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Response to "*In vivo* attenuation and genetic evolution of a ST247-SCCmecI MRSA clone after 13 years of pathogenic broncho-pulmonary colonization in a patient with cystic fibrosis: implications of the innate immune response"

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To the Editor: With interest we read the article of Lopez-Collazo *et al.*[1] The authors reported about the *in vivo* attenuation and evolution of methicillin-resistant *Staphylococcus aureus* (MRSA) isolates, which were recovered from an individual cystic fibrosis (CF) patient 13 years apart. The authors described that these isolates displayed a "slow growth". Furthermore, in this article, Figure 3 shows the colony appearance of the first MRSA isolate revealing very small colonies. Both the slow growth and the small colonies mentioned by the authors are typical features of small colony variants (SCVs).

We recently showed that *S. aureus* isolates with a SCV phenotype and thymidine dependency are attenuated in virulence and showed dramatically changed metabolism.[2] Although SCVs are associated with persistent and recurrent *S. aureus* infections such as osteomyelitis and device-related infections,[3] especially thymidine-dependent SCVs can often be retrieved from the airways of CF patients and are associated with worse lung disease.[4] Many studies pointed out that SCVs originate from normal *S. aureus* phenotypes usually in cases of long-lasting infections and that SCVs may occur and be selected because of their optimized persistence properties.[3]

Therefore, it would be of interest to know whether this patient was colonized/infected by normal *S. aureus* phenotypes earlier. As the patient was 9 years when the first MRSA-SCV was collected, it is very likely that normal *S. aureus* isolates were already present earlier. A comparison of such a normal strain to the latest isolate recovered in 2009 would be even more pronounced, not only in terms of the bacterial adaptation but also in terms of the immune response of the patient. It has been shown that SCVs escape the immune response by downregulation of important virulence genes,[5] which is in line with the authors' observations. Furthermore, a detailed analysis of the whole genome sequencing data of the two SCV isolates would be of major interest. One could assume that the strains carry defined mutations, which might cause the SCV phenotype (e.g., mutations in heme, menadione or thymidine pathways).[3] However, such mutations can only be retrieved if these SCV isolates are compared with a normal isogenic *S. aureus* isolate recovered from the patient or compared with another normal closely related strain.

Moreover, it would be of interest to determine the auxotrophism of these SCVs, as a mean to investigate which substrate will support the phenotypic reversion of the SCVs to determine the underlying mechanism.

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