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***Staphylococcus aureus* Nasal Carriage and its Contributing Factors**

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Summary

Staphylococcus aureus is a medically important pathogen that can also spread amidst the community. Bacteria that reside in anterior nares of hosts serve as reservoirs for both the spread of the pathogen and to predispose the host to subsequent infections. Here, we will review the extent and variability of nasal carriage, and the possible causative factors – both from the host and the bacterium. We also discuss the existing molecular typing techniques used for studying variations among strains of *S. aureus*. Finally, we discuss the possible areas of studies that are open in this field. Given the pathogen's importance in healthcare setting, such areas of study vary vastly, from fundamental research to applied medical care and use of alternative medical regimes for control of SANC. Unsurprisingly, our conclusions also underscore the importance of making policy decisions based on local ethnic and socio-economic population structure.

Executive summary

- Epidemiology and abiotic factors contributing to *Staphylococcus aureus* nasal carriage
 - Prevalence varies in industrialized nations from 26% to 35% of the population.
 - Ethnicity and availability of medical care affect the carriage levels.
 - Developed nations have a higher percentage of carriage than the developing ones.
 - Other factors like intravenous drug usage and HIV contribute to increase in carriage.
- ***Host factors contributing to SANC and persistence***
 - Mutations in immunity related genes affect rates of carriage.
 - Cell surface receptors like TLR-2 are intricately linked to carriage.
- ***Bacterial factors involved in SANC***

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- Clumping factor B and *sdrE* are linked to nasal carriage.
- Horizontal gene transfer plays an important role in establishing SANC.
- Genome scale analysis reveals a possible role for novel protein transport systems in carriage.

- ***Other factors affecting SANC***

- Bacterial interference, by pathogenic and other non-pathogenic species might play a role.
- Intra-strain variability contributes to longitudinal changes in carrier strain profile.

- ***Future directions for studies on SANC***

- Multi-ethnic epidemiological studies to identify population specific variation in SANC.
- Metagenome level analysis of nasal epithelial niche in carriers and non-carriers.
- Microbiome scale sequencing and analysis for interrogating effects of bacterial interference.
- Effects of acute and chronic stress and psychological depression on nasal carriage.

Introduction

Staphylococcus aureus is a versatile pathogen capable of growth and infection under diverse conditions. It has an extremely plastic genome capable of high variability and is known to acquire genes from closely related species. Besides being an interesting model organism to study genomic plasticity and evolution (1) (2), it is also a medically important pathogen. It causes a wide spectrum of afflictions, in post-surgical patients (e.g. post-trauma, burns or wounds) under an immuno-suppressive drug regime, pediatric and geriatric patients, diabetics and immuno-compromised patients (AIDS in particular) among others. The pathogen can be contracted either in a hospital setting (Nosocomial or “Hospital-Acquired”) or from the community (“Community-Acquired”). In the hospital and health care units, it is a growing threat due to rapid acquisition and evolution of drug resistance. Such a threat has been widely acknowledged and studied in great detail, and is outside the scope of this preview. On the other hand, it is known that this pathogen can be transmitted between people in a normal population. Some people carry this pathogen in their anterior nares, thus serving as a reservoir for infections in scenarios mentioned above. Such sub-clinical harboring of the bacterium in the anterior nasal nares is called *nasal carriage* and the hosts are called ‘carriers’. It is notable that most of the carriers are not infected seriously by the bacterium, but rather act as reservoirs for the pathogen assisting its spread in the community. The health implications of *S. aureus nasal carriage* (SANC) with respect to human physiology are well known and reviewed elsewhere (3).

In this review, we will discuss the multifaceted problem of SANC under the following sections: its epidemiology and the trends of SANC observed across several studies, host factors that are known to affect SANC and its implications, techniques for molecular typing of carrier strains, bacterial factors responsible for carriage and recent findings, other factors affecting SANC and finally, future directions that should be taken within the field to understand and control SANC and the spread of *S. aureus*.

Epidemiology and abiotic factors contributing to SANC

SANC is a global phenomenon that is affected by various factors including, but not limited to, age, health, economic status and the country of residence. Carriage in groups of people within continental USA vary from 26% (4) to 32% (5) with local population variations likely accounting for differences in studies (Cepedes 2001). On the other hand, global trends of SANC show a larger variation (10% in adults in Turkey (6)) to close to 25% in another cohort study in Malaysia (7). Despite the global range having remained constant across several studies, the incidence of methicillin resistance in the bacterium has increased noticeably. Two trends are evident in literature with regard to SANC. The first observed trend is that SANC is high in developed countries, [US (32%) (5), Netherlands (35%) (8)] as compared to underdeveloped and developing countries [Nigeria (14%) (9), Malaysia (26%) (7), India (16%) (10), Indonesia (<10%) (11)]. It is possible that more cohort studies in both developed and other countries, in the future might either annul or even reverse this trend. We speculate that high levels of personal hygiene in the developed countries and subsequent low exposure to antigens and pathogens might contribute to decreased clearing of the pathogen in the tested subjects. This would reflect as increased proportion of carriers in the studies cited. Another important trend that is globally observed is the increased rates of carriage amongst intravenous drug users (12) and immuno-compromised individuals (13).

Most of these studies identify a particular target population (encompassing students, hospital workers, infants in neonatal ward, geriatric patients et cetera) and study carriage in that cohort with respect to certain standard variables (viz., age, sex, health status, antibiotic intervention in a given time frame and others) within that group. Very few studies go beyond the normal variables and take into account important differences like ethnicity, economic differences and the availability of health care and literacy. Haplotype (lineage) differences in humans have been associated well with certain medical conditions like atherosclerosis (14) and cancer and diseases like HIV infection (15) and hepatitis (16)(HCV infection). In the light of above argument, it is logical to expect certain haplotypes to be more or less susceptible to *S. aureus* SANC. Indeed, as we shall discuss next, certain haplotypes are known to influence the severity of nasal carriage and colonization. More of these studies must be performed to test the correlation between broad ethnic groups and their resistance to *S. aureus*, the ethical concerns of such a study notwithstanding. Insight into host factors derived from these studies will allow scientists, doctors and policy makers alike, to design an optimal local strategy to control the spread of SANC in their respective communities.

Control measures for SANC in the community is largely passive, and limited to administration of mupirocin in case of a clinical manifestation. Mupirocin has been shown to be generally effective in controlling SANC (17) by delaying the onset of nosocomial infections in hospitalized patients. On the other hand, such preventive removal of nasally carried *S. aureus* is only effective in hosts that have a predisposition against SANC. In permissive hosts, such treatment only allows the recolonization of their epithelium by the same or a different strain (18). In a few cases, sporadic clearing of nasal microbiota led to permissibility to carriage. Summarily, SANC is a complex phenomenon whose global distribution depends on a plethora of variables inherent to the host and the bacterium apart from abiotic / social factors like availability of health care and economic status. In the following pages, we shall review the biotic factors that influence SANC, both belonging to the host as well as to the pathogen.

Host factors contributing to SANC and persistence

The nasal vestibular region serves as the primary reservoir of *Staphylococcus aureus* in humans with nearly 20% of individuals being persistent nasal carriers. Although numerous

studies have been performed on SANC and the associated risk factors, very little is known about the host-pathogen interaction. SANC of host is an equilibrium achieved in the host-pathogen interaction dynamics under non-infective, permissive conditions. Thus successful combat and control of SANC requires a thorough knowledge of possible host factors involved. Several host factors have been suggested to be associated with nasal carriage and can be broadly divided into host genetic factors and immune factors. A list of host factors affecting nasal carriage is given in table 1.

SANC is influenced by certain HLA types, and also by polymorphic variations in several genes. A study conducted on healthy laboratory workers and patients attending an outpatients' clinic found that the presence of the histocompatibility antigen (HLA-DR3) would predispose an individual to SANC (19). Polymorphic variations of numerous genes such as Fc fragment of IgG (Fc γ R), human glucocorticoid receptor, polymorphic variations in the vitamin D receptor (VDR) gene in patients with type I diabetes (20) were also found to be linked with nasal carriage. Further polymorphic differences in IL-4 (C542T genotype) and the human complement cascade activator serine protease C1 inhibitor (C1INH V480M) (21) were linked to increase nasal carriage. Moreover, a link between certain genotypes of human glucocorticoid receptor and nasal carriage has been ascertained (22). This is of particular interest since treatment with corticosteroids is known to subdue immune responses. It is possible that these receptor genotypes are highly responsive to general levels of corticosteroids thereby reducing overall immune resistance in subjects. Besides the blood borne and cellular immunity factors, nasal secretions have an important role in protecting against pathogens. Human nasal fluid is composed of a variety of antimicrobial proteins and peptides such as lysozyme, lactoferrin, Secretory Leukocyte Protease Inhibitor (SLPI), neutrophil alpha-defensins, beta-defensins and cathelicidins (23) (24). Lysozyme is a bacteriolytic enzyme abundant in nasal fluid and hydrolyses the glycosidic linkages in peptidoglycan present on bacterial cell wall. Several studies have found that many of the antimicrobial peptides have little or no activity against many strains of *S. aureus*. Lactoferrin, an iron-binding siderophore with a cationic C-terminal region, contributes to antimicrobial activity as well as Fe²⁺ sequestration. Neither lysozyme nor lactoferrin show significant activity against *S. aureus* by themselves (4). Some of the antimicrobial peptides such as human beta-defensin-1 (HBD-1) are constitutively expressed in the airway fluid, while the expression of HBD-2 and -3 are induced upon pathogenic recognition. Of the three beta-defensins, HBD-3 is most effective against *S. aureus* (25). Even though neutrophil alpha-defensins are upregulated in nasal fluid, most of the *S. aureus* strains are highly resistant to their bactericidal action. Further, there is evidence to suggest that multiple antimicrobial factors in nasal fluid act in synergy to effectively clear the bacteria, at least in case of non-permissive hosts. In fact, beta-defensins, human cathelicidin (LL-37), and lysozyme exhibited a synergistic anti-staphylococcal activity in CF patients when administered via nasal inhalation (26). Dysregulation of expression of one or more of these innate peptides might predispose a person to SANC.

Host factors other than the immunity genes have also been identified, that are involved in SANC. In their studies in mice model, Gonzalez-Zorn and others (27) identified CFTR and TLR-2 as host factors that protect the system against carriage of *S. aureus*. It was shown that nasal colonization / carriage was not only pronounced in mice that were deficient for CFTR, but also in those that lacked TLR-2. It is interesting to note that abrogation of TLR-2 is important for establishing carriage, since our own work suggests that a model carrier clinical isolate was capable of suppressing TLR expression in human nasal epithelial cells (28). Besides, the rate limiting step in host pathogen interaction is the adhesion of bacterium to the host skin surface. It has been known that surface proteins on cornified desquamated nasal epithelium like cytokeratin 10, involucrin and loricin bind to bacterial factors (29) (30).

While animal studies like the ones described above (27) are informative, it is important to recognize a major shortcoming in using animal model systems. In many cases, the pathogen adapts itself to specific host types. Examples of such adaptive behavior have been reported for both cows (31) and other farm animals (32).

Techniques for molecular typing of *S. aureus* and their application to SANC

Before we discuss bacterial factors involved in SANC, it is important to appreciate the basic techniques for molecular typing / classification of *S. aureus*. The most basic type of classification is based on a bacteria's capsular type. However, there are only two capsular types in *S. aureus*, which does not provide the necessary resolution for epidemiological studies. Many molecular typing techniques are available and most of them depend on PCR amplification of one or more genes. The major typing technique that does not depend on PCR uses pulsed field gel electrophoresis (PFGE) to distinguish different strains on basis of their restriction map as well as genome size (33). However, PFGE is a laborious, time-consuming and costly process, which cannot differentiate between strains varying by largely subtle changes. Furthermore, the results of PFGE are not portable, in the sense that two labs working with different strains cannot compare their results directly. Also, it has been shown that some of the modern molecular typing techniques are at least as sensitive to lineage differences as PFGE, while being portable (34). Therefore, much of the strain typing today is being performed at the sequence level. Sequence typing involves the amplification of a variable part of a target gene, the sequencing of the amplicon and comparison of the amplicon sequence across several strains. One might use any gene that exhibits variability at the DNA level for this purpose, though it is preferred that a gene that is necessary for *S. aureus* survival is used. For example, the gene *AgrB* is a hypervariable (2) across different strains of *S. aureus* and the sequence of *AgrB* from each strain under consideration will provide an unambiguous basis for comparison and analysis. Similarly other genes like *sdrC* (35), clumping factors A and B (36), all three of which are hypervariable, are also used for sequence typing.

Even though the use of single gene sequence typing allowed better portability, it was not as accurate in identifying differences, as was PFGE. This can arise, for example, in a scenario, where two strains sharing the same *AgrB* allele have differentially acquired a transposon bearing a group of toxin genes. Such a difference would not be picked up by any molecular typing method, but instead would be immediately visible in PFGE analysis. However, in order to increase the sensitivity of molecular typing methods, the practice of typing multiple genes was introduced. This has been named the multi-locus sequence typing (MLST) (reviewed in (37) and (38)) and since its advent, has been used for the molecular typing of several pathogens including *Streptococcus* spp (39) and *S. aureus* (38) (40). This technique exploits the fact that genes involved in central metabolic pathways are almost always vertically transmitted, as opposed to transposon related genes that are often horizontally transmitted. In *S. aureus*, MLST uses 7 genes that show variation in their sequences but not the length. From each gene, a 450-500 base pair region is amplified by using invariant primers, and amplicons are sequenced. The sequences are then used to distinguish between various strains (by clustering) or to confirm strain identity by comparing against stored MLST information in databases. Given reliability over single gene typing methods, and its speed and cost-efficiency over PFGE, MLST is now a technique of choice for studying lineage variations in *S. aureus*.

Besides those techniques mentioned above, a more standard typing technique was the *spa* typing (41) (42). This uses the polymorphisms in the staphylococcal protein A repeat region. Distinct variations in the tandem repeat region (region X) of this protein have been exploited successfully to distinguish between different strains of MRSA. In a different work, Shopsis and colleagues report a dual assay system that measures both short-term evolution (using

spa typing) and long-term evolution of strains, the latter using variations in the coagulase (*coa*) gene (43). In fact, in a study comparing *spa* and *clfB* typing, Harmsen and colleagues report that the former is a more sensitive and discriminatory technique (44). An ingenious mixture of *spa* typing and MLST, named MLVA (multi-locus variable tandem repeat analysis) has also been introduced (45) and combines the best of both methods, and provides discriminatory powers that are close to PFGE.

Bacterial factors involved in SANC

Apart from the host factors discussed above, several studies have focused on identifying the bacterial factors responsible for SANC. One gene that might have a role in host attachment in general, and nasal carriage might be *sdrE*. The gene *sdrE* encodes a host attachment domain (MSCRAMM) and is highly prevalent in almost all strains of *S. aureus*. Overall, the gene was present in about 90% of the tested individuals in various countries (46). Its presence in carrier strains was significantly higher than the linked genes, *sdrD*. Besides this gene, Schaffer and coworkers reported that the clumping factor B (ClfB) is necessary for nasal colonization in mice. The authors make a strong case for involvement of ClfB in SANC by using deletion mutants for this gene in nasal colonization studies in ICR mice (47). The immunized and control mice were colonized with both wild-type strains (Newman and 502-A) as well as specific mutants (for both *clfB* and *srt* sortase gene). However, abrogation of SANC in these mutants was inversely related with the bacterial dose, and colonization with a strong dose seemed to overcome the effects of ClfB deletion. Such need for ClfB protein in nasal colonization and probably carriage was shown in human subjects too, extending the results of Schaffer and coworkers (48). However, as we would discuss later in the text, ClfB and many other factors, while important, may not be sufficient to establish SANC. Another gene of importance in SANC / colonization, is the fibronectin binding protein (*fnbA*). This gene is present in almost all strains of *S. aureus* isolated from the human nasal epithelium (49), but is also present in invasive strains. It is important to note that the three genes discussed above - clumping factor B, *sdrE*, and fibronectin binding protein A, are MSCRAMM domain genes and augment the pathogen's interaction with the host (50). Many other enterotoxin genes (*se[a-m]*) were tested (51), though no factors were specific for carrier strains were identified. The authors however, show a high prevalence of *se[ceg]* amongst carrier strains.

The MSCRAMM molecules represent a broad category of proteins that play an important role host adhesion and establishment of carriage. Another multi-functional protein of *S. aureus* that plays a role in nasal carriage is the iron-dependent adhesin *isdA* (52), which is expressed specifically under Fe²⁺ depleted conditions. Besides acting as a siderophore (iron acquisition molecule) (53), this protein is also known to counter the innate immune responses of host epithelium (54). Furthermore, *isdA* is reported to protect the bacterium from the effects of host innate molecule apo-lactoferrin (55). Finally, it is to be noted that many of the genes listed above, namely *clfB*, *isdA*, and the *sdr* locus genes, are also necessary for invasion of host tissues and contribute to the formation of abscess lesions (56). Moreover, it is notable that *isdA* is induced by host immune molecules produced by neutrophils (57).

It is again worth noting that these proteins are important for establishing colonization, but none of them are exclusively present in nasal isolates. These genes have been tabulated for quick reference purposes (table 2). All of the above evidence points towards a highly complex interplay of host and bacterial factors in nasal carriage and colonization.

Genome level analysis of *S. aureus* genomes and insights into mechanisms of SANC

Despite many studies analyzing the bacterial factors involved in SANC, all that was known was certain tendencies and propensities of specific toxin genes being present in nasally carried strains. Meanwhile, a flurry of genome sequencing projects specific to the pathogen were well underway and by 2006, at least 7 different genomes were completely sequenced with 5 others well under way.

Extending the aforementioned MLST analysis to hitherto untested clinical nasal isolates, we observed that there was no lineage specificity for SANC (40)[Sivaraman 2008].

Interestingly, our model carrier strain and an invasive strain that was much less capable of adherence / growth on nasal epithelial cells (model non carrier strain) (28) belonged to the same sequence type. Primarily, these results show that factors necessary for SANC were not lineage dependent but likely a part of the “variable genome” (1).

Having established that SANC depends on variable genome, we studied the variations in the genomes of *S. aureus*. It was readily apparent that the loci and the nature of variations were not predictable in the bacterium. Further, even if it were to be predictable, it would have been impossible to identify targets involved in carriage, since the carriage status of sequenced strains was unknown. In order to decipher possible bacterial determinants of SANC, we sequenced two contrasting strains of *S. aureus*: one isolated from a persistent carrier (D30) and another a invasive strain isolated from a infected burn wound (930918-3). The former was the model carrier strain and the latter, a model non-carrier strain (These strains and their properties are described in reference (4)). The physiological differences between these two strains included the capability to produce a protective biofilm (carrier strain was biofilm positive), potentially suppressing innate immunity in the host (28) amongst others.

Genome sequencing and analysis revealed that D30 and 930918-3 were much closely related than was revealed by MLST analysis. For instance, USA300 (58) and NCTC8325 (59) belonged to the same clade on the MLST dendrogram, but differed within themselves by more than 400 genes. On the other hand, we found that D30 and 930918-3 differed only by 84 genes (D30 vs 930918-3) and 256 genes (930918-3 vs D30) respectively (40). Further, most of the differences we observed between the two strains were carried on mobile genetic elements like transposons (in the case of D30) or phages (in the case of 930918-3) (40). We compared the presence / absence of some of the well established factors (clumping factor B, *sdrE*, *tagO* (60), and the enterotoxins) in the completely sequenced genomes. Further genome level enquiry revealed that genes necessary for colonization (viz., *clfB*, *sdrE*, *se[ceg]* and *isdA*) were present in both D30 and 930918-3. This implied that nasal carriage and colonization are distinct phenomena that share majority of their symptoms. Further differential analysis was performed to delineate differences between the two genomes.

Analysis of genes unique to the carrier strain revealed the presence of two major mobile genetic elements possibly contributing to phenotypic differences and hence to carriage. One was designated the bovine pathogenicity island (SaPIBov) (61) and the other was a transposon carrying about a sixth of the total differential genes (13 of 84). There were three genes in the bovine pathogenicity island (BPI12, BPI13:14, and BPI17) present uniquely in the carrier strain. Albeit the SaPIBov is known to enhance infection in bovids (32), the specific role of these three ORFs is not known. Further, there was no sequence similarity for these genes (proteins) with other known toxins.

More interesting was the gene complement found on the transposon. Three notable genes on the transposon were *traG*, *rif* (replication initiation factor), and the *ftsK/spoIIIE* family protein. We speculate that RIF might be involved in supporting bacterial proliferation in the

face of adverse conditions (viz. host immune response). The role of *traG* and *ftsK* family protein appear clearer. The product of *traG* (TraG protein) is a membrane bound ATPase that provides energy for macromolecular transporter systems. In *Escherichia coli*, where it was originally characterized, the protein is a part of a plasmid translocation system (*Tra* system) (62). However, this protein family is found in other membrane bound transportation systems as well. In this particular case in *E. coli*, it was found that the transporter protein had both the *traG* domain and the *traJ* domain, the latter being a *ftsK/spoIII E* family domain. Also, in their work describing a search for a type VII secretion system, Burts and coworkers reveal the presence of a novel protein translocation system in *S. aureus* that contains a similar domain architecture (*traG* and *ftsK / spoIII E*) (63). We suspect that the transposon uniquely found in the model carrier strain D30 carries a novel type VII secretion system and it allows the bacterium to transport toxins and other effector molecules to counteract host responses in the nasal nares.

Other factors affecting *S. aureus* carriage

While our and other's studies cited above, show that disparate factors are responsible for SANC, there are also other factors worth mentioning. The most important amongst them would be strain level variations in the staphylococcal genome. There are several reports that elaborate strain level variations in certain pathogenic genes of the pathogen. Some genes of importance, like the clumping factor B are linked to nasal colonization (47) and others like *agrB* and *clfA* (64) are linked to stress response and biofilm formation, respectively. A combination of these genes would allow the pathogen to sustain and survive in the anterior nares. Of similar interest are the enterotoxin genes that show high levels of inter strain variability (51). In one of our recent studies, we had comprehensively analyzed the common minimal genome of the bacterium, using all the 14 complete genomes available. Our analysis revealed that pathogenicity related genes accumulated more variations at both the DNA and protein level than did the house keeping genes. We suspect that such high levels of variability may also contribute to subtle differences in SANC, apart from host factors (2).

Another much neglected aspect of SANC is the dynamics of carriage status within a host. Apart from factors that are inherent to the host or to the pathogen, there are other niche specific factors that might affect carriage. One such important factor that needs to be studied in greater detail is called bacterial interference. In short, bacterial interference is the phenomenon where a co-existing bacteria of different species affects the survival of an extant species. In a unique study (65), it was shown that there is an inverse correlation in children between the colonization of *S. aureus* and *Streptococcus pneumoniae*, which may indicate a competitive environment for these two microbes in the nares. It has to be noted that the *S. pneumoniae* strain in question was a vaccine strain and hence non invasive / pathogenic. It is known that a pathogenic strain of *S. pneumoniae* does not displace *S. aureus*, as does the vaccine strain (66). Thus, the effect of presence (or absence) of other bacterial species on SANC of *S. aureus* warrants a deeper inquiry. Studies have documented the generic microbiome composition of human skin (67) and its variations depending on biotic and abiotic factors (68). This might serve as a reference platform against which the microbiome(s) of carrier-hosts can be compared.

Future directions for studies on SANC

For many years, characterizing SANC has been restricted to the use of antigen types, single gene sequence types, and of late multigene sequence types. However, as we have argued above, such attempts are insufficient to characterize the biology of this pathogen to an extent that facilitates understanding. High throughput genome level approaches allow one to interrogate the microbial designs leading to carriage, as well as the host response to the threat. Foremost amongst the global questions is the effect of ethnicity on SANC. It will be

invaluable to know race specific susceptibilities to nasal carriage and subsequent complications. As detailed before, this would allow policy makers to draft efficient public awareness campaigns to control and subdue the spread of this pathogen in large pathogens. Besides, it is also important to identify and understand the factors that underlie longitudinal strain shifts. We have observed that in some cases, the strain profile changes in the host. While the health implications are yet undocumented, one can expect a persistent strain to displace a transient one, thereby leading to clonal propagation of persistent strains in community settings. The role of both the host and the bacterium played in such displacement scenarios have to be studied globally, at both the genome level as well as the transcriptome / proteome levels. Metagenome level studies of the epithelial microflora would also establish, with much greater detail, the role played by species that are either accessory or competitive to *S. aureus* in the niche. Also unknown are the changes in transcriptome and or proteome of the pathogen, that accompany the process of carriage initialization and persistence. Last but not least, a comprehensive survey of protein translocation systems in the pathogen would be of immense immediate interest. Besides, it will also be necessary to identify proteins that use these translocation systems and their pathogenic potential. These studies on the bacterial system would allow us to understand comprehensively, the bacterial factors responsible for SANC. Last but not least, the effect of stress, both psychological and environmental on nasal carriage must also be studied. It is known that psychological depression and chronic stress leads to suppression of immune responses (69). It is not known if chronic stress predisposes a host to nasal carriage and subsequent infection. Such a scenario presents an interesting case since a healthy non-permissive host might, under chronic stress, become a permissive one. Such studies would also allow us to define nasal carriage using environmental rather than biological parameters.

In summary, SANC of *S. aureus* is a complex phenomenon that affects our health care capabilities considerably, both in terms of social and economic fallouts. While the reasons behind the phenomenon are unclear, it is unambiguous that steps should be taken to control, and if possible prevent the spread of the pathogen in a community as well as hospital setting.

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Table 1List of host factors known to be associated with nasal carriage / colonization of *S. aureus*

S. No.	Gene	Remarks	Reference
1	HLA – DR3	Predisposes to nasal carriage	(19)
2	Fcγ receptor	Certain polymorphic variations predisposes individuals to SANC	(20)
3	Vitamin D receptor	Polymorphic variations predisposes type I diabetics to SANC	(20)
4	Interleukin 4	C542T genotype is associated with SANC	(21)
5	Protease C1 Inhibitor	C1INH V480M is associated with SANC	(21)
6	Glucocorticoid receptor	Certain polymorphs	(22)
7	Antimicrobial peptides	Required for antimicrobial activity of human airway fluid	(23) (24)
8	Beta-defensin 3	Only HBD effective against <i>S. aureus</i>	(25)
9	Human Cathelicidin	Synergistic action	(26)
10	TLR-2	Abrogation necessary for carriage	(28) (27)
11	Cytokeratin, involucrin, loricin	Non-immune factors	(29) (30)

Table 2

Bacterial factors known to be related to SANC / Colonization

S. No.	Gene	Remarks	Reference
1	Clumping factor B	Necessary for nasal colonization	(47) (48)
2	<i>sdrE</i>	MSCRAMM protein – host cell adhesion	(46)
3	<i>fmbA</i>	Fibronectin binding protein	(49)
4	<i>se[ceg]</i>	Enterotoxins	(51)
5	<i>isdA</i>	Siderophore	(55)
6	SaPIBov	Bovine pathogenicity Island proteins	(61) (32) (40)
7	Type 7 SS	TraG / FtsK complex, present in carrier strain	(63)