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BACTERIAL DIVERSITY AND PROBIOTIC ACTIVITY IN SOLE AQUACULTURE

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Solea senegalensis aquaculture may be affected by bacterial infections and, therefore, negative impacts on the production yields. Because the use of antibiotics should be minimized, supplementation of fish feeds with probiotic cultures is a good practice, increasingly well-accepted. The aim of this study was to characterize the culturable gut microbiota of juvenile sole, subjected to dietary supplementation with commercial probiotics.

Two commercial probiotics were used: a multi-strain probiotic (PA) including *Bacillus* sp., *Pediococcus* sp., *Enterococcus* sp. and *Lactobacillus* sp. and a mono-strain culture (PB) corresponding to *Pediococcus acidilactici*. Both were tested at the recommended dose (PA1 and PB1) and at two-fold lower dosage (PA2 and PB2). Total heterotrophs, and presumable faecal coliforms, pseudomonads and aeromonads, and lactic acid bacteria were enumerated on different culture media.

Culturable proteobacteria, especially enterobacteria and aeromonads, were significantly more abundant in sole gut than lactic acid bacteria (3.71–7.84 and 1.2 – 5.8 log CFU g⁻¹, respectively). The fish that fed with the mono-strain probiotic (PB), mainly at PB2 dose, presented significant increase on the counts of culturable bacteria of all groups tested. In fish fed with the probiotic PA (dose PA1) was observed a decrease in bacterial counts mainly of lactic acid bacteria. In the control diet was observed a reduction in the bacterial counts for all the culture media along the three sampling campaigns diet.

A group of 286 isolates recovered over the three sampling dates was tested for their antimicrobial activity, against *Aeromonas salmonicida* subsp. *salmonicida* LMG3780, *Photobacterium damsela* subsp. *piscicida* LG41/01, *Staphylococcus aureus* DSM 1104 and *Vibrio anguillarum* VCO1.06.05, aiming their possible use as probiotic agents. Antimicrobial activity was detected in 13 strains identified as *Vagococcus fluvialis*, *Enterococcus raffinosus*, *Vibrio furnissi*, *Bacillus methylotrophicus*, *Acinetobacter venetianus*, *Shewanella upenei*, *Shewanella hafniensis*, *Shewanella haliotis* and *Vibrio alfacensis*. All strains presented activity against *A. salmonicida* subsp. *salmonicida* and only two, identified as *V. furnissi* and *A. venetianus*, were active against *P. damsela* subsp. *piscicida*. The fact that none of these species is listed as fish pathogen and the absence of presumed acquired antibiotic resistance phenotypes make these isolates good candidates as probiotics.