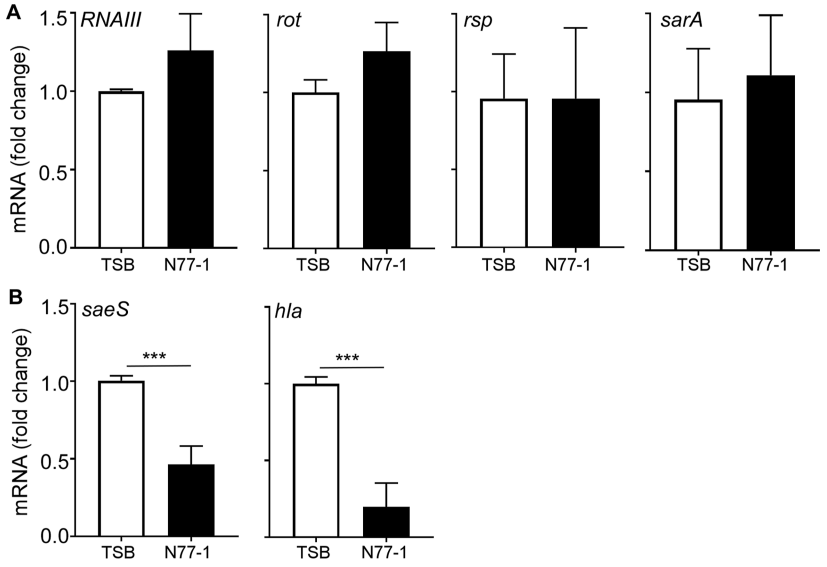

Figure 3. Commensal *S. epidermidis* inhibits the *S. aureus* hemolytic activity by down-regulating the activity of the SaeRS TCS. (A) The transcription of the virulence regulators was tested by RT-PCR in TSB or TSB with 10% spent culture of *S. epidermidis* N77-1. (B) The transcription of *saeS* and *hla* was tested by RT-PCR in TSB or TSB with 10% spent culture of *S. epidermidis* N77-1. The statistical analysis was tested by unpaired, two-tailed Student' *t*-test. ***$P < 0.0001$.

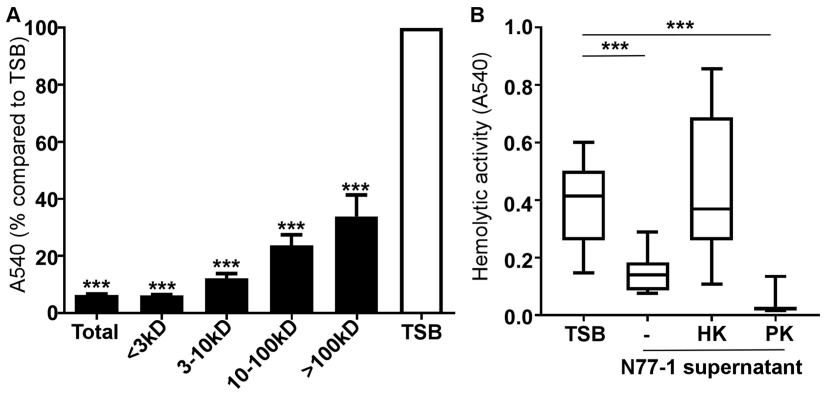

Figure 4. Characterization of the molecules secreted by *S. epidermidis*. (A) Hemolytic activity of *S. aureus* ST398-2012-3 grown in the presence of the *S. epidermidis* supernatant fractionated by molecular weight (kDa). (B) Hemolytic activity of *S. aureus* ST398-2012-3 grown in the presence of *S. epidermidis* supernatant treated with heat (HK) or protease K (PK). The statistical analysis was tested by unpaired, two-tailed Student' *t*-test. ***$P < 0.0001$.

of the main regulators, including *RNAIII*, *rot*, *rsp*, *sarA*, and *saeS* using RT-PCR when co-cultured with the spent culture of *S. epidermis* N77-1. The transcription levels of *RNAIII*, *rot*, *rsp*, and *sarA* were not affected by the presence of *S. epidermidis* N77-1 (Figure 3A). Interestingly, we observed a significantly decreased expression of *saeS* of *S. aureus* co-cultured with *S. epidermidis* (Figure 3B). Consistent with this finding, we also observed a significant inhibition of *hla* transcription in the presence of *S. epidermidis* (Figure 3B). These results suggest that the commensal *S. epidermidis* inhibits the hemolytic activity of *S. aureus* by down-regulating the activity of the SaeRS TCS.

Heat-Sensitive, Protease K-Resistant Small Molecules Secreted by Commensal *S. epidermidis* Inhibit the Hemolytic Activity of *S. aureus*. By far, we observed that the spent culture of *S. epidermidis* inhibits the hemolytic activity of *S. aureus* by down-regulating the activity of the SaeRS TCS. To identify the active component secreted by *S. epidermidis*, the spent culture of *S. epidermidis* was fractionated into four fragments based on molecular weight using ultrafiltration spin columns (Millipore): <3, 3−10, 10−100, and >100 kDa. Surprisingly, all four fragments significantly inhibited the hemolytic activity of *S. aureus*. Notably, the fragment with a molecular weight of less than 3 kDa displayed the most potent inhibitory effect (Figure 4A).

Next, we characterized the active small molecules secreted by *S. epidermidis*. Heat-killed (95 °C for 20 min) spent culture of *S. epidermidis* failed to inhibit the hemolytic activity of *S. aureus* (Figure 4B), indicating that the active molecules are likely to be heat sensitive. Interestingly, treatment with protease K did not affect the inhibitory effect of the spent culture on *S. aureus* (Figure 4B), suggesting that the active molecules are not proteins. In summary, these results suggested that *S. epidermidis* down-regulates the SaeRS TCS of *S. aureus* through the secretion of heat-sensitive small molecules.

Commensal *S. epidermidis* Inhibits the Virulence of *S. aureus* in the Mouse Skin Abscess Model. The SaeRS TCS activity is essential for *S. aureus* virulence.[17] To investigate whether the inhibition of SaeRS TCS activity by

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S. epidermidis contributes to the virulence of *S. aureus* in vivo, we co-infected *S. epidermidis* N77-1 and *S. aureus* ST398-2012-3 strains in a murine skin abscess model. The co-infection of *S. epidermidis* and *S. aureus* resulted in significantly smaller skin lesions. On day 2 post-infection, the average abscess area caused by the *S. aureus* group was 195.3 mm^2, while the abscesses caused by strains from the co-infection group was 34.9 mm^2 on average (Figure 5A,B). In order to verify the involvement of *S. epidermidis* in the decreased virulence of *S. aureus*, we implemented another co-infection group: *S. aureus* with heat-killed *S. epidermidis*. Notably, the co-infection of the heat-killed *S. epidermidis* N77-1 and *S. aureus* led to a significantly larger average abscess area of 169.7 mm^2 compared to the co-infection of regular *S. epidermidis* and *S. aureus* (Figure 5A,B). Although the bacterial loads of *S. aureus* were not affected in the presence of *S. epidermidis* (Figure 5C), the histological analysis displayed more severe inflammation in the skin tissues of mice infected with *S. aureus* alone or with *S. aureus* and heat-killed *S. epidermidis* N77-1 compared to those with regular *S. epidermidis* and *S. aureus* (Figure 5D). These data suggested that *S. epidermidis* inhibits the virulence of *S. aureus*.

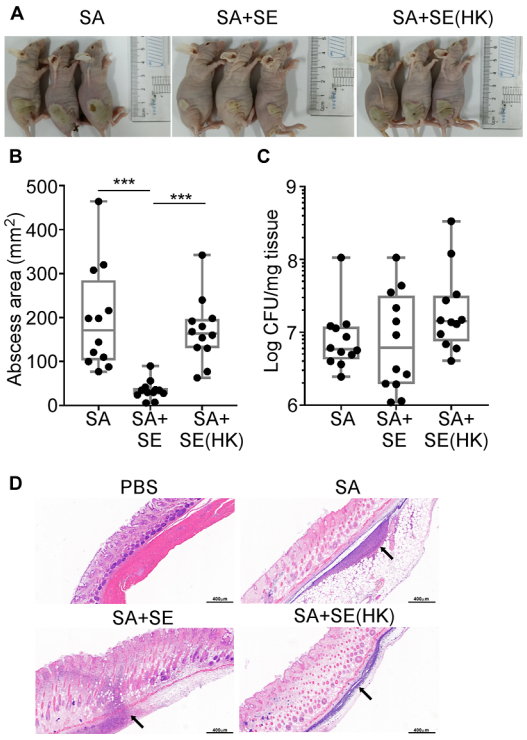

Figure 5. *S. epidermidis* inhibits the virulence of *S. aureus* in the mouse skin abscess model. (A) The virulence of *S. aureus* ST398-2012-3 was tested in a murine skin abscess model. Each mouse was challenged with the tested strain (10^8 cfu) by the subcutaneous injection of the *S. aureus* co-culture with or without *S. epidermidis* N77-1 (10^8 cfu). *S. epidermidis* N77-1 was heat-killed at 95 °C for 20 min when necessary. The representative subcutaneous skin abscesses of the infected mice were observed on day 2 after infection. (B) The abscess areas (length × width) were compared on day 2 after infection. (C) Bacterial loads of abscesses were tested and normalized by tissues (mg) on day 2 after infection. The experiment was repeated three times, and the results were pooled. (D) HE staining of skin abscesses on day 2 after infection. The arrows display the inflammation site in the skin tissues. The statistical significance was measured by the one-way analysis of variance (ANOVA). $^*P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$. SA, *S. aureus* ST398-2012-3; SE, the commensal *S. epidermidis* N77-1; HK, heat-killed.

DISCUSSION

With the prevalence of antibiotic-resistant *S. aureus*, it is urgent to develop new antibacterial strategies. CoNS, especially *S. epidermidis*, develop an arsenal of mechanisms defending against *S. aureus*.[15,16] In the study, we elucidated that heat-sensitive small molecules secreted by *S. epidermidis* effectively inhibit the hemolytic activity of *S. aureus* by down-regulating the SaeRS TCS. Critically, *S. epidermidis* is capable of efficiently inhibiting *S. aureus* virulence in vivo.

In this study, we have demonstrated that the bioactive component secreted by *S. epidermidis* exerts a direct inhibitory effect on the hemolytic activity of *S. aureus* (Figure 2). The human nares have been identified as a reservoir of *S. aureus*, serving as a primary source for subsequent infection.[21] Nasal commensals have developed several strategies to defend against the colonization of *S. aureus*.[3] The antimicrobial peptides produced by *Staphylococcus lugdunensis* and *Staphylococcus hominis* prohibit *S. aureus* growth directly.[4,22] *S. caprae* prevents the colonization of *S. aureus* by blocking Agr the quorum sensing system.[14] As the most abundant species in human nares, *S. epidermidis* defends against pathogens by promoting immune defense. For example, *S. epidermidis* inhibits the colonization of *S. aureus* or *Moraxella catarrhalis* by inducing the production of antimicrobial peptides of host cells.[5] *S. epidermidis* plays a role in antiviral response by promoting interferon-lambda-dependent immunity.[23] Furthermore, the commensal strain of *S. epidermidis* is able to produce molecules with the anti-biofilm activity.[8] All of these studies highlight the search for compounds that can aid people in the fight against *S. aureus* infection.

In the present study, around 46.7% commensal *S. epidermidis* isolates inhibit the hemolytic activity of *S. aureus* Agr-I ST398-2012-3 (Figure 1). The cross-inhibition of the Agr system between CoNS and *S. aureus* has been reported extensively.[13,14] Agr, one of the main regulators for the hemolytic activity of *S. aureus*, is deemed as the most popular mechanism during pathogen competition.[24] By testing the *RNAIII* transcription of *S. aureus* with varying Agr types, we observed that the Agr activity in *S. aureus* ST398-2012-3 (Agr-I) remains unaffected in the presence of *S. epidermidis* (Figure 2). Most of the nasal colonized *S. aureus* isolates were Agr-I type (74.4%), while around 11.7 and 6.7% *S. aureus* belong to Agr-II and Agr-III types (data not shown). Our results suggested that the inhibition of nasal colonized *S. aureus* by *S. epidermidis* is not totally dependent on the Agr system.

Our current investigation revealed that the small molecular secretion by *S. epidermidis* is resistant to the degradation by protease K, implying that the inhibitory effect is not derived from a proteinaceous molecule. One limitation of our study is that we did not identify the specific small molecules responsible for the inhibitory effects of the SaeRS TCS in *S. aureus*. SaeRS TCS is a regulatory system that has been shown to contribute to the virulence of *S. aureus*.[17] SaeS, a histidine kinase sensor, autophosphorylates a conserved histidine residue upon sensing external stimuli. The phosphate group can be transferred to a conserved aspartic residue of the response regulator SaeR.[25] The phosphorylated SaeR regulates the expression of a variety of virulence factors by binding to their promoter regions.[26] Although the histidine kinase SaeS sense the environment by the extracellular linker peptide,[27] the

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exact mechanism of how SaeS recognize the signals is still unknown. The *S. aureus* SaeRS system can be affected by host signals or environmental cues. SaeS is activated by human neutrophil peptides 1, which may contribute to the defense against neutrophils.[28] The silkworm apolipophorin protein can inhibit *S. aureus* virulence through SaeRS TCS.[29] The skin surface with a low pH (5.5) inhibits the Sae activity.[30] The molecule 6-*N*-hydroxyaminopurine (6-HAP) produced by *S. epidermidis* protects against skin neoplasia by inhibiting the DNA polymerase activity.[31] The metabolite short-chain fatty acids (SCFAs) including acetate, propionate, and butyrate existed in the fermentation of *Cutibacterium acnes* and *S. epidermidis* isolates by gas chromatography−mass spectrometry (GC−MS) analysis.[32] The bacterial fatty acid inhibits the SaeS activity by affecting the sensing of the SaeS transmembrane domain.[33] The human skin fatty acid *cis*-6-hexadecenoic acid also displays high antistaphylococcal activity by repressing the SaeRS TCS.[34] It is possible that a particular heat-sensitive metabolite produced by *S. epidermidis* may contribute to the suppression of *S. aureus*. However, the precise identity of the molecule involved requires further elucidation.

In conclusion, our study confirmed that the commensal *S. epidermidis* suppresses the hemolytic activity of *S. aureus* by suppressing the expression of *hla*, which is regulated by the SaeRS TCS. We observed that the SaeRS TCS also plays a role in regulating the bacterial competition. Furthermore, the inhibition of the SaeRS TCS by *S. epidermidis* was observed to impact the virulence of *S. aureus* in a mouse model, indicating a promising therapeutic approach for defending against *S. aureus* infections.

METHODS

Study Design and Sample Collection. This study was approved by the Ethics Committee of Ren Ji Hospital, Shanghai Jiao Tong University School of Medicine, Shanghai, China. All individuals or their legal guardians were provided an informed consent. The animal experiments were performed following the Guide for the Care and Use of Laboratory Animals and were approved by the Ethics Committee of Ren Ji Hospital, Shanghai Jiao Tong University School of Medicine, Shanghai, China. As reported previously, we isolated *S. epidermidis* and *S. aureus* from the nasal swabs of healthy volunteers with different ages.[5] In total, 210 randomly selected *S. epidermidis* isolates and 36 randomly selected *S. aureus* isolates were used for the co-culture assay. For RT-PCR and animal experiments, *S. aureus* ST398-2012-3[35] and *S. epidermidis* N77-1 (this study) were used for the experiments. Generally, *S. aureus* isolates were grown in a tryptic soy broth (TSB; Oxoid) with shaking at 200 rpm at 37 °C.

Co-culture Assay. *S. epidermidis* was cultured for 18 h. After centrifuging (4000*g*, 10 min), the supernatant was collected and filtered through a 0.22 μm container (spent culture). Fresh TSB containing 10% spent culture of *S. epidermidis* was used to culture *S. aureus* isolates. When necessary, the spent culture was heat-killed at 95 °C for 20 min or treated with protease K (PK 50 μg/mL) at 37 °C for 1 h.

Erythrocyte Lysis Assay. Supernatants of *S. aureus* overnight cultures were collected and incubated with human erythrocytes (2% v/v in phosphate buffered saline, PBS) for 1 h at 37 °C in a 96-well round-bottom plate. After centrifugation (1500*g*, 10 min), the supernatant (100 μL) was pipetted into another sterile 96-well flat-bottom plate. The optical density was determined at 540 nm by using an enzyme-linked immunosorbent assay reader (Synergy; BioTek).

GFP Reporter Assay. The report plasmid pOS1-P2-*gfp* for the Agr activity was constructed in our previous study.[36] *S. aureus* was cultured overnight; the bacteria pellets were collected and incubated with lysostaphin (50 μg/mL) at 37 °C for 1 h. Lysates were then diluted to 1 mL PBS. 200 μL liquid was pipetted into a black 96-well microtiter plate in triplicate. Fluorescence (485 nm excitation, 538 nm emission) was measured on an enzyme-linked immunosorbent assay reader (Synergy; BioTek) and normalized by OD_{600}.

Real-Time Quantitative Reverse-Transcription PCR. Bacteria were cultured in TSB overnight. The overnight cultures were diluted (1:100) in fresh TSB or TSB with 10% spent culture of *S. epidermidis* and grown for 8 h at 37 °C with shaking. The pellets were collected by centrifuge at 4000*g* for 10 min and broken by FastPrep-24 (MP 116005500). After centrifugation, the supernatant was used to isolate the total RNA and further synthesize complementary DNA according to the manufacturer's instructions (Qiagen). The cDNA was used as a template for real-time PCR using SYBR-green PCR reagents (Roche) with primers listed in Table S1. Reactions were performed in a MicroAmp Optical 96-well reaction plate using a 7500 Sequence Detector (Applied Biosystems). All qRT-PCR experiments were performed in triplicate using *gyrB* as an internal control gene at the mRNA level.

Mouse Skin Abscess Model. *S. aureus* and *S. epidermidis* were cultured overnight in TSB. The next day, the overnight culture was diluted at 1:100 in fresh TSB and grown at 37 °C for 4 h. After centrifugation (6000 rpm, 10 min), the bacteria pellets were washed twice with sterile PBS and suspended in PBS. When necessary, *S. epidermidis* was heat-killed at 95 °C for 20 min.

Outbred, immune-competent hairless male mice (4−6 weeks old) were anesthetized with Avertin (Sigma T48402) by intraperitoneal injection and administered with *S. aureus* (10^8 cfu in 100 μL of PBS) by s.c. injection on the back. The skin lesion size was measured by length (L) and width (W). On the 2nd day, mice were killed by anesthesia overdose, and the infected skin tissue was excised and homogenized in 500 μL PBS. The homogenized skin tissue was diluted and plated on 5% sheep blood agar to determine cfu. Skin tissues were fixed in 4% paraformaldehyde, paraffin-embedded, sectioned, and stained with hematoxylin & eosin (H&E) staining.

Statistical Analysis. Statistical analysis was performed with the GraphPad Prism 7.0. For the comparison between the two groups, the unpaired, two-tailed Student's *t*-test was performed for statistical analysis. One-way ANOVA was used for the comparison among the three-set data groups. Error bars indicated the standard deviation (±SD), and P values < 0.050 were considered statistically significant. *P < 0.05, **P < 0.01, ***P < 0.001.

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acsomega.3c00263.

Oligonucleotides used in this study (PDF)

All data sets regarding the experiments are available from the corresponding author upon reasonable request

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Author Contributions

N.N., L.W., and Q.W. contributed equally to this work. All authors participated in the project. N.N. performed the animal test. L.W. and Q.W. collected the clinical isolates. S.L., C.J., Y.S., and A.W. did the co-culture assay. Y.W. and N.Z. participated in the data process, analysis, and interpretation. N.N. and Q.L. drafted the manuscript. H.W., M.L., and Q.L. supervised the project and revised the manuscript. All authors approved the final version of the manuscript.

Notes

The authors declare no competing financial interest.

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ABBREVIATIONS

S. epidermidis, *staphylococcus epidermidis*; *S. aureus*, *staphylococcus aureus*; qRT-PCR, real-time quantitative reverse-transcription PCR; CoNS, coagulase-negative *Staphylococci*; PSMs, phenol-soluble modulins; SaeRS TCS, SaeRS two component system; Agr, accessory gene regulator; PK, protease K; HK, heat-killed; 6-HAP, 6-*N*-hydroxyaminopurine; SCFAs, short-chain fatty acids; GC−MS, gas chromatography−mass spectrometry

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