Diversity of *Staphylococcus* species isolated from surface waters

Vanessa Silva[1-4]*, Manuela Caniça[5,6], Vera Manageiro[5,6] Ana Sampaio[7], José L. Capelo[8,9], Gilberto Igrejas[2-4], Patrícia Poeta[1,4]

[1]Microbiology and Antibiotic Resistance Team (MicroART), Department of Veterinary Sciences, University of Trás-os-Montes and Alto Douro (UTAD), Vila Real, Portugal;

[2]Department of Genetics and Biotechnology, University of Trás-os-Montes and Alto Douro, Vila Real, Portugal;

[3]Functional Genomics and Proteomics Unit, University of Trás-os-Montes and Alto Douro (UTAD), Vila Real, Portugal;

[4]Associated Laboratory for Green Chemistry (LAQV-REQUIMTE), University NOVA of Lisboa, Lisboa, Caparica, Portugal;

[5]National Reference Laboratory of Antibiotic Resistances and Healthcare Associated Infections (NRL-AMR/HAI), Department of Infectious Diseases, National Institute of Health Dr Ricardo Jorge, Av. Padre Cruz, 1649-016, Lisbon, Portugal;

[6]Centre for the Studies of Animal Science, Institute of Agrarian and Agri-Food Sciences and Technologies, Oporto University, Oporto, Portugal;

[7]Centre for the Research and Technology of Agro-Environmental and Biological Sciences (CITAB), University of Trás-os-Montes and Alto Douro (UTAD), Vila Real, Portugal;

[8]BIOSCOPE Group, LAQV@REQUIMTE, Chemistry Department, Faculty of Science and Technology, NOVA University of Lisbon, Almada, Portugal;

[9]Proteomass Scientific Society, Costa de Caparica, Portugal

*vanessasilva@utad.pt

Introduction

Antimicrobial resistant bacteria in aquatic systems represents one the greatest challenges and emerging threats since water is one of the most relevant vehicles of bacterial dissemination. Monitoring the presence of antimicrobial resistance human pathogens, in particular staphylococci, may lead to a better understanding of the epidemiology of infections caused by these bacteria. Thus, we aimed to investigate the prevalence of *Staphylococcus* spp. in surface waters in Portugal.

Methods

Seventy-seven water samples (30 rivers, 18 streams, 11 irrigation systems, 10 dams, 5 fountains and 3 lakes) were collected from the hydrographic basins of Douro river in Trás-os-Montes and Alto Douro region, Portugal. Samples were filtered through a cellulose nitrate pore membrane filter. The filters were incubated at 37ºC for 24 h in BHI broth supplemented with 6.5% of NaCl. After that, the inoculum was seeded on Mannitol Salt agar plates and incubated at 37ºC for 24 h. Staphylococci species were identified by MALDI-TOF MS. Antimicrobial susceptibility testing was performed by the Kirby-Bauer disk diffusion method.

Results

Seventy-five staphylococci were isolated from water samples of which 30 were coagulase-positive (CoPS) and 45 were coagulase-negative staphylococci (CoNS). Among the CoPS, 29 were identified as *S. aureus* and one as *S. pseudintermedius*. The CoNS isolated included *S. sciuri*, *S. lentus*, *S. xylosus* and *S. epidermidis*, among others (Figure 1). *S. aureus* were susceptible to almost all antibiotics tested, and CoNS were mostly resistant to penicillin, clindamycin and fusidic acid.

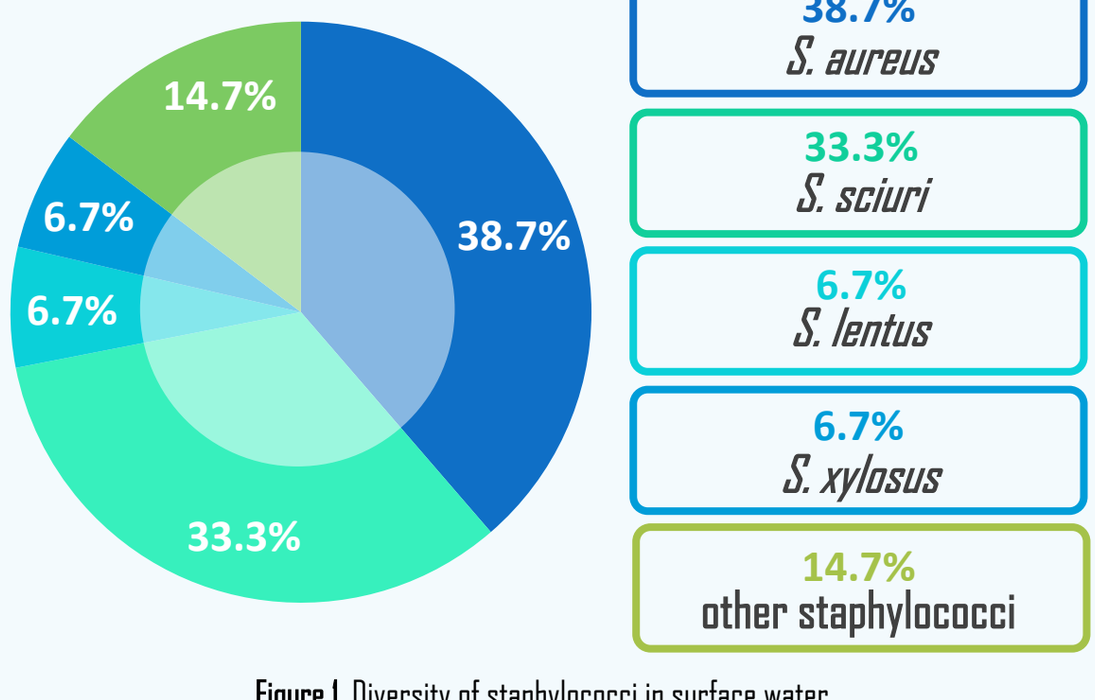

Figure 1. Diversity of staphylococci in surface water.

Conclusions

This study demonstrated that staphylococci, including *S. aureus*, is present in surface waters. A high diversity of staphylococci was detected in surface water samples, in particular for CoNS.

Acknowledgements:

This work was funded by the R&D Project CAREBIO2 - Comparative assessment of antimicrobial resistance in environmental biofilms through proteomics - towards innovative theranostic biomarkers, with reference NORTE-01-0145-FEDER-030101 and PTDC/SAU-INF/30101/2017, financed by the European Regional Development Fund (ERDF) through the Northern Regional Operational Program (NORTE 2020) and the Foundation for Science and Technology (FCT). This work was also supported by the Associate Laboratory for Green Chemistry-LAQV which is financed by national funds from FCT/MCTES (UID/QUI/50006/2020). Vanessa Silva is grateful to FCT (Fundação para a Ciência e a Tecnologia) for financial support through PhD grant SFRH/BD/137947/2018.