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Characterising resistance and survival roles of a *S. aureus* two-component system

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Background: Staphylococcus aureus has evolved multiple mechanisms to survive in different environments, including the ability to resist antimicrobials produced by other bacteria through various mechanisms, such as modification of the cell wall and efflux pumps. We have identified antimicrobial resistance roles for the two-component system (TCS) that is a homologue of both DesKR and YvfTU of Bacillus subtilis. The S. aureus TCS has proposed roles in resistance and thermoregulationExperimental evolution of S. aureus was used to select for resistance to S. epidermidis expressing epifadin, which is a bacteriostatic-like antimicrobial peptide with a short half-life (Torres Salazar et al., 2023). Selection was performed in mixed culture with either epifadin-producing S. epidermidis IVK83 or an epifadin gene mutant strain IVK83 ΔefiTP, as competitor species. S. aureus USA300 evolved resistance to epifadin after 5 days and genome sequencing revealed mutations in the desK-like gene, encoding the histidine kinase of the DesKR-TCS, dependent on the presence of the epifadin operon in S. epidermidis. Mutation of desK-like gene in the S. aureus evolved mutant restored inhibition by S. epidermidis. DesKR-TCS is proposed to have roles in thermoregulation, therefore the stability of the desK-A162V mutation was examined during serial passage at both 25°C and 37°C over 30 days. Loss of epifadin resistance was identified at after 16 days 25°C but not at 37°C. Genome resequencing was used to investigate the mutations associated with phenotype change. These data support the roles of DesKR TCS in both epifadin efflux and thermoregulation together with demonstrating resistance to other antimicrobials.